Prevention of recurrent bacterial cystitis by intravesical administration of hyaluronic acid: a pilot study

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OBJECTIVES

To assess the effect of bladder instillations of hyaluronic acid (HA) on the rate of recurrence of urinary tract infection (UTI).

PATIENTS AND METHODS

Forty women (mean age 35 years) with a history of recurrent UTI received intravesical instillations of HA (40 mg in 50 mL phosphate-buffered saline) once weekly for 4 weeks then once monthly for 4 months. The UTI status was assessed over a prospective follow-up of 12.4 months and compared with the rates of UTI before instillation, determined by a retrospective review of patient charts covering 15.8 months.

RESULTS

After HA treatment no patients had a UTI during the 5-month treatment phase and 28 (70%) were recurrence-free at the end of the follow-up. The mean (SD) rate of UTI per patient-year was 4.3 (1.55) before treatment and 0.3 (0.55) afterward ($P < 0.001$). The median time to recurrence after HA treatment was 498 days, compared with 96 days beforehand ($P < 0.001$). The tolerability was excellent, as side-effects were limited to nine patients who reported mild bladder irritation; no patient interrupted the treatment.

CONCLUSIONS

In this preliminary study, bladder instillations of HA had a significant effect on the rate of UTI in women with a history of recurrent UTIs. The bladder instillation of HA is an acceptable and promising therapeutic alternative in patients with recurrent UTI. Expanded placebo controlled clinical trials examining this application of HA are currently underway.

KEYWORDS

recurrent urinary tract infection, hyaluronic acid, bladder instillations, glycosaminoglycans

INTRODUCTION

UTIs are among the most common bacterial infections, affecting women at a much higher frequency than men [1,2]. Estimates suggest that about a third of women will have at least one episode of UTI requiring antibiotic therapy by the time they are 24 years old, and over a lifetime half will have at least one UTI [1,3]. There is also a high level of recurrence of UTI and 25–35% of initial UTI episodes will be followed by a recurrent infection within 3–6 months [2,4]. The high incidence of the disease leads to a large economic burden. The most recent estimate for the USA (1995) puts the direct cost of community-acquired UTI at $659 million and the indirect cost, through lost productivity, at $936 million [1,4]. This economic burden increases substantially when the costs of nosocomial UTI infections are included, which have been estimated to be $424–451 million per year [1,5].

Although UTIs have traditionally been managed by intermittent or prolonged antibiotic therapy [2,6], increasingly there is renewed interest in the mechanisms of UTI and the development into recurrent infections. Central to infection of the urinary tract is the interaction between the bacteria and the epithelial cells lining the bladder wall [7–9]. One component that is important in this event is the glycosaminoglycan (GAG) layer [10]; together with proteoglycans, GAGs form a glycocalyx that lines the transitional epithelium of the human bladder[10–12]. The presence of an intact GAG layer was proposed to be essential for protecting bladder epithelial cells from injury by toxic components of urine, and there is evidence that the layer blocks the adhesion of bacteria [13–16]. In contrast, a damaged GAG layer can result in direct exposure of epithelial cells to components of urine, and under these conditions there is an increase in both bacterial adherence and infection [16,17]. The GAG layer can be damaged by soluble virulence factors produced by *Escherichia coli*. Tay *et al.* [18], using a rabbit animal model, showed that when bladders were exposed *in vitro* to supernatants of *E. coli*-infected urine there was a significant increase in permeability, reflecting a disruption of the GAG layer, compared with bladder exposed to uninfected urine.

Damage to the GAG layer has been postulated as a causative factor in the development of interstitial cystitis, common UTIs and carcinoma of the bladder [10,19–21]. For interstitial cystitis there is evidence suggesting that reversing the damage, by repairing the GAG layer, results in a diminution of symptoms [17]. The GAG layer has been repaired with a range of epithelial coating techniques including heparin [22], oral pentosan polysulphate [23] or hyaluronic acid (HA) [13]. For the latter, Morales *et al.* [13, 24] showed that in 25 patients with interstitial cystitis, intravesical treatment with HA (40 mg diluted in normal saline to 50 mL, once weekly for 4 weeks, then monthly for 1 year if there was an initial response) resulted in a positive response in 71% of patients after 12 weeks of therapy.

The positive results obtained with intravesical HA therapy in interstitial cystitis suggested that a similar therapeutic approach might be beneficial for treating recurrent UTIs. In the present pilot study we assessed the effect of HA on the rate of recurrence of UTIs in patients with a history of recurrent UTI.

