Failure of Acanthamoeba castellani to Produce Intraocular Infections

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PURPOSE. This study examined possible mechanisms to explain why Acanthamoeba castellani remains restricted to the cornea and rarely produces intraocular infections. The first hypothesis proposed that trophozoites cannot penetrate Descemet’s membrane and the corneal endothelium to enter the anterior chamber (AC). The second hypothesis proposed that the trophozoites can enter the AC; however, the aqueous humor (AH) contains factors that either induce encystment or kill the amoebae.

METHODS. Descemet’s membrane was isolated from pig corneas and was used to determine whether Acanthamoeba trophozoites could penetrate this membrane in vitro. In addition, the capacity of trophozoites to survive in AH was determined in vitro. Trophozoites (10^6) were injected into the AC of hamster eyes, and the number of amoebae in the AC was determined by histopathology 1 to 15 days later.

RESULTS. The amoebae penetrated Descemet’s membrane within 24 hours of in vitro culture. Penetration was prevented by addition of serine protease inhibitors or a chicken monoclonal antibody against the Acanthamoeba serine protease MIP-133. Although AH induced encystment of the amoebae, cysts remained viable. Injection of amoebae into the AC induced a robust neutrophil infiltrate, which was associated with complete clearance by day 15 after AC injection.

CONCLUSIONS. The findings suggest that A. castellani is capable of penetrating Descemet’s membrane and entering the AC. However, a robust neutrophil response is associated with the disappearance of intraocular trophozoites and suggests that cells of the innate immune apparatus are important in preventing Acanthamoeba keratitis from progressing to become an intraocular infection. (Invest Ophthalmol Vis Sci. 2005;46: 2472–2478) DOI:10.1167/iovs.05-0140

Acanthamoeba spp. are ubiquitous free-living protozoa that have been isolated from a variety of habitats, ranging from soil to eyewash stations. The life cycle of Acanthamoeba spp. consists of two stages: the motile trophozoite (10–25 μm) and the dormant cyst (8–12 μm). Acanthamoeba spp. most commonly infects the eye to produce Acanthamoeba keratitis, a progressive, sight-threatening corneal infection. Characteristic symptoms include epithelial ulcers, a ring-like neutrophilic stromal infiltrate, disproportionately severe ocular pain, and resistance to many antimicrobial drugs. Typical treatment consists of topical applications of propamidine isethionate, polyhexamethylene biguanide, or chlorhexidine, alone or in combination. Even with such therapies, many patients receive therapeutic corneal transplants, which can be reinfected by the activation of dormant cysts in the graft bed.

The pathogenic cascade of Acanthamoeba keratitis begins when the trophozoites bind to and desquamate the corneal epithelium. Acanthamoeba trophozoites adhere to mannose glycoproteins on the corneal epithelium via the amoeba’s mannos-binding receptor. Exposure of Acanthamoeba trophozoites to mannose has been shown to induce the production of a 133-kDa protein, termed mannose-induced protein (MIP-133), which is highly cytolytic to corneal epithelial cells in vitro. Acanthamoeba trophozoites penetrate the corneal epithelium and underlying Bowman’s membrane. In addition to killing corneal epithelial cells, Acanthamoeba trophozoites secrete a variety of proteases, including MIP-133, that facilitate invasion of the corneal basement membrane and dissolution of the corneal stroma.

Unlike bacterial keratitis, Acanthamoeba keratitis rarely progresses beyond the cornea to produce intraocular infections and endophthalmitis. Only a handful of cases of Acanthamoeba endophthalmitis have been reported in which Acanthamoeba has been shown to be the causative agent of intraocular infection. Furthermore, it is questionable whether these cases of endophthalmitis were due to the natural progression of the trophozoite beyond the corneal endothelium. This study considered the hypothesis that A. castellani, unlike other ocular pathogens, does not progress and produce intraocular infections. One hypothesis proposed that trophozoites cannot penetrate Descemet’s membrane and the corneal endothelium to enter the anterior chamber (AC). The second hypothesis proposed that the amoebae can enter the AC; however, the aqueous humor (AH) contains factors that either induce encystment or kill the amoebae.

MATERIALS AND METHODS

Acanthamoeba castellani (ATCC 30868), originally isolated from a human cornea, was obtained from the American Type Culture Collection (Manassas, VA). Trophozoites were grown as axenic cultures in peptone-yeast-glucose (PYG) medium at 35°C, as described previously.

