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Are the leading drugs against *Staphylococcus aureus* really toxic to cartilage?☆

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Summary Many studies have shown that the toxic effects of local antibiotics on bone and cartilage limit orthopedic surgeons. In this study, we evaluated three antibacterial agents used locally to treat highly mortal and morbid diseases in the field of orthopedics, such as septic arthritis. Are vancomycin, teicoplanin, and linezolid, which are archenemies of *Staphylococcus aureus*, really toxic to chondrocytes? The purpose of the study was to investigate the effects of antibiotics, which are used against *S. aureus*, on human chondrocytes in vitro.

Primary cell cultures obtained from gonarthrosis patients were divided into two main groups. One of these groups was designated as the control chondrocyte culture. The other group was divided into three subgroups, and each group was exposed

☆ Level of Evidence: Level I, in vitro clinical research study.

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to vancomycin, teicoplanin, or linezolid. Cell culture samples were characterized by immunophenotyping following incubation with the three different antibiotics. Before and after the agents were administered, the cultures were subjected to inverted and environmental scanning electron microscopy. The number of live cells and the proliferation rate were monitored with the MTT-assay.
We found that vancomycin, teicoplanin, and linezolid do not have chondrotoxic effects.
Vancomycin, teicoplanin, and linezolid had no chondrotoxic activity during in vitro culture, which supports the argument that these agents can safely be used in orthopedic surgery, especially against methicillin-resistant *S. aureus* agents.

Introduction

Side effects and adverse reactions occur frequently during systemic antibiotic therapies [1,2]. The prolonged and high-dose administration of antibiotics required to treat septic arthritis and osteomyelitis results in side-effect profiles that also involve toxicity problems [1,3—5]. To avoid such deleterious effects, local antibiotics are often utilized, especially intra-operatively [6—11]. In orthopedics, placing antibiotic-impregnated Spongostan or gels/grafts in various forms into infected regions has been found useful to fight infection or to act as a prophylactic [2,3,7—9]. However, the toxicity of these local antibiotics has been discussed in the literature [1]. Additionally, reports indicating local osteotoxic and chondrotoxic effects have imposed some limitations on orthopedic surgeons [1,10], especially with regard to certain procedures involving open surgeries, such as intra-articular fracture surgery and multiple ligament knee injuries. In such cases, both prophylaxis and the use of antibiotic therapy may be helpful [12,13]. In the present study, vancomycin (VCM) [6], known to be chondrotoxic, and its alternatives teicoplanin (TEIC) and linezolid (LZD), commonly used against *Staphylococcus aureus*, the most common cause of septic arthritis, were specifically chosen due to their common use in orthopedics. The chondrotoxicity of these three antibiotics are investigated in human primary chondrocyte cultures.

Materials and methods

Prior to the present research, approval from the Namik Kemal University Medical Faculty Ethics Committee (Date: 25/12/2014; Number: 127) and the participant written informed consents were obtained. Primary chondrocyte cultures consisted of surgically harvested osteochondral tissues from gonarthrosis patients undergoing routine total knee arthroplasty, ($n = 6$); VCM, TEIC, and LZD agents were added, and cell viability, toxicity, and proliferation analyses were conducted at baseline (0 h), before the application of antibiotics, and after 24 h. Surface morphologies and the extracellular matrix were examined using inverted and environmental scanning electron microscopes (ESEM). To minimize measurement and analysis errors, the same researcher performed the analyses both during and after the primary chondrocyte culturing. The researchers conducting the analyses were blinded to the type of drug used and the experimental setup. All experiments were conducted three times.

Materials

Collagenase type II enzyme (1 mg/mL, Invitrogen Corporation); Hank's Balanced Salt Solution (HBSS-1X, 14025, Gibco); penicillin-streptomycin with fetal bovine serum (FBS); and Dulbecco's Modified Eagle's Medium (DMEM) (1000 mg glucose/L) were obtained from Sigma Chemical, St. Louis, USA. Sodium dodecyl sulfate (SDS) (10% L4522), insulin—transferrin—selenium (ITS) premix, and RPMI-1640 were obtained from Sigma—Aldrich GmbH, Germany. The MTT Kit was a Vybrant MTT Cell Proliferation Assay (Catalog ≠ V-13154) by Cell Biolabs Inc., USA. A laminar flow cabinet (Air Flow-NUVE/NF-800 R) and an incubator (NUVE, 06750) were obtained in Ankara, Turkey. An Olympus CKX41 inverted microscope was used and the images were recorded using an Olympus cell soft imaging system program. A Mindray MR 96A (PRC) enzyme-linked immunosorbent assay (ELISA) device measured viability, cytotoxicity, and proliferation with a commercial kit. This study used a Quanta 250 FEG (Fei Company, Hillsboro, Oregon, USA) ESEM.