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TABLE 1 The status of UTIs and the pathogens identified before and after HA treatment in 40 women

Variable	Before	After	P*
Follow-up, months			
Mean (SD)	15.8 (7.61)	12.4 (3.94)	
Median (range)	13.9 (6.6–38.9)	11.4 (7.2–29.8)	0.0689
Number of infections			
Mean (SD)	5.2 (2.13)	0.4 (0.67)	<0.001
Rate of infection			
Mean (SD) N/year	4.3 (1.55)	0.3 (0.55)	<0.001
Pathogens, n (%)			
Total identified†	182	16	
E. coli	124 (68)	6	
Klebsiella spp	43 (23.6)	4	
Proteus mirabilis	12 (6.6)	5	
Pseudomonas aeruginosa	1 (0.5)	0	
Unknown	2 (1.1)	1	

**Wilcoxon rank-sum test; †Data on pathogens is expressed as the numbers of each pathogen identified and the percentage of the total number.*

PATIENTS AND METHODS

Women (≥18 years old) recruited for the study were referred to the outpatient clinic of the authors' institution specifically for the treatment of recurrent bacterial cystitis, and had been followed in the department for this problem for at least a year. Eligible patients had a documented history of recurrent cystitis, which for this study was defined as at least three episodes of uncomplicated cystitis caused by an identified pathogen in the last year. Only patients with a documented positive culture for each infection were included, a positive culture being defined as the isolation of a uropathogen at $\geq 10^3$ colony-forming units/mL [25].

A data-collection form to extract all UTI-related information from patient charts was designed specifically for the study. Data collection focused on the date of infection, type of bacteria involved and antibiotic sensitivity. The study was approved by the Ethics Committee of the hospital and all patients provided written consent before participating in the study.

All patients had a thorough clinical and radiological evaluation. The former was designed to exclude patients with urethral stenosis or external genitourinary abnormalities. The radiological examination included ultrasonography, IVU and retrograde cystography, and flexible cystoscopy with biopsy when required. The purpose of these examinations was to exclude patients with any congenital abnormalities, e.g. VUR, duplication of the ureter, ureterocele (intravesical or ectopic), ectopic ureteric orifice, urethral diverticulum or other predisposing factors, e.g. interstitial cystitis, stone disease and *in situ* carcinoma of the bladder. Also, patients in whom a neurogenic dysfunction of the lower urinary tract was suspected were evaluated urodynamically to confirm the condition, and these patients were also excluded. Additional patient exclusion criteria included any postvoid residual or the use of spermicides or intrauterine devices as regular birth control methods, as they have been implicated in the pathogenesis of recurrent bladder infections.

For patients meeting the inclusion criteria, a history of previous UTIs was documented by an interview and a retrospective review of their charts at the outpatient clinic. Information recorded included the frequency and timing of past UTIs, the pathogen involved in the infection and, if possible, the antibiotic regimen prescribed for each infection. Every effort was made to compile as complete a history of UTI as possible for each patient. All patients were free of infection at the initiation of the study.

In the prospective phase of the study, eligible patients received intravesical instillations of HA (Cystistat®, Bioniche Life Sciences Inc., Belleville, Ontario, Canada) at a dose of 40 mg in 50 mL PBS, once weekly for 4 weeks and then once monthly for 4 months. This treatment regimen was based on practice patterns developed in our department. The intravesical instillation was administered in the outpatient clinic using an 8 F Nelaton silicon catheter under sterile conditions, after removing residual urine. Local anaesthesia was used with the direct application of xylocaine gel 2% to the urethra 5 min before inserting the catheter. After the instillation the patients were asked to retain the HA solution in their bladder for ≥2 h and then advised to continue their normal everyday habits, including nutrition, smoking, sports and usual sexual activities.

Patients returned to the outpatient clinic for all instillations; on each occasion detailed information was obtained from the patient on adverse events resulting from the procedure. In addition, at each visit the UTI status was determined by urine culture of clean-catch MSU specimens taken before catheterization and HA instillation. UTI status was further assessed at follow-up visits at 1, 4 and 7 months after completing the required course of therapy. All patients were advised to consult our outpatient clinic if they had symptoms suggestive of a UTI, and those with evidence of a recurrent UTI at any time during the study were treated with antibiotics. When a patient developed a UTI, HA instillations were delayed until urine cultures were negative.

For the statistical analysis, the number of UTIs was calculated for the retrospective (before HA) and prospective (after HA treatment) phases of the study. Continuous variables were compared using the Wilcoxon rank-sum test. The time to recurrence of infection before and after HA therapy was analysed using a Kaplan-Meier survival function [26]; in the retrospective assessment (before HA) this was defined as the mean time elapsed between each infection, and in the prospective assessment (after HA) as the time elapsed between the first HA instillation and the first recurrent infection. The log-rank test was used to assess the comparability of the two survival curves. A general linear model was used to assess the influence of factors on time to recurrence.

