



Current Concepts Review

The role of vancomycin-soaking of the graft in anterior cruciate ligament reconstruction

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ABSTRACT

Approximately 1% of anterior cruciate ligament reconstruction (ACLR) procedures develop septic arthritis despite intravenous antibiotic prophylaxis and other preventive measures. Infection is most commonly due to contamination during autograft harvest and preparation by introducing bacteria into the knee during graft insertion. Pre-soaking ACL grafts in 5 mg/mL vancomycin (“vancomycin wrap”) has been utilised to eradicate such bacterial contamination. Many level III studies have reported a marked decrease in infection rates with no increase in graft failure rates. However, the lack of prospective randomised control trials and these studies’ heterogeneity do not allow a universal recommendation for vancomycin pre-soaking of all grafts during ACLR. Randomised controlled trials are needed to confirm efficacy in reducing sepsis rates.

Current concepts

- In 2021, it was reported that 38% of knee surgeons used a pre-implantation ‘wrap’ of the graft in a vancomycin-soaked swab
- Numerous Level III studies, systematic reviews and meta-analyses have demonstrated the effectiveness of the ‘vancomycin wrap’ in reducing infection rates compared to the use of pre-operative prophylactic antibiotics alone.
- Basic sciences and clinical research studies have failed to show any detrimental effect on graft biology, biomechanics, post-operative failure rates, return to sport, and overall knee function.
- Studies suggest that 5 mg/ml of vancomycin to soak the ACL graft are required to be effective at removing bacterial contamination. Studied ‘soak times’ have varied.
- The health economic case for its use seems compelling.

Introduction

Deep infection following anterior cruciate ligament reconstruction (ACLR) is around 1% (range 0.14–1.7%) [1–3] and 0.5% undergo re-operation for infection [4]. The most frequent organisms are coagulase-negative *Staphylococcus* followed by *Staphylococcus aureus* and

Propionibacterium acne [1]. The most common source of graft contamination is the patient’s skin flora [1]. Repeated washouts and long-term antibiotics allow graft survival in many cases [1]. However, it is necessary at times to remove the graft and hardware and perform revision ACLR surgery [1].

After sepsis, the main concerns are graft survival, long-term function, and osteoarthritis prevention [1]. Osteoarthritis risk is low [5–7]. However, the costs of prolonged treatment, work absence, disability, and anxiety are high [1].

Historically, most knee surgeons used pre-operative intravenous antibiotic prophylaxis. In 2012, Vertullo et al [3] reported outcomes after adding vancomycin to the graft (“vancomycin swab”) during ACLR performed by a single surgeon. It reported a 0% infection rate with vancomycin compared with 1.4% without it. Later studies have similarly reported a significant decrease in sepsis utilising this method compared to standard antibiotic prophylaxis [8,9].

Vancomycin has a broad activity spectrum against the most common pathogens associated with ACLR deep sepsis [10]. Similar vancomycin use in spinal and cardiac surgery did not increase antibiotic resistance [11,12]. If vancomycin usage reduces infections by 1%, over 1500 prophylactic post-operative antibiotic doses (offset by vancomycin costs) could be avoided for every 1000 ACLRs performed. This has economic and potential drug resistance benefits [1].

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Future perspectives

- New publications upon the subject are appearing frequently and, accordingly, the advice to surgeons needs constantly updating
- There is a need for Level I prospective randomised controlled correctly powered studies.
- Such research needs to answer the following questions
 - Optimal concentration and soaking time for topical antibiotic use
 - ◻ Does increased soaking time (and hence procedure time) paradoxically increase infection risk?
 - The necessity for universal use or should this be dependent upon such factors as:
 - ◻ Different types of graft
 - ◻ Primary or revision surgery
 - ◻ Individual patient risk factors

By 2016, Eriksson and Karlsson [1] stated that vancomycin was effective in decreasing ACLR sepsis rates from low to minimal. However, this claim was based on low-level evidence studies. A systematic PubMed search using the keywords “ACL AND vancomycin” identified 44 articles. Thirty-seven were deemed suitable for analysis. Six articles were not directly related to the subject leaving 31 for further investigation. Were previous claims justified and could any adverse effects be identified?

