

Pseudomonas Cepacia (PC) Contamination of a Cornea Conserved in Organ Culture

Pseudomonas-cepacia-Kontamination einer Hornhautbank-Hornhaut

Background

▼ In Switzerland three Eye Banks are operational for tissue banking. The Jules-Gonin Eye Hospital Eye Bank is one of the main of them and is able to provide each year about 120 corneal grafts for human therapy. The storage method used is organ culture at + 31 °C. In the medium, detection for bacterial contamination is available by change of color. The contamination rate of corneoscleral rims after penetrating keratoplasty is about 0.53 to 0.7% [1, 4].

To our knowledge, this is the first description of a corneal button contaminated by *Pseudomonas Cepacia* and grafted on a recipient for human therapy. We describe the clinical features, the method used to identify the infection, the treatment and the procedure that was adapted to prevent a recurrence.

History and signs

Corneal processing (Donor)

Corneas were collected from cadaver donors in the Centre Hospitalier Universitaire Vaudois (CHUV). The eyeballs were removed after 5% povidone-iodine decontamination for 3 minutes, and the corneas with the scleral ring were excised in the eye bank under a vertical laminar flow in the clean room. After its excision, the corneoscleral rims were immediately immersed in 100 ml of conservation medium (CM) (CorneaMax, EUROBIO laboratories, France). In our case, no special features were noticed at the initial exam and when the corneas were prepared. After a quarantine of 7 days an endothelial cell count was performed without trypan blue in the clean room. During the banking the color and the turbidity of the CM were analyzed each 3 days. Two days before transplantation, a last quality check was performed including endothelial cell examination, with a color and a turbidity check of the CM. Furthermore, a direct microbiological control was performed on 5 ml of CM in an independent laboratory (Bactolab SA in Lausanne). Only then, the corneal button was ready to be used for human therapy and was grafted.

med on 5 ml of CM in an independent laboratory (Bactolab SA in Lausanne). Only then, the corneal button was ready to be used for human therapy and was grafted.

Clinical features of the recipient

The recipient was a 59-year-old diabetic woman. On past ophthalmologic history, she had a corneal graft on her right eye for keratoconus in 1982, and 6 years later, she underwent a refractive radial and arcuate keratotomy on the same eye for residual ametropia. In 2006, she underwent a second penetrating keratoplasty on her right eye for corneal scar, complicating the refractive surgery performed in 1988.

The day after her second penetrating keratoplasty, clinical examination revealed a visual acuity limited to Hand Motion (► Fig. 1). At slit lamp control, we noticed diffuse stromal corneal opacities in the graft with a recipient corneal ring remaining transparent. A strong inflammation was present in the anterior chamber: cells 3+ and Tyndall 2+.

Method used to identify the infection

Microbiologic controls were performed with smears on the cornea, in the fornix, in the anterior chamber and in the CM.

The diagnosis of a primary corneal infection was confirmed regarding to the association of the clinical features with PC positive smears from CM and ocular surface of the recipient. Moreover, the CM culture of the donor's fellow eye was also positive for PC. The antibiogram results obtained 3 days after the surgery showed sensitivity to quinolons and chloramphenicol and resistance to the other current antibiotics including vancomycin and aminosids.

Therapy and outcome

▼ In emergency, a third penetrating keratoplasty was performed once the diag-

nosis confirmed (► Fig. 2). It was associated to an intraocular injection of both aminosids and vancomycin. Then we introduced intensive topical and parenteral antibioticotherapy adapted to the antibiogram. The evolution was favorable and leaded to a fast visual recovery (0.2 decimal equivalents at 1 month and 0.4 at 3 months) with no relapse of the infection during the follow up (► Fig. 3).