RESULTS

In all, 40 women (mean age 35 years, range 18–45) were recruited for the study (Table 1).

All patients had been attending the clinic for at least a year and clinical records over this period were available for each. There were no menopausal women in the study and none of them had previous urinary tract surgery or radiotherapy (results not shown). However, as shown in Table 1, these patients had an extensive history of recurrent UTIs. Based on a retrospective assessment (mean 15.8 months) the 40 patients had 208 UTIs for a total observation period of 52.7 patient-years (5.2 infections/patient). On this basis the overall mean (SD) rate of infection, expressed as the number of infections per year and per patient, was 4.3 (1.55). As shown in Table 1, of 182 pathogens identified during the infections, *E. coli* and *Klebsiella* were the most common, representing 68% and 24% of the total pathogens, respectively. All the patients received standard antibiotic treatment and in the case of *E. coli*-derived infections the most common antibiotics used were ampicillin (30% of confirmed infections), cotrimoxazole (18%) and quinolone (15%). For this aspect of the study all information on past UTIs was available for all patients.

Over the course of the 5-month intravesical instillation with HA none of the patients had a UTI. In the extended follow-up (mean 12.4 months) only 12 patients (30%) had a UTI recurrence for a total observation period of 41.5 patient-years. As shown in Table 1, when the variables were compared before and after treatment for all patients there were statistically significant changes in the number of infections over the follow-up (mean decrease 4.8 infections) and in the rate of infections (mean decrease of 4 UTIs per patient/year). The follow-up after treatment (12.4 months) was not significantly different ($P = 0.069$) from that over which the retrospective assessment took place (15.8 months). Of the 12 patients who had recurrent UTIs in after treatment, eight had one UTI while the remaining four had two.

As shown in Table 1, for the 12 patients with recurrent UTIs after treatment the pathogens identified were similar to those before treatment. However, although *E. coli* was still the most common (six of 16), in five infections *Proteus mirabilis* was identified as the causative agent. This represented a 4.7-fold increase in the role of *P. mirabilis* in UTI after treatment, as before treatment this species was identified in only 6.6% of UTIs. The increase in involvement of *P. mirabilis* in recurrent UTI after HA treatment was at the expense of *E. coli* infections, which decreased from 68% before to six of 16 after treatment, whereas infections by *Klebsiella* were similar in both periods (23.6% and four of 16, respectively).

TABLE 2 The epidemiological characteristics of the two groups defined by their response to HA

Variable	Recurrence	No recurrence	P*
N	12	28	
Age, years			
Mean (SD)	36.4 (7.15)	34.4 (6.11)	0.2151
Median (range)	37.3 (19.9–44.5)	35.5 (18.8–45.3)	
Rate of infection before treatment, number/year			
Mean (SD)	4.8 (2.23)	4.1 (1.15)	0.6580
Median (range)	4.2 (2.4–8.9)	3.9 (2.1–6.9)	

**Wilcoxon rank-sum test.*

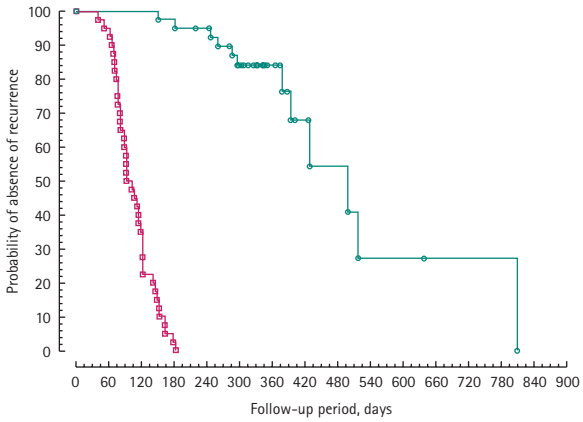

FIG. 1.
Time to recurrence of infection for the periods before (red solid squares) and after HA instillation (green open circles); the results are expressed as the probability of no recurrence of UTI with time. The median (range) time to recurrence was 96 (40–182) and 498 (150–810) days for before and after instillation, respectively.

To characterize any differences between patients with and with no recurrent UTI after HA treatment, the demographic and clinical variables for both groups were compared (Table 2). Patient age and annual rate of UTI for both groups before HA treatment were not significantly different.

