

Prostate autoimmunity: from experimental models to clinical counterparts

Expert Rev. Clin. Immunol. 5(5), 577–586 (2009)

Giuseppe Penna,
Benedetta Fibbi,
Mario Maggi and
Luciano Adorini[†]

[†]Author for correspondence
Intercept Pharmaceuticals,
Via P. Togliatti 22 bis,
06073 Corciano (Perugia), Italy
Tel.: +39 075 792 1934
Fax: +39 075 792 1696
ladorini@interceptpharma.com

Different murine models of autoimmune prostatitis have been developed and characterized, proving the autoimmune origin of this pathology. Autoimmune prostatitis models have also provided a wealth of information on the mechanisms involved in disease development, shedding light on inciting autoantigens, regulatory and pathogenic T cells, and mediators of prostatic autoimmunity. Unfortunately, the clinical counterparts of experimental autoimmune prostatitis are still poorly defined. In this review, we will discuss evidence for the autoimmune origin of chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS) and the chronic inflammatory nature of benign prostatic hyperplasia (BPH). The autoimmune pathogenesis of CP/CPPS and the chronic inflammation characteristic of BPH will be reviewed within the context of the recent demonstration that human prostate stromal cells from BPH tissue can act as antigen-presenting cells and are not only able to activate CD4⁺ T lymphocytes, but can also produce IL-12 and IL-23, which are key cytokines for the induction of pathogenic Th1 and Th17 cells.

KEYWORDS: antigen-presenting cells • benign prostatic hyperplasia • chronic prostatic inflammation • chronic prostatitis/chronic pelvic pain syndrome

Prostatitis and benign prostatic hyperplasia (BPH) are common pathologies of the prostate gland. Although it is generally accepted that prostatitis and BPH are distinct clinical entities, these two common male problems frequently coexist [1] and could be strongly related, sharing common pathogenetic mechanisms based on the capacity of prostate cells to act as inducers and targets of chronic inflammation. The immunology of the prostate has recently emerged as a new field of research in urology, highlighting the complex network of cytokines and chemokines involved in the prostatic inflammatory response [2]. Studies are now permitting us to delineate a causal relationship between prostate inflammation and highly prevalent prostatic diseases, including BPH, chronic prostatitis and even prostate cancer.

Prostatitis, defined as a clinical syndrome mainly characterized by pelvic pain, is the most common urologic disorder in males younger than 50 years of age, with an overall prevalence of approximately 8% [3]. The NIH consensus definition and classification identifies four categories of prostatitis [4]. Category I includes acute bacterial prostatitis and category II includes chronic bacterial prostatitis. These are rare

conditions characterized by a defined infectious etiology. Category III, also known as chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS), is defined by pelvic pain in the absence of demonstrable bacterial infection, and is further subdivided into category IIIA, or inflammatory, and category IIIB, or noninflammatory, based on the presence of leukocytes in expressed prostatic secretion or seminal plasma. Category IV, or asymptomatic inflammatory prostatitis, is defined by the presence of leukocytes in seminal secretions or by inflammatory infiltrates detected in histological specimens, in the absence of typical chronic pelvic pain. CP/CPPS (NIH category III prostatitis), which includes the overwhelming majority of prostatitis patients, is common in medical practice and its prevalence in the general population ranges from 5 to 14.2% [5]. Therefore, according to these categories, approximately 60–80% of all prostatitis syndromes could be classified as CP/CPPS. Interestingly, BPH fits well the criteria defined for category IV, although it is not yet officially defined as prostatitis. It is tempting to speculate that NIH categories III and IV actually represent forms of prostate autoimmunity.

The prostate, weighing only a few grams at birth, grows during puberty, reaching approximately 20 g in the young adult. In contrast to the pubertal growth phase, which involves the entire gland, during the fifth decade of life in approximately 75% of men there is a second growth phase, selectively involving the periurethral zone. Enlargement of this zone gives rise to BPH, the most common age-related disease of males [6]. BPH can cause lower urinary tract symptoms owing to compression of the prostatic urethra, which, in the earliest stages, is compensated by an increased activity of the bladder detrusor muscular system. However, later on, complete emptying of the bladder is prevented, owing to the slackening of the neck musculature, potentially leading to emergency surgery for acute urinary retention [6].

The prostate, once considered an immunoprivileged site [7], is actually a fully immunocompetent organ, owing to the presence of an intraglandular immune system participating in the maintenance of reproductive tract sterility, and preventing autoimmune responses towards intraprostatic and sperm antigens [8]. In the normal human prostate, leukocytes are localized within the interstitium around the epithelial ducts and between epithelial cells, comprising T lymphocytes (with a prevalence of CD8⁺ vs CD4⁺ cells), macrophages, B lymphocytes and mast cells [9,10].

The primary event in the activation of the inflammatory network in the prostate is still unclear, but infections could play an important role. Several studies have demonstrated the presence of a heterogeneous set of bacterial and viral strains in BPH samples [11]. Prostate cells express all Toll-like receptors (TLRs) [12–15], enabling them to recognize pathogen-associated molecular patterns, which are structural components highly conserved among pathogens that trigger the activation of innate and adaptive immune responses [16]. TLR agonists have been found to induce the production of proinflammatory cytokines and chemokines by BPH stromal cells, able to recruit infiltrating cells and promote prostate cell growth, supporting a link between infection, the inflammatory response and prostate pathology [15].

Prostatic immune responses can be also triggered by self-antigens. A relationship between the autoimmune response and the pathogenesis of BPH was established through the detection of increased levels of anti-prostate-specific antigen (PSA) antibodies in sera from BPH patients, with the authors concluding that BPH could be considered an organ-limited autoimmune condition characterized by the absence of polyclonal B-cell activation [17]. Further work from the same authors has shown that PSA can induce *in vitro* proliferation of peripheral blood mononuclear cells (PBMCs) from BPH patients and enhance their secretion of TNF- α , suggesting a role for autoimmune dysregulation in BPH pathogenesis [18]. The hypothesis of an autoimmune pathogenesis in BPH is, however, not formally proven, although it is supported by the prevalence of Th1 and Th17 subsets, which are typical of autoimmune diseases [19], in BPH tissues [11]. Conversely, the autoimmune origin of CP/CPPS appears to be quite firmly established [20,21]. If this is the case, as CP/CPPS is eight-times more frequent than bacterial prostatitis [3,5], autoimmune responses could represent a major cause of prostatitis.

The predominant role of autoimmunity in CP/CPPS development has been confirmed by several murine models of spontaneous age-related and antigen-induced experimental autoimmune prostatitis (EAP) [20,22]. These findings in experimental models of autoimmune prostatitis, together with the induction of pathogenic T-cell responses by BPH stromal cells [15], suggest common mechanisms promoting and sustaining BPH and CP/CPPS.

Experimental models of prostate autoimmunity

Several experimental models of autoimmune prostatitis have been developed and extensively characterized [20,22]. Immunization of murine strains with a variety of prostate antigens, including male accessory gland extracts, total prostate homogenate, PSA, prostatic acid phosphatase (PAP) and prostatic steroid-binding protein (PSBP), can induce autoantigen-specific T-cell and antibody responses associated with histological evidence of prostate inflammation (TABLE 1). In some cases, prostatitis has been transferred to naive [23] or immunodeficient [24] syngeneic recipients by the adoptive transfer of CD4⁺ T cells from diseased animals, a hallmark of autoimmune disease.

