Use of vaccines for prophylaxis of urinary tract infections

O uso de vacinas na profilaxia das infecções do trato urinário

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Abstract

The urinary tract is the most common site of bacterial infections. Urinary tract infections (UTIs) in women without urinary tract anatomic abnormalities require frequent and repeated use of antibiotics, increasing the prevalence of antimicrobial-resistant microorganisms. The possibility of an alternative approach, with the use of vaccines produced from inactivated bacteria or structural components of these microorganisms, is a reality. Confirming the results observed experimentally, controlled clinical studies of oral or vaginal immunotherapy have shown reductions in the number of episodes of recurrence, without significant side-effects. We reviewed the mechanisms of aggression and defense involved in the pathogenesis of UTIs in women with anatomically normal urinary tracts, the evolution of knowledge about the immunotherapy of UTIs, and the vaccines already available or under development for the treatment of this important clinical condition.

Keywords: Urinary tract infections. Therapeutics. Recurrence.

Resumo

O trato urinário é o sítio mais comum de infecção bacteriana. As infecções do trato urinário (ITU) recorrentes em mulheres sem anormalidades anatômicas do trato urinário demandam uso frequente e repetido de antibióticos, aumentando a prevalência de micro-organismos resistentes aos antimicrobianos. A possibilidade de abordagem alternativa, com a utilização de vacinas produzidas a partir de bactérias inativadas ou componentes estruturais desses micro-organismos, é uma realidade palpável. Confirmando resultados observados experimentalmente, estudos clínicos controlados têm mostrado redução dos episódios de recorrência, sem efeitos colaterais significativos, com imunoterapia oral ou vaginal. Nesta revisão, foram apresentados os mecanismos de agressão e defesa envolvidos na gênese das infecções urinárias em mulheres com trato urinário normal, a evolução do conhecimento sobre a imunoterapia nas ITU e as vacinas já disponíveis ou em desenvolvimento para o tratamento dessa importante condição clínica.

Palavras-chave: Infecções urinárias. Terapêutica. Recidiva.

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Introduction

Urinary tract infections (UTIs), characterized by the presence of a pathogenic microorganism somewhere in the urinary tract,[1] may be caused by any pathogen (fungi, parasites, viruses or bacteria) which can colonize this anatomic site. The most common agents are the enterobacteria,[2] *Escherichia coli* alone accounting for 80% of the cases.[3-6] UTI development is determined by the virulence of the invading microorganism, inoculum size and failure of the host`s defense mechanisms.[3,4,7]

Pathogenic bacteria have virulence factors which allow them to adhere to the urinary epithelium, with further multiplication and colonization of the urinary tract.[7,8] Adhesion is mediated by specific interactions between components of the bacterial surface (adhesins) and receptors on the host`s cells. Uropathogenic *E. coli* adhesins correspond to hair-like fimbriae or non-hair-like proteins on the external surface of the membrane.[5] Three different types of fimbriae have been identified: type 1 (FimH), type P (PapG) and

type S. Over 90% of *E. coli* causing pyelonephritis have type P fimbriae, which interact with glycolipid receptors. Type 1 fimbriae bind to glycoprotein receptors which express mannose in their binding sites. Binding of type 1 fimbriae to mannose allows *E. coli* to colonize the urinary epithelium, while P fimbriae trigger the inflammatory cascade. Although immunization against P fimbria receptors may prevent infection, strategies targeting colonization seem to be more effective.[3,9] Different fimbria types may also be found in a same strain of *Proteus mirabilis* (MR/P, UCA, PMF), all associated with the mechanism of UTI caused by this agent.[10,11]

An ability to compete with the host for iron storages is another virulence mechanism. Some pathogenic bacteria have aerobactin and enterobactin, substances that play a role in iron uptake.[5,12]