The absence of recurrent UTI was compared before and after HA instillation by plotting Kaplan-Meier curves for each period (Fig. 1). Based on the follow-up for patients with or with no recurrence the median time to recurrence after the first instillation of HA predicted by the model was 498 days; the median time to recurrence before HA was 96 days, a significant difference ($P < 0.001$). In an analysis of factors before HA treatment that influenced time to recurrence afterward (variables analysed included age, number of infections before treatment, presence of at least one *E. coli*, *Klebsiella*, *Proteus* or *Pseudomonas* infection) only the number of infections had a significant correlation ($P = 0.0063$).

All patients tolerated the HA instillation and no serious adverse events were reported. Nine women (23%) reported mild bladder irritation but the symptoms did not last for >6 h after the instillation and only three of the women required anti-inflammatory medication to relieve the symptoms. None of the patients stopped HA therapy.

DISCUSSION

This study presents clear evidence that instilling HA into the bladder of women with a history of recurrent UTIs is feasible and well accepted by patients, and significantly reduces the incidence of recurrent lower UTIs. Over the 12.4 month follow-up from starting HA instillations the number and rate of infections decreased by 13- and 14.3-fold,

respectively (assessed over 15.8 months). Only 12 patients (30%) had a recurrence over the follow-up.

The phase of HA treatment lasted only 5 months, with weekly administrations for the first month followed by monthly treatments for 4 months. This regimen was based on pragmatic experience with HA therapy in patients with UTI at the authors' hospital, during which no patients had a recurrent UTI. However, the protective effect of HA was maintained even after direct treatment had stopped. Thus at a median time to progression of 498 days, the 5-month treatment phase maintained protection against UTI for 348 days, a very marked recurrence-free period.

Contemporary treatment options for women with a history of recurrent UTI usually include intermittent or prolonged antibiotic therapy, with variations in specific antibiotics, their dose and duration of therapy [1,27]. However, these approaches have had limited, and usually temporary, success, and persistence of infection and the emergence of resistant bacteria are common problems [2,6]. Alternatively, oestrogen replacement therapy has been suggested as a strategy to decrease the incidence of recurrent UTIs in postmenopausal women, by reversing the changes in vaginal pH, and this is an example of an intervention not based on antibiotics [28,29]. Raz and Stamm [28] found that in women treated with intravaginal oestriol cream there was a reduction in UTI recurrence and at 6 months ≈80% of the treated patients remained infection-free. However, no menopausal women were included in the present study and hence these patients would be ineligible for this antibiotic-free treatment.

The third therapeutic approach targets bacterial adherence to bladder mucosa. The most successful have used cranberry juice, effective through its phenolic components [30]. The principle of GAG substitution for preventing UTIs was shown experimentally in animals for heparin [15] and for sodium pentosan polysulphate [16]. Nevertheless, to our knowledge, the present study is the first to report the efficacy of GAG layer substitution for preventing recurrent bacterial cystitis in humans.

The mechanism of action of the protective effect of HA seen in the present study is not completely understood, but other successful treatment of interstitial cystitis with bladder instillation of HA [17] was assumed to result from repair of a damaged bladder GAG layer, and presumably a similar mechanism operates here. In the present case the damaged GAG layer facilitates bacterial adherence, leading to recurrent UTI, a process reversed by repairing the GAG layer with HA [15,17]. Based on this premise the preliminary data suggest that the more UTIs patients have the greater the damage to the GAG layer and the lower the probability that HA treatment can reverse that damage. For the present data, interestingly more *E. coli* have been shown to attach to vaginal epithelial cells from women with recurrent UTIs than to cells from normal women [9,31].

One of the potential limitations of the present study was the retrospective assessment of UTI history before HA therapy, as such data collection can be compromised by poor patient records. However, these patients had been receiving treatment in the clinical department at the hospital for a mean of 15.8 months and the UTI history was well documented for all patients. In addition, the single-arm study design and the relatively few patients may also be interpreted as limitations. Indeed, by including a retrospective and a comprehensive prospective component in the study, each patient served as her own control, strengthening the validity and interpretation of the data generated in this pilot study. The data were adequate to show a statistically significant change in the incidence of recurrent UTI from before to after treatment. As a result of these encouraging results a prospective, randomized, placebo-controlled study is currently underway.

In conclusion, bladder instillations of HA reduce the incidence of recurrent UTI, possibly through a protective effect on the GAG layer, and may offer an alternative to the widespread use of antibiotics, which are not always successful or well accepted by patients.

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CONFLICT OF INTEREST

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Abbreviations: **HA**, hyaluronic acid; **GAG**, glycosaminoglycan.