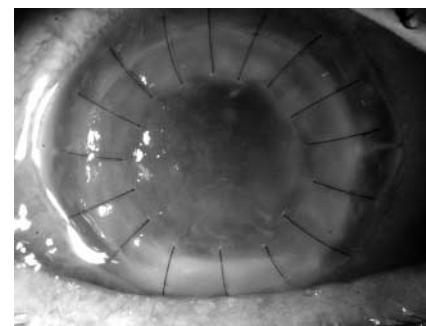


Fig. 1 Initial presentation of the patient the day after the second corneal graft: visual acuity limited to hand motion, corneal opacification, whiteness and dense infiltration of the graft with a recipient corneal rim remaining transparent.

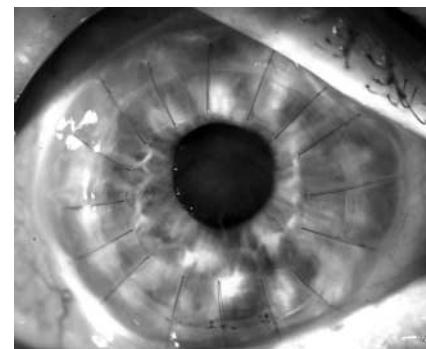


Fig. 2 One month after the third graft. Visual acuity was 0.2 (decimal).

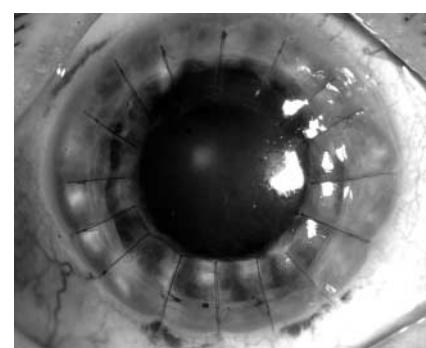


Fig. 3 Three months after the third graft, the visual acuity was 0.4 (decimal) with no relapse of the infection.

Discussion



Biosafety of organ or tissue used for transplantation

Up to the new laws for organ and tissue transplantation adopted by the Swiss Confederation in July 2008, no specific guidelines were listed for the banking. We usually adopt the European Laws as a guideline for our practice. With the new set of laws, the processes for organs or tissues transplantations have been defined more precisely and good manufacturing practices (GMP) are controlled by Swiss Medic. These GMP take in consideration, organization of the Eye Bank, quality control of the environment, traceability of the reagents, methodology in the corneal processing and report of the incidents (Swiss Federal Laws for tissues, cells and organs transplants).

Contamination of a tissue during the banking is an important concern. In a series of 2608 corneas managed in the Besançon Eye Bank, 5.3% of the corneas were positive for bacterial contamination [8]. To detect an infection during the banking, the most frequent protocol in European Eye Bank is a rapid microbiologic testing, of a CM sample, by an independent laboratory two days before surgery. However, in the United States, conservation at + 4 °C is the standard method for corneal banking and no bacterial evaluation is performed before grafting.

Procedure that was adapted to prevent a recurrence

The case we report was unique in our practice, that the reason why the bank activity was not stopped then. In fact, we rapidly postulated that PC was probably carried by the donor and was transmitted to the recipient after the organ culture period.

After this incident, we adapted our procedure and decided to advance the last check quality point to 5 days before grafting. With this procedure, we will not have only a quick microbiological testing on the CM, but also a culture on a longer period of time able to identify suspect bacterial growth.

Pseudomonas Cepacia

PC is a virulent pathogen responsible for nosocomial infections in hosts with altered immunity [5, 9]. It has been implicated in endophthalmitis, keratitis and conjunctivitis and is resistant to conventional antipseudomonal therapy [3, 6].

Morel et al. reported a series of corneal contamination with trypan blue used for endothelial cell count during banking. 169 corneoscleral rims analyzed with a contaminated trypan blue were grafted. Fortunately, no cases of infection were reported during the 3 months of follow-up [8].

In the case we report, PC could survive in CM because it was resistant to the antibiotics present in the CorneaMAX (penicillin and streptomycin). Furthermore, the antibiotics we used in emergency were not efficient because of PC resistances.