The main defense mechanism against uropathogens is constant urine flow.[13] The low pH, polymorphonuclear cells, Tamm-Horsfall glycoprotein, urea concentration and osmolarity are specific characteristics that inhibit bacterial adhesion to the bladder mucosa.[4,7,8] Bacterial destruction involves the complement system and local mucosa IgA production, directed against the bacterial surface. Acquired specific IgM and IgG-mediated serum immune response follows acute pyelonephritis, between 7 and 10 days after infection onset. Urinary antibodies (secretory IgA and serum IgM and IgG) bind to bacterial structures, such as fimbriae and O and K antigens, so as to facilitate their elimination.[4,7,8]

Increasing pathogen resistance and the slow pace of development of new antimicrobials may hamper the treatment not only of UTIs but also of other infections. The development of new strategies to reduce the induction of bacterial resistance without affecting treatment efficacy is paramount.[14-18] Several studies have shown the action of cranberry extract, a natural compound, in the prevention of recurring urinary infections.[19-22] The use of vaccines for UTI prophylaxis may be a promising alternative.

We review the main scientific evidence concerning immunotherapy as a strategy for UTI prevention.

History

The use of probiotics, composed of *E. coli* strains, was started in the 1920s for treatment of chronic infectious and inflammatory bowel diseases.[23] The rationale for the use of bacterial substrates as a way of stimulating the immune system and reducing recurring UTIs arose 40 years ago, when knowledge about the potentially involved immune mechanisms was limited.

E. coli extract was reported in the 1980s to reduce the incidence of UTIs in adults, children and pregnant women.[24] Frey *et al*., in 1986, observed a significant reduction of antibiotic use by women with recurring cystitis treated with a bacterial extract.[25] Extracts composed of whole bacteria or their fragments protected some individuals. Fimbriae and several other molecules with expression on the bacterial surface were studied as potential targets for the development of vaccines.[26] In the past few years, better understanding of the immune response mechanisms has consolidated the use of these substances as immunostimulants.[27]

Scientific background

Vaccines are biological preparations used to establish or improve humoral immunity against a specific disease.[28] Adaptive immune response starts when the invading pathogen manages to evade the defense mechanisms linked to the innate immune response. The efficacy of the former is related to the interaction of three cell types: antigen-presenting cells, thymus-derived T lymphocytes and bone-marrow-derived B lymphocytes. Antigen-presenting cells capture, process and present the antigen to the T cells, for recognition by the surface cell receptor. B cell surface receptors, that is, immunoglobulins, directly recognize the antigen, in a process that leads to the production of plasma cells that can secrete antibody sub-classes (IgA, IgE, IgG and IgM). These antibodies have a role in the prevention or limitation of the initial phases of the infection, and are involved in the destruction of the infected cells through antibody-dependent cytotoxicity or complement-mediated lysis.[28]

Immunologic memory allows a fast increase of the response after another antigen challenge. This effect plays an important role in the function of the immune system, being one of the cornerstones of vaccination.[28]

Available vaccines or vaccines being developed

OM-89 (Uro-Vaxom)

OM-89, a bacterial extract consisting of components from 18 strains of uropathogenic *E. coli*, can stimulate the immune system through several mechanisms. The product is marketed in capsules for oral administration.[24,26,27,29-32]

Several *in vitro* studies have shown that OM-89 leads to the production of tumor-necrosis factor alpha (TNF-α), gamma interferon and interleukins 1 and 6 by peripheral blood monocytes, besides stimulating B

lymphocytes, the production of antibodies against *E. coli* and the phagocytic activity of macrophages and natural killer cells.[26,29-34]

Sedelmeier & Bessler concluded that multiple oral administrations of *E. coli* extract induce the dose-dependent production of specific serum antibodies of the IgG and IgM types.[34] Huber *et al.* showed that the antibodies obtained could recognize and bind to the 18 *E. coli* strains and other bacterial strains commonly isolated from UTI patients, such as *Enterococcus faecalis*, *Klebsiella pneumoniae* and *Proteus mirabilis*.[35] Nauck *et al.* confirmed OM-89 ability to stimulate the activity of rabbit polymorphonuclear leukocytes against *E. coli* strains.[36]