Molecular insights

A study utilising vancomycin (5 mg/mL) investigated whether human hamstring tendon (HT) autograft molecular function in ACLR was altered compared to controls [12]. Tenocyte viability *in vitro* after 60 min of treatment was diminished ($p < 0.05$) — but not maintained at 120 min. Vancomycin-treated tendon exhibited no significant augmentation in apoptotic gene expression, or apoptotic protein levels in tissue or supernatant, *ex vivo*. Vancomycin was related to a decrease in inflammatory proteins from treated tendon supernatants (IL-6; $p < 0.05$). It concluded that ACL “vancomycin wrap” did not alter ACL HT autograft molecular structure that might ameliorate graft integrity.

A 2020 study exposed human patellar tendon-derived tenocytes to varying and relatively high vancomycin concentrations. There were no significant changes in tenocyte viability after 2 and 6 h of incubation at any vancomycin concentration between 0 and 12,800 $\mu\text{g/mL}$. Incubation with vancomycin for 24 h led to a significant decrease in cell viability at higher concentrations [13].

Biomechanical and microbiological effects

Experimental studies

Schütter, Scharm, Stein et al. [14] examined porcine flexor tendons using “vancomycin wraps” with concentrations ranging from 1 to 10 mg/ml. Sodium chloride was utilised as a control [14]. After *Staphylococcus epidermidis* contamination, 63 tendons were wrapped into sterile compresses containing vancomycin for 10 and 20 min. Vancomycin soaking for 10 min failed to remove bacteria at any antibiotic concentration. At 20 min, contamination was encountered using 1 mg/ml and 2.5 mg/ml (85.7% and 42.9%, respectively). With 5 mg/ml and 10 mg/ml concentrations, contamination was eliminated after seven days of culture. No differences in peak load or Young's modulus were encountered between groups indicating no deterioration in biomechanical function.

Lamplot used bovine patellar tendons harvested and prepared as tendon-only grafts or bone-tendon-bone (BPTB) grafts. Vancomycin-tendon wraps (5 mg/mL) were rested for 30 min at room temperature and compared with normal saline controls. Vancomycin soaking appeared to have no immediate detrimental effects on either tendon-only or bone-

tendon-bone (BTB) grafts biomechanical properties using tensile testing machinery and assessing elongation strain and Young's modulus [15].

In a controlled *in vitro* biomechanical study utilising live donor graft, vancomycin pre-soaking human semitendinosus graft (STG) did not alter its biomechanical properties. 30 STGs harvested were dissected equally and randomly allocated to a vancomycin group or control group. The vancomycin group samples were presoaked in a solution of 5 mg/mL for 10 min. The control group samples were presoaked in a physiological serum for 10 min. Mechanical testing determined the stress-strain curve, Young's modulus, ultimate tensile stress, ultimate tensile elongation-before failure and elastic limit. No significant difference was observed between the 2 groups for all investigated parameters [16].

Vancomycin presoaking reduces the risk of infection after ACLR

Experimental study

Grayson found that soaked tendon grafts can act as vancomycin reservoirs. The antibiotic amount released and the elution profile depended upon rinsing, tendon volume, and antibiotic concentration. This study's clinical relevance was that vancomycin presoaking of ACLR autografts may reduce infection incidence without local or general toxicity risk [17].

Clinical studies—primary ACLR

Several publications have shown that pre-operative intravenous antibiotic prophylaxis combined with a “vancomycin wrap” reduces the post-operative ACLR deep infection rates [3,8,9,18–24] (Table 1).

Banios et al. [23] reported that knee infection risks were significantly higher with HT grafts (compared to BPTB grafts) and in revision cases. This was supported by a systematic review of level III and IV studies and meta-analysis (level IV evidence) [25]. Nonetheless, the study authors cautioned that it remained controversial whether vancomycin-soaking should be advised for all ACLRs or reserved for patients at risk. No differences were identified between patients (with or without vancomycin-soaked grafts) in the incidence of ACLR revision, IKDC (International Knee Documentation Committee) score, Tegner score, tendon biomechanics, and cartilage integrity. They reported an odds ratio of 0.04 in favour of post-operative infection reduction by adding intra-operative vancomycin-soaking of grafts.