In addition to autoantigen-induced autoimmune prostatitis, the development of spontaneous prostatitis is well documented in several rat strains [20,22] and in the nonobese diabetic (NOD) mouse [24]. The NOD mouse is genetically prone to develop different organ-specific autoimmune diseases, including Type 1 diabetes, thyroiditis, sialitis, oophoritis, adrenalitis and orchitis [25]. Interestingly, this mouse strain also spontaneously develops autoimmune prostatitis, defined by intraprostatic lymphomononuclear cell infiltrates. Spontaneous autoimmune prostatitis is preceded by a marked autoreactive T-cell response to PSBP, indicating a lack of tolerance to this prostatic self-antigen in the NOD mouse [24]. The intraprostatic inflammatory infiltrate spontaneously developing with age in the NOD mouse is characterized by Th1-type CD4⁺ T cells, which are able to adoptively transfer autoimmune prostatitis in immunodeficient NOD.SCID recipients, confirming the autoimmune nature of the spontaneous disease [24]. The NOD mouse is highly susceptible to the induction of EAP by injection of mouse prostate homogenate in CFA, a protocol inducing a florid intraprostatic leukocyte infiltrate in 100% of male NOD mice [24,26]. EAP can also be induced in NOD mice by injection of PSBP [24,27] (also known as prostatein), a major autoantigen in murine EAP [23,28]. The male NOD mouse responds to PSBP by developing specific cellular and humoral autoimmune responses associated with inflammatory infiltrates in the prostate [24,27]. NOD mice lacking expression of β 2-microglobulin or the MHC class II β chain have demonstrated an essential role for CD4⁺ T cells in the development of EAP [27], and functional Th1 cells specific for immunodominant PSBP peptides have been identified within the prostate of peptide-primed NOD mice [29]. Collectively, these data demonstrate the autoimmune nature of spontaneous and induced prostatitis in the NOD mouse, confirming and extending previous evidence obtained in rats and mice (TABLE 1).

Experimental autoimmune prostatitis can also be induced in different mouse strains by thymectomy at day 3 of age [30]. EAP in this model initiates at puberty, reaching an incidence of 68%

Table 1. Autoimmune responses to prostate antigens in experimental models and in leukocytes from normal donors and/or chronic prostatitis/chronic pelvic pain syndrome patients.

Prostate antigen	Species	Readout responses	Ref.
MAG extract	Mouse	T-cell proliferation, IFN- γ production	[26]
MAG extract	Rat	DTH levels, proliferation, CTL response, antibody production, mast cell degranulation, MIF production	[28,73–81]
Prostate homogenate	Mouse	IFN- γ , IL-17 production	[24, 29,82]
Prostate homogenate	Rat	Anti-PSBP response	[23]
PSA	Mouse	T-cell proliferation	[42]
PSA	Human	T-cell proliferation, IFN- γ production, CTL response	[36,39,40,42,83–87]
PSM	Human	T-cell proliferation	[36]
Kallikrein 4	Human	T-cell proliferation	[88]
PAP	Mouse	T-cell proliferation	[43]
PAP	Rat	CTL response	[89]
PAP	Human	T-cell proliferation, IFN- γ production	[40,43,90]
PSBP	Mouse	IFN- γ production	[24,27]
PSBP	Rat	DTH, proliferation, antibody response	[23,28]

All animal studies included histological assessment of intraprostatic leukocyte infiltration.

CTL: Cytotoxic T lymphocyte; DTH: Dihydrotestosterone; MAG: Male accessory gland; MIF: Migration inhibitory factor; PAP: Prostatic acid phosphatase; PSA: Prostate-specific antigen; PSBP: Prostatic steroid-binding protein; PSM: Prostate-specific membrane antigen.

in 50–150-day-old mice. Thus, mice thymectomized at a critical neonatal age develop EAP by autosensitization to antigen(s) normally expressed in the differentiated prostate [30]. Prostate autoimmunity can be completely prevented by a single intraperitoneal injection of spleen cells from syngeneic adult mice on day 4, strongly suggesting a role for active tissue-specific regulatory T cells (Tregs) in prostate self-tolerance [31]. Subsequent work has demonstrated that tissue-specific CD4⁺ Tregs activated extrathymically by prostate antigens maintain peripheral tolerance to prostatic autoantigens [32]. Based on these studies, it is conceivable that immune tolerance to self-antigen(s) in the prostate is maintained by tissue-specific Tregs, which migrate from the thymus to the periphery later than day 3 after birth, and can inhibit the activation of autoreactive T cells.

Finally, alterations in steroid hormone levels have been shown to induce autoimmune prostatitis in the rat, based on the evidence for prostatitis induced by the adoptive transfer of splenic CD3⁺ T cells from 17 β -estradiol-treated rats into naive recipients [33]. Several proinflammatory genes were upregulated in the prostate after only 4 days of treatment with 17 β -estradiol, including *IL-1 β* , *IL-6*, *MIP-2* and inducible nitric oxide synthase (*iNOS*). After 4 weeks of treatment with 17 β -estradiol and dihydrotestosterone propionate, significant increases in the transcript levels of *IL-4*, *IL-5*, *IL-6*, *MIP-2*, *eotaxin* and *iNOS* were detected, showing the inflammatory nature of this hormonally induced prostatitis [34]. In addition, elevated activities of matrix metalloproteinases (MMPs), including MMP-2, MMP-7 and MMP-9, correlate with leukocyte infiltration in the lateral lobe inflammatory response induced in the rat by chronic treatment with 17 β -estradiol [35].

Human prostate stromal cells are antigen-presenting cells potentially able to induce autoimmune prostatitis

Prostate stromal cells obtained from BPH tissue actively contribute to the organ-specific inflammatory process by acting as antigen-presenting cells (APCs) and as targets of TLR agonists, leading to the production of proinflammatory cytokines and chemokines able to mediate prostate chronic inflammation and hyperplasia [15]. As mentioned previously, prostate stromal cells express all the known TLRs, and their triggering by bacterial or viral agonists leads to the secretion of proinflammatory cytokines such as IL-6, and chemokines such as CXCL10 and IL-8. In addition to the growth-promoting properties of IL-8 and IL-6 on prostate cells, the capacity of IL-8 and CXCL10 to recruit inflammatory cells could play a role in inducing and maintaining chronic inflammatory conditions of the prostate, such as those observed in BPH and CP/CPPS patients [15].

The concept of prostate autoimmunity, largely based on experimental models (TABLE 1), has recently received critical support following the observation that human prostate cells can act as APCs. Stromal BPH cells exhibit key features of APCs: the expression of MHC class II molecules and costimulatory molecules, and the production of high levels of IL-12/IL-23p40 and IL-12p75. Stromal BPH cells present alloantigen to CD4⁺ cells, inducing the secretion of IFN- γ and IL-17. These cytokines strongly enhance the production by prostate cells of IL-8, an autocrine/paracrine growth factor for prostate cells and a potent chemokine recruiting to the prostate inflammatory cells (FIGURE 1). Although chronic inflammation does not necessarily imply an autoimmune pathogenesis, these findings suggest a plausible association between prostatic APC-induced T-cell responses, chronic prostate inflammation and sustained prostatic cell growth in BPH.