Human corneal endothelial (HCE) cells were isolated from a corneal explant and immortalized with the human papillomavirus E6 and E7 genes, as described previously. HCE cells were cultured in complete Eagle’s minimum essential medium (MEM, BioWhittaker, Walkersville, MD) containing 1% l-glutamine (BioWhittaker), 1% penicillin, streptomycin, amphotericin B (Fungizone; BioWhittaker), 1% sodium pyruvate (BioWhittaker), and 10% fetal calf serum (FCS, HyClone Laboratories, Logan, UT).

Primary human iris-ciliary body (ICB) cells were harvested from a corneal scleral rim and cultured in Dulbecco’s modified Eagle’s me-
dium (DMEM; BioWhittaker) containing 1% t-glutamine, 1% penicillin, streptomycin, amphotericin B, 1% sodium pyruvate, and 10% human serum type AB (Sigma-Aldrich, St. Louis, MO).

**Animals**

Chinese hamsters were purchased from Cytogen Research and Development, Inc. (West Roxbury, MA) and used at 4 to 6 weeks of age. All corneas were examined before experimentation, to exclude animals with preexisting corneal defects. Animals were handled in accordance with the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research.

**Chicken Anti-MIP-133 Antiserum**

The specificity of the chicken anti-MIP-133 antiserum (Aveslabs, Tigard, OR) was confirmed through Western blot analysis and ELISA, as described previously. Anti-MIP-133 antiserum inhibits cytopathic effects (CPEs) against corneal epithelial cells in vitro and effectively blocks migration through a collagenous matrix (Matrigel; Collaborative Biomedical Products, Bedford, MA).

**Assay for CPE**

The CPE on corneal cells was determined with a previously described photometric assay. Briefly, trophozoites (2.5 × 10^4) were added to 96-well plates, with or without confluent monolayers of HCN or ICB cells and incubated for 48 hours at 37°C. Wells were Giemsa stained (Shandon, Inc., Pittsburgh, PA) and the optical density was read at 590 nm in a microplate reader (Molecular Devices, Menlo Park, CA). The OD of trophozoites alone was subtracted from the OD of experimental wells to determine the OD of live cells.

**Migration Assays**

Corneas were removed from porcine eyeballs (Owen’s, Dallas, TX) and placed in 5% dispase (Invitrogen-Gibco Corp., Carlsbad, CA) in PBS (pH 7.2) and incubated at 37°C for 30 minutes. Descemet’s membrane was peeled from porcine corneas and placed on a nitrocellulose membrane (15 mm diameter, 3.0 μm pore size; Millipore Corp., Bedford, MA) in a Boyden chamber (Costar Corp., Cambridge, MA). Acanthamoeba trophozoites (10^6) were placed in the top chamber in 200 μL PYG and incubated at 37°C for 24 hours. Acanthamoebae were counted in the bottom chamber with a hemocytometer. Viability was ascertained by trypan blue exclusion. Inhibition assays involved incubating trophozoites with the serine protease inhibitors, 1 mM PMSF and 1 mM 1,10-phenanthroline (Shandon, Piscataway, NJ); or 50 μg/mL cytchalasin D at 37°C for 30 minutes before their addition to the upper chambers.

To ensure that the protease inhibitors were nontoxic to trophozoites, 10^6 trophozoites were incubated in 1 mL of PYG with either one mM PMSF and one mM 1,10-P, a 1:10 dilution of chicken anti-MIP-133 antiserum (0.2 mg/mL), or a 1:100 dilution of chicken preimmune serum control (0.2 mg/mL) at 37°C for 24 hours. The number of viable trophozoites was determined by trypan blue exclusion and direct trophozoite counts on a hemocytometer.

To determine whether the protease inhibitors would impede trophozoite motility, trophozoites (10^6) were incubated in 1 mL of PYG with one mM PMSF and one mM 1,10-P; a 1:10 dilution of chicken anti-MIP-133 antiserum (0.2 mg/mL); a 1:100 dilution of chicken preimmune serum control (0.2 mg/mL); or 50 μg/mL cytchalasin D at 37°C for 30 minutes before their addition to the upper level of a migration chamber (Transwell; 6.5 mm diameter, 3.0 μm pore size; Corning Costar, Corning, NY). The trophozoites were then placed in the top chamber in 200 μL PYG and incubated at 37°C for 24 hours.