Tammen studied 120 patients with recurring UTI. Recurrence rate was significantly lower in the group receiving *E. coli* extract, both during the 3-month treatment period and during the following months.[37] Schulman *et al.* reported a 0.7 recurrence rate in treated patients, against 1.5 in the placebo group.[38] Magasi *et al.* also showed a significant effect of the *E. coli* extract in the prevention of recurring UTI. During the study period, 13.8% of the patients who received the extract had a recurrence, against 79.6% in the placebo group ($p < 0.0005$).[39]

Bauer *et al.* analyzed five double-blind, placebo-controlled studies demonstrating a protection afforded by OM-89 against recurrent UTIs.[32] In a multicenter, double-blind, placebo-controlled study of 454 women, the same authors found a significant reduction of the recurrence rate of UTIs with *E. coli* extract.[24]

Similar results were obtained by Naber *et al.*, in a meta-analysis that included 5 double-blind, placebo controlled clinical trials. UTI incidence was significantly lower in OM-89-treated patients.[27]

In the past decades, *E. coli* extract has proved efficient and safe. The most frequently reported side-effects were headache and gastrointestinal upset, although at a rate similar to that observed in controls. There have been no worrying or unexpected side-effects.[26,30,32]

Solco Urovac

Solco Urovac is a vaccine consisting of 10 strains of inactivated uropathogenic bacteria. Six *E. coli* serotypes and strains of *Proteus mirabilis, Morganella morganii, Klebsiella pneumoniae* and *Enterococcus faecalis* make up the vaccine, which is administered as a vaginal suppository. Its efficacy was demonstrated in the second phase of two independent studies which indicated that Solco Urovac may be an alternative to the antibiotic-based prophylactic regimens in women with recurrent UTIs.[40,41]

Hopkins *et al.*, in a double-blind, placebo-controlled study, randomized 75 patients into three groups: group I, receiving primary immunization with the vaccine without booster doses, group II, receiving primary immunization and booster doses, and group III, receiving a placebo. UTI rate was higher in group III, compared with the group receiving the booster doses. Sexually active women under the age of 52 years who received the vaccine and booster doses had a significantly lower rate of *E. coli* – related UTIs. Fever, vaginal bleeding and rash, nausea and headache were the most frequently reported side-effects. There was no significant difference in the rate of side-effects and in the serum and vaginal levels of antibodies among the three groups.[40]

Similar results were reported by Uehling *et al.* in a double-blind, placebo-controlled study. The UTI-free time-interval was longer in patients who received the vaccine.[41]

FimH Adhesin

Type 1 fimbriae, found in some strains of *E. coli*, are heteropolymers composed of a larger subunit (FimA) and three smaller subunits (FimF, FimG and FimH).[42] There is evidence suggesting that these fimbriae are an important factor to initiate bacterial UTIs, as they promotes epithelial adhesion and urinary tract colonization. Thankavel *et al.* assessing the humoral response to intramuscular and subcutaneous immunizations, observed immunogenicity of the FimH subunit in rats. Serum antibodies against components of the FimH subunit protected against bacterial colonization *in vivo*. Protection involved blockade of bacterial adhesion to the uroepithelium. Immunized animals had a lower incidence of *E. coli* caused experimental cystitis, with a markedly higher bladder level of antibodies against FimH.[42] Likewise, Langermann & Ballou showed that the systemic immunization of rats with a FimH-based vaccine resulted in higher serum levels of IgG antibodies and blockade of bacterial adhesion.[43]

Langermann *et al.* administered an FimH-based intramuscular vaccine to primates.[44] After 2 initial doses and a booster one after 48 weeks, the authors concluded that the vaccine induced protective immunity. Serum and vaginal IgG antibodies against FimH were identified in the immunized primates only. The levels of antibodies in the mucosal secretions may be more significant that the serum levels for protection against mucosal infection. A limitation of the study was the small number of primates. These are promising data which offer a rationale for the development of controlled clinical trials.