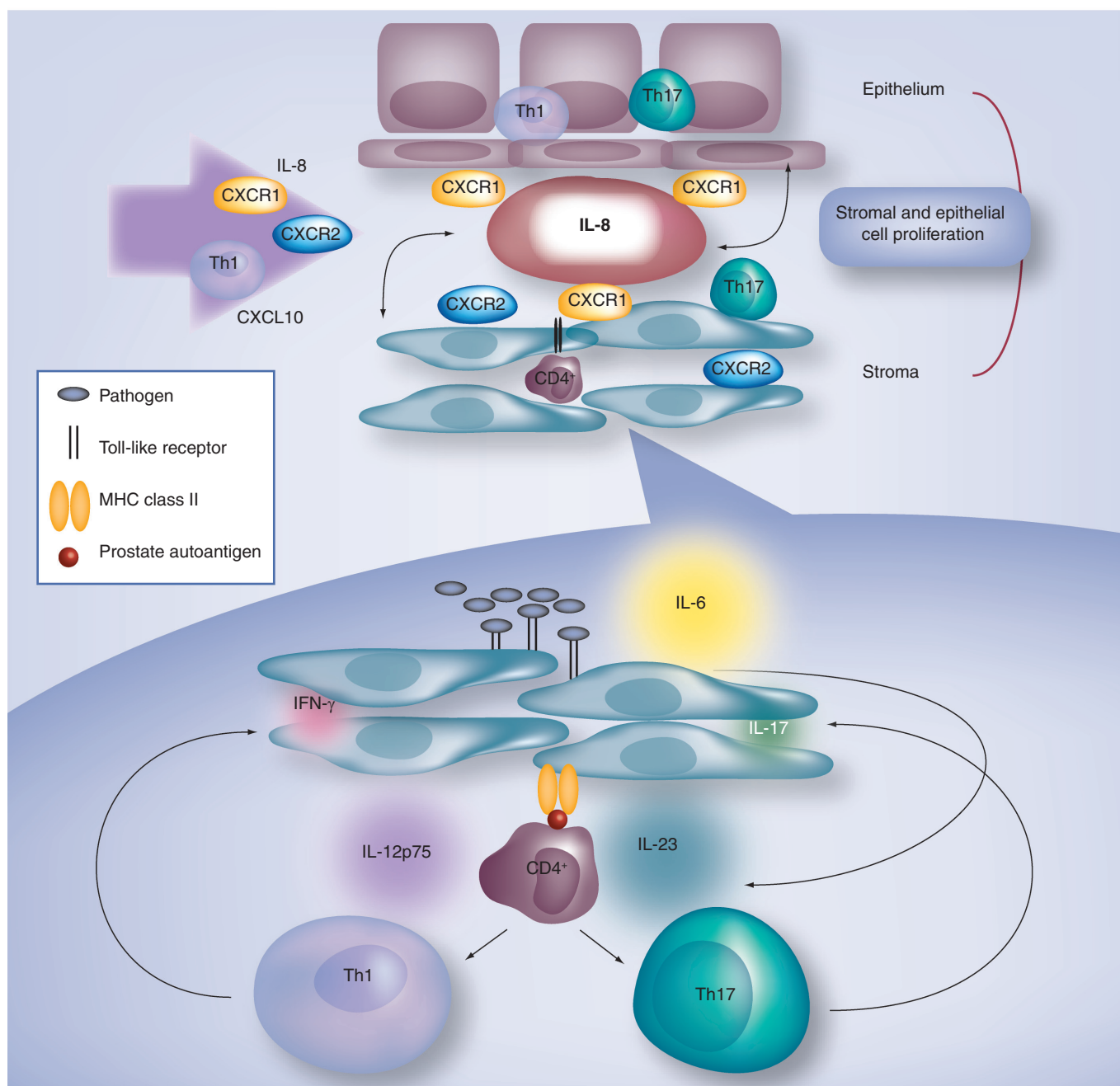


Figure 1. IL-8 has a central role in prostatic chronic inflammation, recruiting inflammatory cells and mediating prostate cell proliferation. Epithelial and stromal prostate cells actively secrete the proinflammatory chemokine IL-8 in response to different stimuli, including IFN- γ and IL-17 produced by prostate-infiltrating Th1 and Th17 cells, respectively. IL-8 recruits to the prostate lymphomononuclear cells expressing the cognate receptors CXCR1 and CXCR2, and directly (via an autocrine/paracrine action) and indirectly (via FGF-2 induction) promotes prostate cell proliferation. Therefore, IL-8 appears to represent a key link between chronic inflammation and overgrowth of benign prostatic hyperplasia (BPH) tissues. Inset: BPH stromal cells are inducers and targets of prostatic chronic inflammation. BPH stromal cells express all the Toll-like receptors (TLRs), which are pattern recognition receptors that recognize structural components highly conserved among pathogens. TLR activation by infectious stimuli (bacteria, viruses or fungi) induces production by stromal BPH cells of proinflammatory cytokines (IL-6) and chemokines (CXCL10 and IL-8) able to recruit infiltrating cells and promote prostate cell growth. BPH stromal cells express MHC class II and costimulatory molecules, functioning as nonprofessional antigen-presenting cells (APCs), leading to the activation of CD4 $^{+}$ T lymphocytes and promoting their differentiation into Th1 and Th17 effector cells by secreting IL-12 and IL-23, respectively. In turn, activated Th1 and Th17 lymphocytes produce proinflammatory cytokines, such as IFN- γ and IL-17, able to mediate pathogenic effects while sustaining and amplifying immune-mediated inflammatory responses in the prostate.

The nature of the inciting autoantigen(s) in prostate autoimmunity is still unclear, but several candidates have been proposed (TABLE 1). Autoreactive CD4⁺ and CD8⁺ T cells specific for prostate antigens exist in normal individuals, as demonstrated by the capacity of peripheral T cells from healthy controls to respond to PSA and prostate-specific membrane antigen [36]. This autoreactive T-cell repertoire could potentially become activated by a number of prostate antigens to mount an autoimmune response (TABLE 1). Indeed, PBMCs and CD4⁺ T cells from CP/CPPS patients proliferate in response to seminal plasma [37,38] and specific prostate antigens such as PSA and PAP [39,40], indicating expression of the autoreactive T-cell repertoire in disease pathogenesis. High-titer IgG autoantibodies to prostate-associated proteins are found in patients with CP/CPPS [41], further indicating a T-cell-dependent autoimmune process.