A 2021 systematic review and meta-analysis (Level III evidence) found that soaking ACL tendon grafts with vancomycin pre-implantation were related to a nearly 15-fold decrease in the odds of infection compared to not soaking [26]. The review analysed 10 articles—nine used a concentration of 5 mg/ml and one used a concentration of 1 mg/ml. Soaking grafts in vancomycin resulted in fewer infections (0.013% vs. 0.77%; $p < 0.001$). No differences were found in IKDC scores one-year following ACLR for subjects with grafts pre-soaked in vancomycin vs. those without [25]. Few studies included investigated patient-reported outcomes and re-tear rates after vancomycin soaking causing difficulties concluding whether the practice affected re-rupture rates.

Optimal soaking time has not been definitively defined in the literature. Since operating time has been reported to independently correlate with postoperative infection [20], avoiding excessive soaking time is important. Similarly, any clinical study needs to consider co-morbidities. Factors such as BMI [20] and older patients [4] have been associated with greater complications for ACLR.

Clinical studies—revision ACLR

A large level II study reported that vancomycin soaking of the graft diminished postoperative septic arthritis in revision ACLR [27]. Analysing 2155 revision cases, vancomycin soaking (5 mg/ml) reduced infection rates from 0.9% to 0.0% ($p = 0.029$). HT was utilised in 60.8% and quadriceps tendons with patellar bone block tendons in 39.2% with no significant differences between groups.

Table 1

Clinical studies showing that vancomycin soaking reduces the risk of infection after primary anterior cruciate ligament reconstruction (ACLR).

AUTHORS	YEAR	TYPE OF STUDY	LEVEL OF EVIDENCE	N	CONCLUSIONS
Vertullo et al. [3]	2012	Case series	IV	1135	Prophylactic vancomycin pre-soaking of HT autografts diminished the infection incidence more than intravenous antibiotics alone.
Perez-Prieto et al. [8]	2016	Case-control	IV	810	Autograft pre-soaking with vancomycin in combination with intravenous antibiotic prophylaxis diminished the incidence of knee infection following ACLR more than antibiotic prophylaxis alone.
Phegan et al. [9]	2016	Controlled observational study	III	1585	Compared to intravenous antibiotics alone, pre-soaking of HT autografts with topical vancomycin diminished the incidence of postoperative infection.
Perez-Prieto et al. [18]	2018	NA	II	50	In 14% of cases, ACL graft harvesting and manipulation caused bacterial contamination. In this study, such contamination was fully eliminated after soaking in the vancomycin solution.
Figuerola et al. [19]	2019	Cohort study	III	490	Pre-soaking of HT autografts in vancomycin for ACLR prevented the appearance of postoperative septic arthritis compared to not soaking of the grafts.
Baron et al. [20]	2019	Therapeutic study	III	1640	The utilisation of vancomycin-soaked grafts was associated with a 10-fold decrease in infection after ACLR.
Wan et al. [21]	2020	Retrospective study	NA	305	Prophylactic vancomycin soaking of HT autograft was more efficacious in reducing the infection incidence than preoperative antibiotic alone.
Schuster et al. [22]	2020	NA	III	10,516	Soaking the graft in vancomycin solution before implantation diminished the incidence of postoperative septic arthritis after ACLR.
Banios et al. [23]	2021	NA	NA	1242	Septic arthritis after ACLR can be significantly diminished (or even eliminated) by soaking ACL autografts in a 5 mg/ml vancomycin solution.
Hees et al. [24]	2021	NA	III	1636	Graft soaking in vancomycin (1 mg/ml) was efficacious in preventing septic arthritis after ACLR.

N = Number of patients; HT = Hamstring tendons; NA = Non-available; ACL = Anterior cruciate ligament.