Trophozoites were counted in the bottom chamber by light microscopy (magnification, ×100).

**Effect of Aqueous Humor on Trophozoites**

Trophozoites (10^6) were incubated in PYG, PBS, or porcine AH, and viability of organisms was determined through trypan blue exclusion at 24 hours, 48 hours, or 7 days.

AH was concentrated 10× using microcentrifugal concentrators with membranes having a molecular mass cutoff of approximately 5 kDa (Millipore Corp., Bedford, MA). Trophozoites (10^6) were incubated in PYG, AH, 10× AH, or PBS for 7 days. Serial dilutions of the trophozoites, ranging from 10^3 to 10^7, were made and placed on a lawn of Escherichia coli on nutrient agar as described previously. Subsequently, the plates were observed for the presence of Acanthamoeba-produced trails at 24, 48, and 72 hours.

Trophozoites (2.5 × 10^7/well) were incubated in a 24-well plate in PYG or AH alone or in AH on a confluent monolayer of ICB cells. Because trophozoites kill the monolayer of ICB cells within 48 hours, it was necessary to transfer trophozoites to new wells containing fresh PYG, fresh AH, or fresh AH on a new monolayer of ICB cells at 24, 48, and 72 hours. The contents of each well were suspended in the respective medium, and the viability of cysts and trophozoites was determined at 24, 48, 72, and 96 hours by trypan blue exclusion. Trophozoites and cysts were differentiated from ICB cells by morphology.

**AC Injections**

Trophozoites were injected into the AC of the Chinese hamster eye by using a previously described method for transplanting a precise number of tumor cells into the AC of the mouse eye. Briefly, hamsters were deeply anesthetized with ketamine (100 mg/kg of body weight; Fort Dodge Laboratories, Fort Dodge, IA) given intraperitoneally. Trophozoites (10^6/5 μL of PYG) were inoculated into the AC using a 0.1 mL syringe (Hamilton Co., Reno, NV) fitted with a 35-gauge glass needle. Control experiments involved injecting PYG alone (5 μL) into the AC of the eye. Eyes were examined three times per week for 3 weeks for the appearance of clinical disease.

**Histologic Examination**

Eyes were removed at 30 minutes and on days 1, 4, and 15 after inoculation of trophozoites into the AC and stored in 10% Carson’s formalin for 24 hours. Specimens were then embedded in paraffin, cut into 4-μm sections with a rotary microtome (Shandon; Thermo Shandon, Pittsburgh, PA) and placed on positively charged glass slides (VWR Scientific, Suwanee, GA). Slides were then bleached overnight in 3% hydrogen peroxide in 1% sodium phosphate-buffered saline. Sections were stained with hematoxylin and eosin, coverslipped, and examined by light microscopy.

**Statistical Analysis**

Student’s unpaired t-test was used to determine statistical significance between experimental and control groups.

**Results**

Trophozoite-Mediated CPE on HCN Cells In Vitro

Previous studies have shown that trophozoites are capable of killing a variety of target cells in vitro. To ascertain whether A. castellanii rarely progresses beyond the corneal endothelium to produce intraocular infections, it was important first to determine whether trophozoites could either disrupt or kill HCN cells in vitro. A. castellanii trophozoites produced extensive CPEs on HCN cells in vitro, killing approximately 90% of cells within 48 hours, compared with untreated control cells (data not shown).
Trophozoite Penetration of Descemet's Membrane and the Corneal Endothelium

It is possible that Descemet’s membrane and the corneal endothelium serve as a physical barrier to *Acanthamoeba* trophozoites, preventing entry of trophozoites into the AC and subsequent intraocular infection. Accordingly, the ability of *Acanthamoeba* trophozoites to penetrate Descemet’s membrane was examined in vitro. More than one-third of the trophozoites were capable of penetrating Descemet’s membrane within 24 hours of incubation (Fig. 1A). Serine protease inhibitors, PMSF, and 1,10-P inhibited the penetration of Descemet’s membrane by approximately 75%, whereas the cysteine protease inhibitor cystatin had no significant effect. Previous studies have shown that *A. castellanii* trophozoites elaborate a 133-kDa serine protease, MIP-133, that is cytolytic against corneal epithelial cells and de-grades collagen in vitro. Therefore, the ability of anti-MIP-133 antiserum to prevent the penetration of Descemet’s membrane was examined. Anti-MIP-133 antiserum treatment decreased the penetration of Descemet’s membrane by approximately 50%, whereas the preimmune normal serum (NS) control had no significant effect (Fig. 1B).