Among self-antigen candidates, PSA has been demonstrated to activate CD4⁺ T cells from patients with granulomatous prostatitis via two different antigenic peptides presented by HLA-DRB*1501 [42]. Two immunodominant PAP peptides, naturally processed from native PAP in HLA-DRB1*1501 tg mice, were also capable of stimulating CD4⁺ T lymphocytes from HLA-DRB1*1501-positive granulomatous prostatitis patients and normal donors [43]. Interestingly, PSA, a major secretory protein of the prostate, can also activate innate immunity, inducing a pro-inflammatory response that results in IFN- γ secretion by natural killer cells [44]. Thus, PSA could contribute to prostate inflammation, both as an autoantigen and as an activator of innate immune responses.

The hypothesis that prostate cells can induce and sustain intraglandular autoimmune responses is strongly supported by our demonstration that human stromal prostate cells are APCs able to activate alloreactive CD4⁺ T cells and secrete IL-12p75 and IL-23, which could trigger and amplify the differentiation of pathogenic Th1 and Th17 cells specific for prostatic autoantigens (FIGURE 1) [15]. Interestingly, significantly elevated levels of IL-12p75 have been observed in the seminal plasma of both CP/CPPS and BPH patients compared with controls [45], providing a clinical correlate for enhanced secretion of the key Th1-inducing cytokine by prostate stromal cells. Increased IL-12p75 levels in seminal plasma are also consistent with the INF- γ -secreting lymphocytes specific for the prostate autoantigens PSA and PAP detected in 34% of CP/CPPS patients [40].

CP/CPPS as an autoimmune disease

Chronic prostatitis/chronic pelvic pain syndrome is still an enigmatic syndrome, with immunological, neurological and endocrine dysfunctions proposed to be involved in disease development [21]. However, evidence indicative of a predominant autoimmune component in the pathogenesis of CP/CPPS is emerging [20,46]. The autoimmune nature of CP/CPPS is supported by experimental models of prostatitis (TABLE 1), which clearly demonstrate that the immunization of rats or mice with prostate gland extracts or defined prostate antigens can induce T-cell and antibody responses to prostate autoantigens associated with a robust intraprostatic inflammatory response.

The isolation of autoreactive CD4⁺ and CD8⁺ T lymphocytes specific for prostatic antigens from normal individuals [36], the capacity of seminal plasma [37,38] and specific prostate antigens [39,40] to induce the proliferation of PBMC and CD4⁺ T cells, and the autoreactive CD4⁺ cell-mediated B-cell activation, with the production of high-titer autoantibodies to prostatic proteins [41], all support the pathogenic role of autoimmune responses to prostate antigens in CP/CPPS.

Chronic prostatitis/chronic pelvic pain syndrome is, by definition, a nonbacterial prostatitis. However, it is conceivable that prostate infections may contribute to disease pathogenesis by triggering an organ-specific autoimmune response. Microbial products could activate signal transduction via TLRs expressed by prostate cells, leading to the production of proinflammatory cytokines and chemokines that contribute to create the conditions for an autoimmune attack. This pathogenic mechanism, in concert with genetic susceptibility and the induction of adaptive immune responses, may precipitate autoimmune diseases, as is shown by the induction of autoimmune myocarditis by a combination of TLR stimulation and CD40-mediated triggering of self-peptide-loaded dendritic cells (DCs) [47]. Activation of TLRs in target tissues could also be involved in the pathogenesis of systemic autoimmune diseases, as suggested for TLR-9 in systemic lupus erythematosus [48]. TLRs expressed by human prostate cells are functional, and their triggering by viral or bacterial products, such as poly I:C and lipopolysaccharide, induces the production of proinflammatory chemokines such as IL-8 and CXCL10, and cytokines such as IL-6 [15]. In addition to the growth-promoting properties of IL-8 and IL-6 on prostate cells, the capacity of IL-8 and CXCL10 to recruit inflammatory cells could play a role in inducing and maintaining chronic inflammatory conditions of the prostate that could promote autoimmune responses (FIGURE 1).

Besides infections, intraprostatic reflux, frequent or prolonged contraction of the smooth muscle in the urethra/bladder neck and prostatic stroma, increased intraprostatic pressure and hormonal imbalances have been recognized as potential risk factors for CP/CPPS [49]. No correlation has been found between leukocyte and bacterial counts and the severity of CP/CPPS symptoms [50] or the aggressiveness and extent of inflammation with leukocyte counts in prostatic fluid [51], implying that factors other than inflammation and infection contribute to the symptoms associated with CP/CPPS [52]. Among them, modulation of estrogen and the androgen/estrogen ratio, known to be involved in the control of inflammatory responses [53], can induce experimental prostatitis of a reported autoimmune nature [33]. All these risk factors could potentially favor an autoimmune prostatic response, and intraprostatic reflux of urine or semen may induce epithelial injury that could precipitate an immunological response.

BPH as an autoimmune disease?

Benign prostatic hyperplasia is the most common age-related disease of males, occurring clinically in approximately 50% of men at 70 years of age [54]. BPH is defined by hyperproliferation of the stromal and epithelial cells of the prostate, caused by complex cellular alterations, including changes in proliferation,

differentiation, apoptosis and senescence [55]. A variety of growth factors have been implicated in the pathogenesis of BPH, and two members of the FGF gene family, FGF-2 and FGF-7, are considered to play key roles in prostate cell growth [56]. However, the complex regulatory mechanisms of growth control in BPH are still incompletely understood.

Although primarily characterized by prostatic cell proliferation, an inflammatory component has been extensively documented in BPH [11,57,58]. Strikingly, histological evidence of prostate inflammation was identified in all patients tested, irrespective of bacterial infection [59]. Interestingly, analysis of prostate biopsies in a subgroup of over 1000 randomly selected patients from the Medical Therapy of Prostatic Symptoms study indicates that the presence of inflammatory infiltrates in the prostate of BPH patients is associated with an increased rate of disease progression and a higher risk of acute urinary retention [60]. In addition, analysis of baseline data from the Reduction by Dutasteride of Prostate Cancer Events (REDUCE) trial indicates an association between inflammation and BPH symptoms [52]. As for CP/CPPS, it is unclear whether an infectious origin could represent the primary event inducing inflammation in BPH pathogenesis. Microbial organisms demonstrated thus far in BPH appear to be rather heterogeneous, and no study has reported a single strain of microorganism present in all BPH samples, while viruses have been detected less commonly in association with BPH [11].

Inflammatory infiltrates in BPH have been found to consist primarily of T cells; mostly CD4⁺CD45RO⁺ cells, B cells and macrophages [11]. The upregulation of several proinflammatory cytokines has been described in BPH, in particular IL-2, IFN- γ [61], IL-15 [62] and IL-17 [63], leading to the hypothesis that BPH may represent an 'immune inflammatory' disease [11]. This is an attractive hypothesis as the association of BPH with chronic inflammation could offer a sound framework for understanding the disease pathogenesis. Notably, upregulation of IL-17 in BPH tissues was associated with pathological conditions [63] well before the establishment of a pathogenic role of Th17 cells in autoimmune diseases.