Iron receptors

Alteri *et al.*, studying strains of uropathogenic *E. coli*, identified six proteins (ChuA, Hma, Iha, IreA, IroN and IutA) involved in iron uptake by bacterial cells. The purified proteins were further associated with an adjuvant and intranasally administered to a group of rats, with successive booster doses, 7 and 14 days after initial vaccination. The animals then underwent experimental UTI, to assess the vaccine-generated immune response. The authors concluded that vaccination with iron receptors can produce protective immunity against experimental UTI. Antibody production played an important role in the protection against infection, and correlated with a lower rate of bladder colonization. Although all antigens led to a significant increase of the serum levels of antigen-specific IgG and IgM antibodies, the animals immunized with Hma, IreA and IutA had a more striking increase of the IgG titers, compared with the IgM ones.[45]

Russo *et al.* undertook an experiment in rats, to test the role of the IroN receptor in the protection against UTI *in vivo*, through specific antibody-mediated immune response against this receptor. Compared to controls, the immunized animals had a significant increase of serum IgG antibody titers.[46]

There is evidence suggesting that this class of molecules can afford protection against *E. coli* infections. Further studies investigating their applicability to clinical practice and possible effects in humans are necessary.

Proteus mirabilis

Bacterial adhesion to the uroepithelium is a crucial step in the development of UTI caused by *Proteus mirabilis*. Different types of fimbriae can be found in the same strain of *P. mirabilis* (MR/P, UCA, PMF).

Li *et al.*, in an experiment with rats, assessed the efficacy of the administration of different vaccines composed of *P. mirabilis*, MR/P fimbriae or MrpH adhesin, a fragment of the MR/P fimbriae.[10] Intranasal immunization prevented UTI induced by inoculation of *P. mirabilis* into the urinary tract, and conferred a strong antibody response, with increased levels of serum, urinary, vaginal, biliary and bladder antibodies. Subcutaneous immunization induced a larger production of IgG antibodies, which did not necessarily result in more effective protection. The whole-organism vaccine is effective through the subcutaneous and intranasal routes, whereas the MR/P fimbriae-based vacine is effective through the intranasal and transurethral routes. Both vaccines effectively protected against *P. mirabilis*. Differently from what happened with the intranasal route, in the animals which received the whole *P. mirabilis* vaccine, the subcutaneous route failed to stimulate the production of urinary, bladder, vaginal and biliary antibodies. IgA production in the aforementioned sites was another characteristic of the animals which received the *P. mirabilis*-based and MrpH-based vaccines intranasally. Antibody production after the MR/P fimbriae-based vaccine was less vigorous compared with that produced after the whole-cell vaccine. Rats which were MR/P-immunized through the transurethral and intranasal routes had a significant reduction of urinary tract colonization with *P. mirabilis*.[10]

Scavone *et al.* investigated the immune response to the transurethral and intranasal administration of MrpA, UcaA and PmfA recombinant proteins, obtained from the MR/P, UCA and PMF fimbriae, respectively. Both administration routes stimulated the humoral response, with local and systemic antibody production, as well as the development of cell-mediated immune response. The intranasal route seems to more effectively stimulate urinary tract antibody production and protect against experimental UTI. Rats immunized through the intranasal route had a significant production of serum IgG and IgA, and the animals which received the PmfA and MrpA vaccines through the transurethral route had lower rates of renal colonization with *P. mirabilis*.[11] These same authors had already demonstrated that systemic immunization through the subcutaneous route with fimbriae subunits of *P. mirabilis* (MrpA, UcaA and PmfA) leads to a significant humoral response that can protect the immunized rats against ascending UTI caused by the same microorganism. The most promising results were obtained with administration of the MrpA antigen, a subunit of the MR/P fimbriae.[47]

Conclusion

There is evidence supporting the view that vaccines are a promising strategy for the prophylaxis of UTIs, as they have antigenic potential and can induce protective immunity. Further studies refining our knowledge of the cell-based and inflammatory mechanisms developed in animals and humans in response to these strategies should optimize our immunization approaches to UTIs. The clinical benefits of the vaccines for specific UTI groups, such as pregnant women, children and those with indwelling bladder catheters still need to be confirmed by controlled studies. The available results support the use of this therapy in young and post-menopausal women.

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