The inhibition of trophozoites by protease inhibitors was not due to adverse effects on trophozoite viability or motility (Fig. 2). However, cytochalasin D inhibited trophozoite migration through a migration membrane (Transwell; Corning).

**FIGURE 1.** Effect of protease inhibitors and anti-MIP-133 antiserum on trophozoite penetration of Descemet’s membrane in vitro. *Acanthamoeba* trophozoites (10^6/200 μL PYG) were placed on Descemet’s membrane in the top chamber of a Boyden chamber and incubated at 37°C for 24 hours. (A) Inhibition assays involved incubating trophozoites with serine protease inhibitors (1 mM PMSF/1 mM; 1,10 P) or a cysteine protease inhibitor control (10 μM cystatin) at 37°C for 30 minutes before their addition to the upper chamber. (B) Inhibition assays involved incubating trophozoites with chicken anti-MIP-133 antiserum (0.2 mg/mL; ImS) or chicken preimmune normal serum control (0.2 mg/mL; NS) at 37°C for 30 minutes before their addition to the upper chamber. *P < 0.05.

**FIGURE 2.** Effects of protease inhibitors on trophozoite viability and motility. (A) Effect of serine protease inhibitors and a cytoskeleton inhibitor on trophozoite viability. *Acanthamoeba* trophozoites (10^6) were incubated in 1 mL of PYG, (1) alone or with (2) cytochalasin D (50 μg/mL), (3) serine protease inhibitors, (4) anti-MIP-133 antiserum (0.2 mg/mL), or (5) preimmune normal serum control (0.2 mg/mL) at 37°C for 24 hours. The number of viable trophozoites was determined by trypan blue exclusion. Data are the mean ± SE of triplicate counts. The results are not significantly different from the PYG control. (B) Effect of protease inhibitors on trophozoite migration through a migration chamber. *Acanthamoeba* trophozoites (10^6/200 μL PYG) were incubated (1) alone or with (2) cytochalasin D control, (3) serine protease inhibitors, (4) cysteine protease inhibitor, (5) anti-MIP-133 antiserum, or (6) preimmune normal serum at 37°C for 30 minutes before their addition to the upper chamber of a two-chamber migration system. A 3-μm pore size membrane separated the upper and bottom chambers. After a 24-hour incubation at 37°C, *Acanthamoebae* were counted in the bottom chamber. Data are the mean ± SE of triplicate counts. *P < 0.05.*
Costar) by 70%, without evidence of toxicity to trophozoites (Fig 2A).

**Effect of Aqueous Humor on Trophozoite Viability**

AH contains several factors that are known to be toxic to microorganisms, including complement and immunoglobulin. Approximately 37% of trophozoites encysted in AH after 48 hours, whereas only 10% encysted in PYG during the same time frame (Fig. 3). After 7 days, 100% of trophozoites encysted in AH, PBS, and PYG.

Ten-fold dilutions of cysts were made ranging from $10^5$ to $10^7$ cysts after the 7-day incubation in AH, PBS, or PYG. Each dilution of cysts was plated on a lawn of E. coli on nonnutrient agar to determine cyst viability. By 72 hours, only the dilution containing 10 cysts in the AH group did not produce trails on the lawn of E. coli, indicating that at least 99.999% of the cysts were viable after the 7-day incubation in AH (data not shown). These results were similar to those in the PBS control group.

Trophozoites were incubated in $10^2$ trophozoites. The number of trophozoites in the AC decreased by 70%, without evidence of toxicity to trophozoites in AH, PBS, and PYG.

FIGURE 3. Trophozoite encystment in AH in vitro. Trophozoites ($10^6$) were incubated in 1 mL of either PYG (negative control), aqueous humor (AH), or phosphate buffered saline (PBS; positive control for encystment). Trophozoites and cysts were counted at 24 and 48 hours and 7 days, and viability was determined by trypan blue exclusion. *P < 0.01.