PC is innately resistant to a wide range of antibiotics including polymyxin, aminoglycosides and the antipseudomonal penicillins [10]. Historically the most effective antibiotics were cotrimoxazole and chloramphenicol [10]. However, these agents showed limited clinical efficacy. Ceftazidime, temocillin, imipenem and ciprofloxacin display some in vitro activity against the bacterium [10]. PC is a pathogen that divides only poorly and very slowly [7]. It is probably the reason why we could not detect it before grafting.

Tissue banking contamination is a serious complication. The critical contamination periods are before removal of the eye, during mortuary washing and during decontamination of the fornices [2]. A special care must be undertaken during those critical stages of the procedure.

Pseudomonas cepacia is resistant to conventional antipseudomonal therapy. Changing the graft in emergency and adapt the treatment according to the antibiogram was probably the only issue to solve this tricky situation. This case led us to reconsider the procedure we use to control possible contamination during the banking.

Conflict of interest: None

M. Berguiga¹, S. Uffer¹, F. Hafezi³,

F. Majo^{1,2}

¹ Hôpital Ophtalmique Jules-Gonin, Faculté de Médecine de Lausanne, Switzerland (Chairman: Professor Zografos)

² Service de Chirurgie Expérimentale, Centre Hospitalier Universitaire Vaudois, Lausanne, Switzerland (Chairman: Professor Barrandon)

³ IROC, Institut für Refraktive und Ophtalmochirurgie, Zürich, Switzerland

References

- 1 Borderie VM, Laroche L. Microbiologic study of organ-cultured donor corneas. *Transplantation* 1998; 66: 120–123
- 2 Builles N, Perraud M, Reverdy ME et al. Reducing contamination when removing and storing corneas: A multidisciplinary, transversal, and environmental approach. *Cornea* 2006; 25 (2): 185–192
- 3 Del Piero E, Pennet M, Leopold I. *Pseudomonas cepacia* endophthalmitis. *Ann Ophthalmol* 1985; 17: 753–756
- 4 Hagenah M, Bohnke M, Engelmann K et al. Incidence of bacterial and fungal contamination of donor corneas preserved by organ culture. *Cornea* 1995; 14: 423–426
- 5 Kaitwacharachai C, Silpapojakul K, Jitsurong S et al. An outbreak of *Burkholderia cepacia* bacteremia in hemodialysis patients: an epidemiologic and molecular study. *Am J Kidney Dis* 2000; 36: 199–204
- 6 Lennette EH, Balows A, Hausler WJ. Aerobic bacteria: In manual of clinical microbiology American society for microbiology. Washington, D.C, 1985; 4th ed: 362
- 7 Levy JH, Katz HR. *Pseudomonas cepacia* keratitis. *Cornea* 1989; 8 (1): 67–71
- 8 Morel P, Roubi N, Talon DR et al. Bacterial contamination of a Cornea Tissue bank: implications for the Safety of the Graft Engineering. *Cornea* 2003; 22 (3): 221–225
- 9 Snell GI, De Hoyos A, Krajden M et al. *Pseudomonas cepacia* in lung transplant recipients with cystic fibrosis. *Chest* 1993; 103: 466–471
- 10 Spencer RC. The emergence of epidemic, multiple-antibiotic-resistant *Stenotrophomonas maltophilia* and *Burkholderia cepacia*. *J Hosp Infect* 1995; 30 (Suppl): 453–464

Bibliografie

DOI 10.1055/s-0028-1109284

Klin Monatsbl Augenheilkd 2009; 226: 348–349
© Georg Thieme Verlag KG Stuttgart · New York
ISSN 0939-2661

Korrespondenzadresse

François Majo, MD PhD

Head of: Medical and Surgical Cornea, Refractive Surgery and Eye banking, Hôpital Ophtalmique Jules-Gonin
Avenue de France 15
1004 Lausanne
Switzerland
fmajo@bluewin.ch