Avoidance of potential allergic reactions

Correct graft handling is important in avoiding allergic reactions to vancomycin. Direct contact between graft and staff should be minimised. Standard staff protective equipment should always be used. Pre-mixed bottles should be utilised and grafts soaked in antibiotic swabs within a plastic bag. This ensures staff exposure is kept to a minimum [1]. Observation of any adverse drug reaction from the patient should be standard practice.

Relationship of graft type to infection rates

HT autografts have been associated with higher ACLR infection rates than other graft types [1,23,28]. Accordingly, vancomycin-impregnated HT autografts were recommended [27]. These findings were supported by a 2020 systematic review and meta-analysis (level IV evidence). Reviewing 68,453 ACLRs, the overall infection rate was 0.9%. HT autografts had an infection frequency of 1.1%, BPTB autografts 0.7%, and allografts 0.5%. Pre-soaking HT autografts reduced infection rates to 0.1%. It was concluded that pre-soaking HT autografts diminished infection frequencies 10-fold [29].

Lack of toxicity

Data has not reported any adverse effects, such as graft degeneration, increased laxity, or a change in functional outcome at short-to-medium-term follow-up [16,30,31]. Experience from spinal surgery has not shown any negative consequences from vancomycin use [17,32].

Return to sport and knee function

Bohu et al. [33] compared return to sport (RTS) and knee function 1-year after ACLR using autografts with and without vancomycin pre-soaking. Ten minutes of graft soaking were used for the “soak” group. The study included 1674 patients. 0.59% without vancomycin had a deep infection compared to no infections with vancomycin. No significant

differences in return to running were found between the two groups one year after ACLR (75.9% vs. 76.1%, non-significant). Significantly more subjects in the vancomycin group returned to their pre-injury sport ($p = 0.04$). Knee function was comparable between the two groups. The article concluded that vancomycin-soaked grafts during ACLR diminished the risk of postoperative knee infection without affecting RTS or knee function.

Re-rupture and arthrofibrosis

Offerhaus et al. [34] investigated whether vancomycin pre-soaking (5 mg/ml) was associated with a higher frequency of complications—including graft failure or arthrofibrosis (level III evidence study). 1779 patients were included. Infection rates without vancomycin were 2.38% compared to 0% with antibiotics ($p < 0.01$). 500 patients were analysed further. These showed a significant reduction in graft failure: 3% in the vancomycin group, compared to 10% in the control group ($p < 0.05$). No differences were encountered in the frequency of postoperative arthrofibrosis, Tegner scores, or subjective results.

Perez-Prieto assessed the vancomycin soaking effect on re-rupture rates and functional outcomes [35]. Group 1 had pre-operative intravenous antibiotics alone. Group 2 had pre-operative intravenous antibiotics together and vancomycin soaking. The minimum follow-up was five years. 4.7% experienced re-rupture in group 1 and 3.9% in group 2 (non-significant). The IKDC was 82 in group 1 and 83.9 in group 2 ($p = 0.049$); the Tegner score was 4 in both groups (ns.), and the Lysholm score was 90.3 in group 1 and 92 in group 2 ($p = 0.015$).

These studies indicate that the vancomycin soaking for ACL autografts was a safe and cost-effective method for daily clinical practice—in terms of reduced infection, re-rupture rates, and functional outcomes.

Cost-efficacy

Ruelos et al. [36] performed an analysis (level IV evidence) of whether vancomycin graft soaking was cost-effective for infection prevention after arthroscopic ACLR. Infection treatment costs were

calculated using two alternative protocols: irrigation and debridement with revision ACLR or ACL graft retention. Intraoperative vancomycin was shown to be cost-effective if it prevented 1 infection in 550 cases (ARR = 0.182%), considering costs of \$24,178 and \$44/1000 mg for revision ACL reconstruction and vancomycin, respectively. If the ACL graft is retained after infection, intraoperative vancomycin was considered cost-effective if it prevented infection in 146 cases (ARR = 0.685%), given costs of \$6424 and \$44/1000 mg for arthroscopic debridement and vancomycin prophylaxis, respectively. Vancomycin soaking appeared a highly cost-effective prophylactic method for the prevention of infection after arthroscopic ACLR.