**Trophozoites in the AC**

It is possible that trophozoites can enter the AC; however, the AH that fills this compartment of the eye contains factors that either induce encystment or kill the amoebae in vivo. To determine whether trophozoites can survive in the AH in the AC, $10^6$ trophozoites were injected into the AC of Chinese hamsters on day 0. Eyes were enucleated and examined after 30 minutes, 1 day, 4 days, and 15 days. (A) Normal Chinese hamster AC and iris (I). (B) AC containing trophozoites within 30 minutes after AC injection. (C) Trophozoites (arrows) localized on the iris surface (I) within 30 minutes after AC injection. (D) Trophozoites and inflammatory cells in the AC within 1 day after AC injection. (E) Neutrophils (arrows) circumscibing trophozoites (T) within 1 day after AC injection. Note the trophozoite (T) nestled in the iris (I). (F) Abundance of inflammatory cells in the AC 4 days after AC injection. (G) Paucity of trophozoites in the AC within 4 days after AC injection. (H) Absence of both trophozoites and inflammatory cells in the AC 15 days after AC injection. (I) Normal iris architecture 15 days after AC injection. Note the absence of inflammatory cells. Control experiments involving the injection of PYG alone (5 μL) into the AC of the eye did not result in inflammation (data not shown). Magnification: (A, B, D, F, H) ×100; (C, E, G, I) ×600.

by day 4 after inoculation. By day 15, both trophozoites and inflammatory cells were not detected anywhere in the eye.

**Effect of AH on Encystment of Trophozoites Incubated with ICB Cells**

Trophozoites rapidly encysted when incubated in AH in vitro, yet there was no evidence of encystment of trophozoites after injection into the AC of Chinese hamsters. We believe the encystment induced by AH in vitro is the consequence of nutrient deprivation. The extensive accumulation of trophozoites on the iris and the conspicuous absence of trophozoite encystment suggest that ICB cells may serve as a source of nutrition for trophozoites that enter the AC. Accordingly, the capacity of trophozoites to kill ICB cells was examined in vitro. Within 48 hours of in vitro incubation, trophozoites lysed approximately 80% of the ICB cells compared with the effect on untreated control cells (data not shown).

Additional experiments were performed to determine whether ICB cells could serve as a source of nutrition and offset encystment associated with AH. Trophozoites were incubated in PYG or AH alone or AH on a confluent monolayer of ICB cells, and cultures were examined for evidence of encystment. AH did not induce encystment if trophozoites were incubated on a confluent monolayer of ICB cells (Fig 5). By 48 hours, 40% of the trophozoites incubated in AH alone had encysted. By contrast, only 6% of the trophozoites that were incubated in AH on a confluent monolayer of ICB cells
encysted. Almost 100% of the trophozoites incubated in AH alone had encysted by 96 hours, whereas only 13% of the trophozoites incubated in AH on a confluent monolayer of ICB cells had encysted. The capacity of ICB cell cultures to prevent AH-induced encystment was comparable to that found with PYG culture medium control and supports the notion that ICB cells prevent encystment by preventing nutrient deprivation.

**DISCUSSION**

The purpose of this study was to determine why *Acanthamoeba* keratitis does not usually progress to produce intraocular infections. Only a few cases of endophthalmitis have been reported in which *Acanthamoeba* spp. have been implicated in intraocular infections. However, each of these cases of endophthalmitis is questionable. For example, in 2001, a patient who had received four corneal transplants due to *Acanthamoeba* keratitis presented with intraocular infection. It is possible that this case of endophthalmitis was due to the introduction of trophozoites into the eye at the time of corneal transplantation and not the natural progression of the trophozoite beyond the corneal endothelium.

To date, it is not known why *Acanthamoeba* keratitis rarely progresses beyond the cornea to produce an intraocular infection. One hypothesis proposed that the amoebae could not progress beyond the corneal endothelium and Descemet’s membrane to enter the AC. Because trophozoites were capable of killing human corneal endothelial cells and penetrating Descemet’s membrane in vitro, we conclude that trophozoites can progress beyond the corneal endothelium and Descemet’s membrane to enter the AC. This is not surprising, as Descemet’s membrane contains type IV collagen, which can be degraded by *Acanthamoeba* MIP-133 protease. Because serine protease inhibitors, including anti-MIP-133 antibody, prevent trophozoite penetration of Descemet’s membrane, it is likely that trophozoites use MIP-133 to penetrate this membrane. This correlates with previous findings showing that anti-MIP-133 antiserum prevents trophozoite penetration of a collagenuous matrix (Matrigel; Collaborative Biomedical Products), which was used as a model of the corneal basement membrane. As the serine protease inhibitors were not toxic at the doses used and did not prevent trophozoite migration, it is likely that the inhibitors were able to prevent the serine proteases involved in the penetration of Descemet’s membrane in vitro, which supports the hypothesis that trophozoites can employ the serine protease MIP-133 to penetrate the cornea and enter the AC.