Significantly increased levels of the proinflammatory cytokines IL-1 α , IL-1 β , IL-6 and IL-12p70, and the chemokines CCL1, CCL4, CCL22 and IL-8, are present in the seminal plasma of BPH patients [45]. The concomitant increase in several inflammatory cytokines and chemokines in BPH patients is consistent with an important chronic inflammatory component in the pathogenesis of the disease, and expression-profiling data demonstrate a strong correlation between inflammation and symptomatic BPH [64]. IL-8 is expressed *in situ* by epithelial and stromal prostate cells and is functional, as shown by the recruitment of CXCR1- and CXCR2-positive leukocytes as well as CD15⁺ neutrophils [45]. The potential value of IL-8 as a surrogate marker of disease is further supported by the positive correlation of IL-8 levels with symptom scores and serum PSA values in BPH patients [45]. Therefore, IL-8 may not only serve as a reliable biomarker applicable to the diagnosis, prognosis and assessment of treatment efficacy in BPH, as well as in CP/CPPS patients, but also actually represents an important driver of prostate inflammation, and is an interesting therapeutic target in itself for the treatment of this condition. IL-8-mediated

BPH cell growth can be induced by a combination of IFN- γ and IL-17, thus establishing a possible relationship between the autoimmune response induced by BPH cells and prostate cell growth (FIGURE 1). Consistent with an IL-8-dependent link bridging the inflammatory response and cell growth in BPH cells, the vitamin D receptor (VDR) agonist elocalcitol, a well-defined anti-inflammatory and antiproliferative agent inhibiting prostate growth in experimental models [65,66] and in BPH patients [67], has been shown to inhibit IL-8-mediated prostate growth and inflammation through multiple mechanisms of action [29,68].

Therefore, BPH has all the features of a chronic inflammatory disease. It remains to be seen whether or not this chronic inflammation is of an autoimmune origin, but the circumstantial evidence available would suggest this is a distinct possibility.

Expert commentary & five-year view

A key novel element in the pathogenesis of CP/CPPS and BPH is provided by the observation that human prostate cells can act as APCs that are able to stimulate alloreactive CD4⁺ T cells, and potentially also autoreactive T cells specific for prostate antigens, to produce IFN- γ and IL-17. The induction of a BPH cell-driven autoimmune response, as well as the triggering of TLRs expressed by BPH cells, upregulate the production of IL-8, IL-6 and CXCL10, key factors for sustaining prostate inflammation, recruiting inflammatory leukocytes and promoting prostate cell hyperplasia.

Thus, BPH and CP/CPPS could share common pathogenetic mechanisms, based on the capacity of human stromal prostate cells to act as inducers and targets of chronic inflammation. Critically important for the development of both diseases could be the capacity of prostate cells to act as APCs and to respond to TLR agonists, producing proinflammatory cytokines and chemokines able to recruit infiltrating cells and promote prostate cell growth. The antigen-presenting capacity of prostate cells is also a key element defining a putative autoimmune origin for CP/CPPS and BPH. The autoimmune nature of CP/CPPS is fairly well established, with multiple prostate antigens already identified as being capable of inducing a memory T-cell response in peripheral blood lymphocytes from CP/CPPS patients (TABLE 1). Conversely, firm evidence for an autoimmune origin of BPH is still lacking, although a single study has reported autoreactive T cells specific for prostate antigens in BPH patients [18]. Nevertheless, the available evidence for chronic inflammation in BPH and the similarities between BPH and CP/CPPS, a disease in which the autoimmune origin is fairly obvious, also suggest an autoimmune pathogenesis for BPH.

Future work will clarify these issues, in particular the putative autoimmune nature of BPH, and hopefully provide a more detailed analysis of the inciting prostatic autoantigens. In any case, the marked inflammatory component shared by CP/CPPS and BPH should spur attempts to treat these conditions using anti-inflammatory agents. In this respect, the limited experience available is too fragmentary for a thorough assessment [69–71]. Nevertheless, the VDR agonist elocalcitol, an antiproliferative

and anti-inflammatory agent able to inhibit prostate growth and inflammation in preclinical models of BPH [65,72], has been shown to arrest prostate growth in BPH patients [67], indicating the beneficial effects of targeting inflammation in this disease and further supporting the relevance of the inflammatory response in the pathogenesis of BPH. In the next few years, we will certainly witness additional work along these lines.

Financial & competing interests disclosure

The authors have no relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript. This includes employment, consultancies, honoraria, stock ownership or options, expert testimony, grants or patents received or pending, or royalties.

No writing assistance was utilized in the production of this manuscript.

Key issues

- Prostate autoimmunity has been clearly demonstrated in a variety of animal models.
- The demonstration that human prostate cells are antigen-presenting cells strongly supports the autoimmune nature of chronic prostate inflammatory diseases such as chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS). Based on the available evidence, CP/CPPS can be considered an autoimmune disease.
- The inciting autoantigen in CP/CPPS is not known, but several candidates have been proposed among the prostate-specific antigens.
- Besides infections, intraprostatic reflux, frequent or prolonged contraction of smooth muscle in the urethra/bladder neck and prostatic stroma, increased intraprostatic pressure and hormonal imbalances have all been recognized as potential risk factors for CP/CPPS, and could precipitate autoimmune responses against prostate antigens.
- Chronic inflammation is associated with benign prostatic hyperplasia (BPH), and BPH stromal cells function as antigen-presenting cells that are able to activate T cells. However, an autoimmune origin of the chronic inflammatory response in BPH has not yet been formally demonstrated.
- Appropriate anti-inflammatory therapies are predicted to be beneficial in CP/CPPS and BPH patients.

References

Papers of special note have been highlighted as:

- of interest
- of considerable interest

- Nickel JC. The overlapping lower urinary tract symptoms of benign prostatic hyperplasia and prostatitis. *Curr. Opin. Urol.* 16(1), 5–10 (2006).
- Steiner GE, Djavan B, Kramer G *et al.* The picture of the prostatic lymphokine network is becoming increasingly complex. *Rev. Urol.* 4(4), 171–177 (2002).
- Krieger JN, Lee SW, Jeon J *et al.* Epidemiology of prostatitis. *Int. J. Antimicrob. Agents* 31(Suppl. 1), S85–S90 (2008).
- Krieger JN, Nyberg L Jr, Nickel JC. NIH consensus definition and classification of prostatitis. *JAMA* 282(3), 236–237 (1999).
- Schaeffer AJ. Clinical practice. Chronic prostatitis and the chronic pelvic pain syndrome. *N. Engl. J. Med.* 355(16), 1690–1698 (2006).
- Roehrborn CG. Pathology of benign prostatic hyperplasia. *Int. J. Impot. Res.* 20(Suppl. 3), S11–S18 (2008).
- Whitmore WF, Gittes RF. Studies on the prostate and testis as immunologically privileged sites. *Cancer Treat. Rep.* 61(2), 217–222 (1977).
- Vykhovanets EV, Resnick MI, Marengo SR. The healthy rat prostate contains high levels of natural killer-like cells and unique subsets of CD4⁺ helper-inducer T cells: implications for prostatitis. *J. Urol.* 173(3), 1004–1010 (2005).
- Steiner G, Gessl A, Kramer G *et al.* Phenotype and function of peripheral and prostatic lymphocytes in patients with benign prostatic hyperplasia. *J. Urol.* 151(2), 480–484 (1994).
- Kramer G, Steiner GE, Handisurya A *et al.* Increased expression of lymphocyte-derived cytokines in benign hyperplastic prostate tissue, identification of the producing cell types, and effect of differentially expressed cytokines on stromal cell proliferation. *Prostate* 52(1), 43–58 (2002).
- Kramer G, Mitteregger D, Marberger M. Is benign prostatic hyperplasia (BPH) an immune inflammatory disease? *Eur. Urol.* 51(5), 1202–1216 (2007).
- Well-argued review, arguing for a pathogenetic role of chronic inflammation in benign prostatic hyperplasia (BPH). Discusses key immunologic features of the inflammatory response in BPH.
- Konig JE, Senge T, Allhoff EP, König W. Analysis of the inflammatory network in benign prostate hyperplasia and prostate cancer. *Prostate* 58(2), 121–129 (2004).
- Nishimura M, Naito S. Tissue-specific mRNA expression profiles of human Toll-like receptors and related genes. *Biol. Pharm. Bull.* 28(5), 886–892 (2005).
- Gatti G, Rivero V, Motrich RD, Maccioni M. Prostate epithelial cells can act as early sensors of infection by up-regulating TLR4 expression and proinflammatory mediators upon LPS stimulation. *J. Leukoc. Biol.* 79(5), 989–998 (2006).
- Penna G, Fibbi B, Amuchastegui S *et al.* Human benign prostatic hyperplasia stromal cells as inducers and targets of chronic immuno-mediated inflammation. *J. Immunol.* 182(7), 4056–4064 (2009).
- Provides the first evidence for the antigen-presenting capacity of prostate cells, a finding with far-reaching implications for the autoimmune pathogenesis of prostate diseases such as chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS) and BPH.
- Medzhitov R. Recognition of microorganisms and activation of the immune response. *Nature* 449(7164), 819–826 (2007).
- Zisman A, Zisman E, Lindner A *et al.* Autoantibodies to prostate specific antigen in patients with benign prostatic hyperplasia. *J. Urol.* 154(3), 1052–1055 (1995).
- Zisman A, Lindner A, Zisman E, Lindner U, Mozes E. Prostate-specific antigen induces proliferation of peripheral blood lymphocytes and cytokine secretion in benign prostate hypertrophy patients. *Eur. Urol.* 36(3), 258–265 (1999).
- Dardalhon V, Korn T, Kuchroo VK, Anderson AC. Role of Th1 and Th17 cells in organ-specific autoimmunity. *J. Autoimmun.* 31(3), 252–256 (2008).
- Motrich RD, Maccioni M, Riera CM, Rivero VE. Autoimmune prostatitis: state of the art. *Scand J. Immunol.* 66(2–3), 217–227 (2007).

- **Thorough review, based on animal and human studies, supporting the autoimmune pathogenesis of CP/CPPS.**
- 21 Pontari MA, Ruggieri MR. Mechanisms in prostatitis/chronic pelvic pain syndrome. *J. Urol.* 179(5 Suppl.), S61–S67 (2008).
- 22 Vykhovanets EV, Resnick MI, MacLennan GT, Gupta S. Experimental rodent models of prostatitis: limitations and potential. *Prostate Cancer Prostatic Dis.* 10(1), 15–29 (2007).
- **Comprehensive review of the animal models for prostatitis.**
- 23 Liu KJ, Chatta GS, Twardzik DR *et al.* Identification of rat prostatic steroid-binding protein as a target antigen of experimental autoimmune prostatitis: implications for prostate cancer therapy. *J. Immunol.* 159(1), 472–480 (1997).
- **First report identifying and characterizing prostatic steroid-binding protein (PSBP) as a potent prostatitis-inducing autoantigen.**
- 24 Penna G, Amuchastegui S, Cossetti C *et al.* Spontaneous and prostatic steroid binding protein peptide-induced autoimmune prostatitis in the nonobese diabetic mouse. *J. Immunol.* 179(3), 1559–1567 (2007).
- **Evidence for spontaneous prostatitis development in NOD, a mouse strain prone to the development of multiple autoimmune diseases, and detailed characterization of PSBP peptide-induced autoimmune prostatitis.**
- 25 Kikutani H, Makino S. The murine autoimmune diabetes model: NOD and related strains. *Adv. Immunol.* 51, 285–322 (1992).
- 26 Rivero VE, Cailleau C, Depiante-Depauli M, Riera CM, Carnaud C. Non-obese diabetic (NOD) mice are genetically susceptible to experimental autoimmune prostatitis (EAP). *J. Autoimmun.* 11(6), 603–610 (1998).
- **First report of induced experimental prostatitis in the NOD mouse. This study represents a cornerstone in the definition of experimental autoimmune prostatitis features in this mouse strain.**
- 27 Rivero V, Carnaud C, Riera CM. Prostatein or steroid binding protein (PSBP) induces experimental autoimmune prostatitis (EAP) in NOD mice. *Clin. Immunol.* 105(2), 176–184 (2002).
- 28 Maccioni M, Rivero VE, Riera CM. Prostatein (or rat prostatic steroid binding protein) is a major autoantigen in experimental autoimmune prostatitis. *Clin. Exp. Immunol.* 112(2), 159–165 (1998).
- 29 Penna G, Amuchastegui S, Cossetti C *et al.* Treatment of experimental autoimmune prostatitis in nonobese diabetic mice by the vitamin D receptor agonist elocalcitol. *J. Immunol.* 177(12), 8504–8511 (2006).
- 30 Taguchi O, Kojima A, Nishizuka Y. Experimental autoimmune prostatitis after neonatal thymectomy in the mouse. *Clin. Exp. Immunol.* 60(1), 123–129 (1985).
- 31 Taguchi O, Nishizuka Y. Self tolerance and localized autoimmunity. Mouse models of autoimmune disease that suggest tissue-specific suppressor T cells are involved in self tolerance. *J. Exp. Med.* 165(1), 146–156 (1987).
- 32 Taguchi O, Kontani K, Ikeda H *et al.* Tissue-specific suppressor T cells involved in self-tolerance are activated extrathymically by self-antigens. *Immunology* 82(3), 365–369 (1994).
- 33 Seethalakshmi L, Bala RS, Malhotra RK *et al.* 17 β -estradiol induced prostatitis in the rat is an autoimmune disease. *J. Urol.* 156(5), 1838–1842 (1996).
- 34 Harris MT, Feldberg RS, Lau KM, Lazarus NH, Cochrane DE. Expression of proinflammatory genes during estrogen-induced inflammation of the rat prostate. *Prostate* 44(1), 19–25 (2000).
- 35 Wilson MJ, Woodson M, Wiehr C, Reddy A, Sinha AA. Matrix metalloproteinases in the pathogenesis of estradiol-induced nonbacterial prostatitis in the lateral prostate lobe of the Wistar rat. *Exp. Mol. Pathol.* 77(1), 7–17 (2004).
- 36 Corman JM, Sercarz EE, Nanda NK. Recognition of prostate-specific antigenic peptide determinants by human CD4 and CD8 T cells. *Clin. Exp. Immunol.* 114(2), 166–172 (1998).
- 37 Alexander RB, Brady F, Ponniah S. Autoimmune prostatitis: evidence of T cell reactivity with normal prostatic proteins. *Urology* 50(6), 893–899 (1997).
- **First evidence for the autoimmune origin of CP/CPPS. A seminal study presenting direct evidence for autoimmune responses in CP/CPPS patients.**
- 38 Batstone GR, Doble A, Gaston JS. Autoimmune T cell responses to seminal plasma in chronic pelvic pain syndrome (CPPS). *Clin. Exp. Immunol.* 128(2), 302–307 (2002).
- 39 Ponniah S, Arah I, Alexander RB. PSA is a candidate self-antigen in autoimmune chronic prostatitis/chronic pelvic pain syndrome. *Prostate* 44(1), 49–54 (2000).
- 40 Motrich RD, Maccioni M, Molina R *et al.* Presence of INF γ -secreting lymphocytes specific to prostate antigens in a group of chronic prostatitis patients. *Clin. Immunol.* 116(2), 149–157 (2005).
- **Identification of IFN- γ -producing cells specific for two prostate antigens, prostate-specific antigen and prostatic acid phosphatase, in CP/CPPS patients.**
- 41 Dunphy EJ, Eickhoff JC, Muller CH, Berger RE, McNeel DG. Identification of antigen-specific IgG in sera from patients with chronic prostatitis. *J. Clin. Immunol.* 24(5), 492–502 (2004).
- 42 Klyushnenkova EN, Link J, Oberle WT *et al.* Identification of HLA-DRB1*1501-restricted T-cell epitopes from prostate-specific antigen. *Clin. Cancer Res.* 11(8), 2853–2861 (2005).
- 43 Klyushnenkova EN, Kouivaskia DV, Kodak JA, Vandenbark AA, Alexander RB. Identification of HLA-DRB1*1501-restricted T-cell epitopes from human prostatic acid phosphatase. *Prostate* 67(10), 1019–1028 (2007).
- 44 Kodak JA, Mann DL, Klyushnenkova EN, Alexander RB. Activation of innate immunity by prostate specific antigen (PSA). *Prostate* 66(15), 1592–1599 (2006).
- 45 Penna G, Mondaini N, Amuchastegui S *et al.* Seminal plasma cytokines and chemokines in prostate inflammation: interleukin-8 as a predictive biomarker in chronic prostatitis/chronic pelvic pain syndrome and benign prostatic hyperplasia. *Eur. Urol.* 51, 524–533 (2007).
- **First study reporting elevated levels of cytokines and chemokines in seminal fluids from BPH patients. Provides clinical validation for the hypothesis of a pathogenetic role of inflammation in BPH.**
- 46 Rivero VE, Motrich RD, Maccioni M, Riera CM. Autoimmune etiology in chronic prostatitis syndrome: an advance in the understanding of this pathology. *Crit. Rev. Immunol.* 27(1), 33–46 (2007).
- 47 Eriksson U, Ricci R, Hunziker L *et al.* Dendritic cell-induced autoimmune heart failure requires cooperation between adaptive and innate immunity. *Nat. Med.* 9(12), 1484–1490 (2003).
- 48 Anders HJ, Vielhauer V, Eis V *et al.* Activation of Toll-like receptor-9 induces progression of renal disease in MRL-Fas(lpr) mice. *FASEB J.* 18(3), 534–536 (2004).
- 49 Pontari MA. Etiologic theories of chronic prostatitis/chronic pelvic pain syndrome. *Curr. Urol. Rep.* 8(4), 307–312 (2007).