Survey of the ACL study group

An ACL study group 2021 survey assessed surgical practice patterns regarding pre-soaking ACL tendon grafts in vancomycin (level IV evidence study) [37]. A web-based questionnaire was used. Questions included vancomycin soaking use during ACLR, graft soaking protocols, vancomycin concentration, graft choices, and any technical concerns. Sixty-six (57%) completed the survey. 38% of vancomycin pre-soaked their ACL grafts pre-implantation—including 60% of European surgeons. 76% had adopted this method in the last 5 years. Most wrapped the graft in a vancomycin-soaked gauze before implantation (56%), utilise a concentration of 5 mg/mL (68%), and soak hamstring grafts (92%). No comments were made relating to differential practices between HT and BPTB grafts. Worries expressed were graft mechanical properties (35%), antibiotic costs (23%), availability (12%), and antibiotic resistance (9%).

Discussion

Vancomycin wrap: should it be the New gold standard?

A 2021 editorial by Pfeiffer, supported by 22 bibliographical references, reported that increasing numbers of surgeons using vancomycin graft soaking in ACLR had resulted in a significant reduction in post-operative septic knee arthritis [38]. *In vitro* findings show no detrimental effect of vancomycin on tendon properties [16]. Published concentrations of antibiotics (≥ 5 mg/ml) appear to be effective in eliminating graft contamination with no evidence of chondrotoxicity demonstrated [9,19,20,22,24].

According to Figueroa [39], the use of HT autografts has been related to an increased risk of infection following ACLR compared to other graft types. The reason for this is not clear, although contamination following harvesting and graft preparation is the most probable cause.

Vertullo [40] commented that septic arthritis is a severe complication of ACLR occurring in about 1% - despite appropriate intravenous antibiotic prophylaxis and other standard prophylactic measures, such as ventilation and patient and operating room sterility [1]. Infection is most likely to be caused by graft contamination during harvest and preparation. Pre-soaking ACL grafts in 5 mg/mL vancomycin were developed to eliminate bacterial contamination and supported by level III evidence from multiple observational trials reporting a significant decrease in infection frequencies without evidence of augmented graft failure or reduced functional outcomes [9,19,20,22,24]. The editorial concluded that the time for this method to become a global recommendation had arrived [40].

However, the technique is not universally accepted by knee surgeons [37]. The reason may be related to the lack of level 1 evidence. Observational studies utilising a historical cohort as a comparator are at risk of various biases. Level I evidence is needed for the technique to be accepted as a global recommendation in infection control guidelines. Future research on “vancomycin wrap” should focus on randomised controlled trials [40].

Despite the positive effects of vancomycin pre-soaking in reducing ACLR infection rate, the lack of prospective randomised control trials and

the heterogeneity of the different reported surgical groups makes it not possible to advise vancomycin pre-soaking of the graft globally for all ACLR subjects [38].

Conclusions

Vancomycin graft pre-soaking seems to be a reasonable technique to diminish the frequency of deep infection following ACLR. Nonetheless, standard prophylaxis (sterile operating room environment, appropriate patient preparation, including pre-washing with chlorhexidine, ventilation, and so on) remain key to preventing sepsis. Such prophylaxis together with vancomycin graft pre-soaking appears to further diminish the risk of infection and protect the graft from the subject's skin flora. However, the lack of prospective randomised controlled trials and the heterogeneity of the different reports published to date does not allow yet the unequivocal advice that all subjects undergoing ACLR should be subjected to vancomycin pre-soaking of their implanted graft.

Author contributions

ECR-M conceived the original article, wrote the first draft, and undertook the initial literature search. WJR reviewed the original article and relevant publications and undertook the drafting of the submitted manuscript. All authors attest to the accuracy and integrity of the manuscript.

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ECR-M and WJR do not have any competing interests or disclaimers.

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