The second hypothesis proposed that the amoebae can enter the AC; however, the AH contains factors that either induce encystment or kill the amoebae. Our results show that trophozoites encyst in AH in vitro, but are still viable after a 7-day incubation in 10× AH, suggesting that it is unlikely that AH contains a factor that is toxic to trophozoites. We speculate that AH-induced encystment of trophozoites in vitro may be due to nutrient deprivation. Injection of trophozoites into the AC did not induce their encystment or death. However, the presence of trophozoites elicited a robust neutrophil infiltrate, which undoubtedly contributed to the elimination of trophozoites. Because trophozoites did not die in the AC and were capable of killing ICB cells in vitro, we hypothesized that trophozoites could feed on the iris cells in vivo. Exposure to AH did not induce encystment if trophozoites were incubated with ICB cells as a source of nutrition in vitro. Thus, it is likely that trophozoites feed on iris cells in vivo, thereby preventing their encystment in the AC.

Previous studies have suggested that the innate immune apparatus is involved in the resolution of *Acanthamoeba* keratitis in animals. In fact, recruitment of neutrophils into the cornea through intrastromal injections of recombinant MIP-2, a chemotactic factor for neutrophils, results in transient infiltration of neutrophils followed by a rapid resolution of corneal infection. Moreover, neutrophils are capable of killing trophozoites in vitro, and deletion of neutrophils with anti-Chinese hamster neutrophil antibody results in an early onset and more severe infection, compared with untreated hosts. Furthermore, activated macrophages are also capable of killing trophozoites in vitro, and deletion of conjunctival macrophages with the macrophagocidal drug dichloromethylene diphosphonate (clodronate), results in a chronic, more severe form of *Acanthamoeba* keratitis.

Scleritis has been reported in patients with *Acanthamoeba* keratitis; however, the scleral inflammation was attributed to an immune-mediated response to necrotic organisms and was not believed to be the result of active infection. This finding is consistent with the present data indicating that intraocular infection is associated with transient inflammation and elimination of trophozoites.

Treatment of *Acanthamoeba* keratitis often involves the use of topical steroids to control the inflammation associated with disease. However, dexamethasone treatment increases...
the incidence, severity, and duration in the Chinese hamster model of Acanthamoeba keratitis, presumably by inhibition of macrophage and neutrophil activation. Moreover, dexamethasone induces excystment and proliferation of trophozoites, and also stimulates the Acanthamoeba-mediated cytopathic effect of corneal epithelial cells in vitro. Consistent with previous findings, the results of the present study suggest steroid treatment may increase the corneal load of trophozoites and exacerbate ocular disease.

Although the innate immune apparatus is clearly involved in the resistance to Acanthamoeba infections, the role of the adaptive immune apparatus is not clear. The immune response to corneal infection with Acanthamoeba spp. does not normally elicit delayed-type hypersensitivity (DTH) or serum IgG antibody against Acanthamoeba antigens. Furthermore, lymphocytes are sparse in the histopathological specimens of Acanthamoeba keratitis biopsy specimens from patients and experimental animals. Collectively, these results suggest that T cells fail to inhibit the establishment or progression of Acanthamoeba keratitis.

In summary, our results suggest that Acanthamoeba trophozoites use MIP-1α, a serine protease, to penetrate Descemet’s membrane and the corneal endothelium. However, once in the AC, an intense neutrophil infiltrate is evoked with the eventual clearance of intraocular trophozoites, implicating the innate immune apparatus in the resolution of intraocular infection with Acanthamoeba trophozoites. The findings in this study suggest that the innate immune apparatus can circumvent ocular immune-privilege to control an intraocular infection. However, it is unclear why the innate immune apparatus is apparently incapable of resolving Acanthamoeba infections in the human cornea. Future studies are necessary to determine why the innate immune apparatus is effective at eliminating trophozoites from inside the eye, yet ineffective at controlling infection of Acanthamoeba trophozoites at the corneal surface.

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References