- 50 Schaeffer AJ, Knauss JS, Landis JR *et al.* Leukocyte and bacterial counts do not correlate with severity of symptoms in men with chronic prostatitis: the National Institutes of Health Chronic Prostatitis Cohort Study. *J. Urol.* 168(3), 1048–1053 (2002).
- 51 Tsuboi N, Nishimura T, Chen H *et al.* Relation of leukocytosis in prostatic fluid and inflamed prostatic tissue. *J. Nippon. Med. Sch.* 74(3), 210–216 (2007).
- 52 Nickel JC, Roehrborn CG, O'Leary MP *et al.* The relationship between prostate inflammation and lower urinary tract symptoms: examination of baseline data from the REDUCE trial. *Eur. Urol.* 54(6), 1379–1384 (2008).
- 53 Straub RH. The complex role of estrogens in inflammation. *Endocr. Rev.* 28(5), 521–574 (2007).
- 54 Webber R. Benign prostatic hyperplasia. *Clin. Evid.* (15), 1213–1226 (2006).
- 55 Lee KL, Peehl DM. Molecular and cellular pathogenesis of benign prostatic hyperplasia. *J. Urol.* 172(5 Pt 1), 1784–1791 (2004).
- 56 Lucia MS, Lambert JR. Growth factors in benign prostatic hyperplasia: basic science implications. *Curr. Urol. Rep.* 9(4), 272–278 (2008).
- 57 Nickel JC. Inflammation and benign prostatic hyperplasia. *Urol. Clin. North Am.* 35(1), 109–115; vii (2008).
- **Authoritative review discussing the multiple aspects of the association between inflammation and BPH.**
- 58 Sciarra A, Mariotti G, Salciccia S *et al.* Prostate growth and inflammation. *J. Steroid. Biochem. Mol. Biol.* 108(3–5), 254–260 (2008).
- 59 Nickel JC, Downey J, Young I, Boag S. Asymptomatic inflammation and/or infection in benign prostatic hyperplasia. *BJU Int.* 84(9), 976–981 (1999).
- 60 Roehrborn CG. Definition of at-risk patients: baseline variables. *BJU Int.* 97(Suppl. 2), 7–11; discussion 21–12 (2006).
- 61 Steiner GE, Stix U, Handisurya A *et al.* Cytokine expression pattern in benign prostatic hyperplasia infiltrating T cells and impact of lymphocytic infiltration on cytokine mRNA profile in prostatic tissue. *Lab. Invest.* 83(8), 1131–1146 (2003).
- 62 Handisurya A, Steiner GE, Stix U *et al.* Differential expression of interleukin-15, a pro-inflammatory cytokine and T-cell growth factor, and its receptor in human prostate. *Prostate* 49(4), 251–262 (2001).
- 63 Steiner GE, Newman ME, Paikl D *et al.* Expression and function of pro-inflammatory interleukin IL-17 and IL-17 receptor in normal, benign hyperplastic, and malignant prostate. *Prostate* 56(3), 171–182 (2003).
- **Reports abundant expression of IL-17 and the IL-17 receptor by BPH-related T cells, well before the establishment of an important role for Th17 cells in the pathogenesis of autoimmune diseases.**
- 64 Prakash K, Pirozzi G, Elashoff M *et al.* Symptomatic and asymptomatic benign prostatic hyperplasia: molecular differentiation by using microarrays. *Proc. Natl Acad. Sci. USA* 99(11), 7598–7603 (2002).
- 65 Crescioli C, Ferruzzi P, Caporali A *et al.* Inhibition of prostate cell growth by BXL-628, a calcitriol analogue selected for a Phase II clinical trial in patients with benign prostate hyperplasia. *Eur. J. Endocrinol.* 150(4), 591–603 (2004).
- 66 Adorini L, Penna G, Amuchastegui S *et al.* Inhibition of prostate growth and inflammation by the vitamin D receptor agonist BXL-628 (elocalcitrol). *J. Steroid Biochem. Mol. Biol.* 103(3–5), 689–693 (2007).
- 67 Colli E, Rigatti P, Montorsi F *et al.* BXL628, a novel vitamin D3 analog arrests prostate growth in patients with benign prostatic hyperplasia: a randomized clinical trial. *Eur. Urol.* 49(1), 82–86 (2006).
- 68 Morelli A, Vignozzi L, Filippi S *et al.* BXL-628, a vitamin D receptor agonist effective in benign prostatic hyperplasia treatment, prevents RhoA activation and inhibits RhoA/Rho kinase signaling in rat and human bladder. *Prostate* 67(3), 234–247 (2007).
- 69 Bates S, Talbot M. Short course oral prednisolone therapy in chronic abacterial prostatitis and prostatodynia: case reports of three responders and one non-responder. *Sex. Transm. Infect.* 76(5), 398–399 (2000).
- 70 Nickel JC. The three As of chronic prostatitis therapy: antibiotics, α -blockers and anti-inflammatories. What is the evidence? *BJU Int.* 94(9), 1230–1233 (2004).
- 71 Dhar NB, Shoskes DA. New therapies in chronic prostatitis. *Curr. Urol. Rep.* 8(4), 313–318 (2007).
- 72 Adorini L, Penna G. Control of autoimmune diseases by the vitamin D endocrine system. *Nat. Clin. Pract. Rheumatol.* 4, 404–412 (2008).
- 73 Galmarini M, Riera CM, Pesoa S, Vullo C, Vottero-Cima E. Induction of humoral autoimmune response to rat male accessory glands. *Medicina (B. Aires)* 41(3), 349–353 (1981).
- 74 Pacheco-Rupil B, Depiante-Depaoli M, Casadio B. Experimental autoimmune damage to rat male accessory glands. II. T cell requirement in adoptive transfer of specific tissue damage. *Am. J. Reprod. Immunol.* 5(1), 15–19 (1984).
- 75 Depiante-Depaoli M, Pacheco-Rupil B, Britos S, Casas A. Experimental autoimmune damage to rat male accessory glands. I. Transfer of autoimmune response by spleen cells. *Am. J. Reprod. Immunol.* 5(1), 9–14 (1984).
- 76 Depiante-Depaoli M, Pacheco-Rupil B. Experimental autoimmunity to rat male accessory glands (MAG): circulating antibodies, immunoglobulins bound to target glands, and immunoglobulins-secreting cells. *Am. J. Reprod. Immunol. Microbiol.* 7(1), 32–38 (1985).
- 77 Casas-Ingaramo A, Depiante-Depaoli M, Pacheco-Rupil B. Activation of cytotoxic cells by syngeneic prostate antigens in experimental autoimmune vesiculo-prostatitis. *Autoimmunity* 9(2), 151–157 (1991).
- 78 Rivero VE, Iribarren P, Riera CM. Mast cells in accessory glands of experimentally induced prostatitis in male Wistar rats. *Clin. Immunol. Immunopathol.* 74(3), 236–242 (1995).
- 79 Maccioni M, Rivero V, Riera CM. Autoantibodies against rat prostate antigens. Association of specific IGG2b and IGG2c with the DTH response. *J. Autoimmun.* 9(4), 485–491 (1996).
- 80 Galmarini M, Ferro ME, Riera CM. Delayed hypersensitivity and lesions following isoimmunization with modified rat male accessory glands: kinetics of induction. *J. Reprod. Immunol.* 13(2), 147–157 (1988).
- 81 Donadio AC, Gagliano H, Remedi MM, Nowotny E, Depiante-Depaoli M. Time-course study of cellular immune response and testosterone metabolism in an autoimmune model for chronic prostatic inflammation. *J. Urol.* 160(4), 1546–1550 (1998).
- 82 Keetch DW, Humphrey P, Ratliff TL. Development of a mouse model for nonbacterial prostatitis. *J. Urol.* 152(1), 247–250 (1994).
- 83 Correale P, Walmsley K, Nieroda C *et al.* *In vitro* generation of human cytotoxic T lymphocytes specific for peptides derived from prostate-specific antigen. *J. Natl Cancer Inst.* 89(4), 293–300 (1997).

- 84 Xue BH, Zhang Y, Sosman JA, Peace DJ. Induction of human cytotoxic T lymphocytes specific for prostate-specific antigen. *Prostate* 30(2), 73–78 (1997).
- 85 Correale P, Walmsley K, Zaremba S *et al.* Generation of human cytolytic T lymphocyte lines directed against prostate-specific antigen (PSA) employing a PSA oligopeptide peptide. *J. Immunol.* 161(6), 3186–3194 (1998).
- 86 Berlyn KA, Schultes B, Leveugle B *et al.* Generation of CD4⁺ and CD8⁺ T lymphocyte responses by dendritic cells armed with PSA/anti-PSA (antigen/antibody) complexes. *Clin. Immunol.* 101(3), 276–283 (2001).
- 87 Harada M, Kobayashi K, Matsueda S *et al.* Prostate-specific antigen-derived epitopes capable of inducing cellular and humoral responses in HLA-A24⁺ prostate cancer patients. *Prostate* 57(2), 152–159 (2003).
- 88 Hural JA, Friedman RS, McNabb A *et al.* Identification of naturally processed CD4 T cell epitopes from the prostate-specific antigen kallikrein 4 using peptide-based *in vitro* stimulation. *J. Immunol.* 169(1), 557–565 (2002).
- 89 Fong L, Ruegg CL, Brockstedt D, Engleman EG, Laus R. Induction of tissue-specific autoimmune prostatitis with prostatic acid phosphatase immunization: implications for immunotherapy of prostate cancer. *J. Immunol.* 159(7), 3113–3117 (1997).
- 90 McNeel DG, Nguyen LD, Disis ML. Identification of T helper epitopes from prostatic acid phosphatase. *Cancer Res.* 61(13), 5161–5167 (2001).
- Mario Maggi
Andrology Unit, Department of Clinical Physiopathology, University of Florence, 50139 Florence, Italy
Tel.: +39 055 427 1415
Fax: +39 055 427 1413
m.maggi@dfc.unifi.it
 - Luciano Adorini
Chief Scientific Officer,
Intercept Pharmaceuticals, Via P. Togliatti 22 bis, 06073 Corciano (Perugia), Italy
Tel.: +39 075 792 1934
Fax: +39 075 792 1696
ladorini@interceptpharma.com

Affiliations

- Giuseppe Penna
Intercept Pharmaceuticals, 06073 Corciano (Perugia), Italy
Tel.: +39 075 792 1934
Fax: +39 075 792 1696
gpenna@interceptpharma.com
- Benedetta Fibbi
Andrology Unit, Department of Clinical Physiopathology, University of Florence, 50139 Florence, Italy
Tel.: +39 055 427 1487
Fax: +39 055 427 1375
benedetta.fibbi@unifi.it

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.