

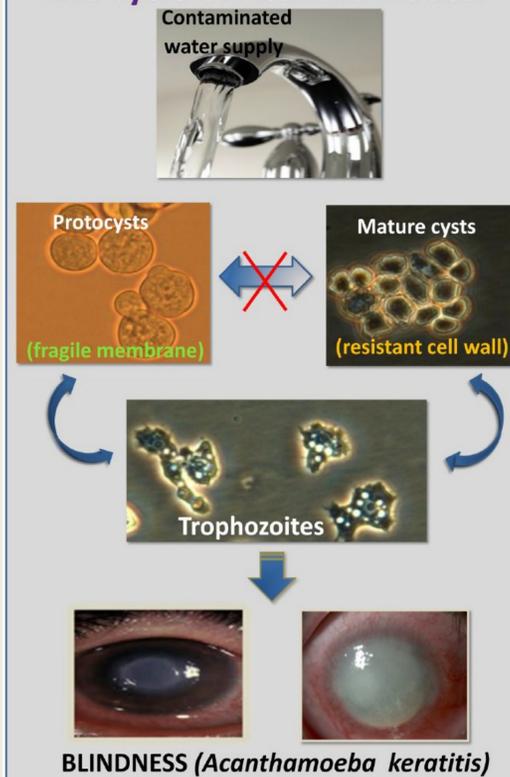
BACKGROUND

Infection of the cornea by a common parasite called *Acanthamoeba* leads to blindness. Almost 80% of those patients diagnosed are **contact lens** users.

This sporadic outbreak of a painful, sight threatening, parasitic infection of the cornea follows minor corneal injuries upon exposure of contact lenses to contaminated natural, man-made & clinical water sources.

A major pathogenic factor of *Acanthamoeba* is **Mannose-Binding Protein (MBP)**, a trans-membrane surface protein, which initiates the parasitic adhesion, penetration and corneal tissue destruction.

Life cycle of *Acanthamoeba*



Previously, only TWO distinct life forms of *Acanthamoeba* have been identified:

- actively motile **trophozoites** stage that causes destruction of corneal tissue;
- double-walled/ dormant **mature cysts** that is highly resistant to drugs.

JUSTIFICATION

- Current therapies have failed due to rapid transition of trophozoites into the mature cysts.
- *Acanthamoeba* shares similar biochemical process with humans so it is difficult to target with drugs.
- Our recent discovery of a third life stage termed **protocysts**, characterised by fragile cell wall and is likely to be much more susceptible to antiparasitic drugs.

CONCLUSION/FUTURE WORK

- Conditions have been established that trigger changes in *Acanthamoeba* life stages
- *Acanthamoeba* mannose-binding protein will be characterised to understand how it binds to sugars.
- Anti-MBP antibodies and carbohydrate polymers will be used to block *Acanthamoeba* adhesion.

RESEARCH QUESTIONS

- Is the protocysts life stage, more vulnerable towards effective treatment of *Acanthamoeba* infection?
- Can blocking parasitic adhesion via MBP prevent disease?

AIMS

To understand:

- The conditions that trigger *Acanthamoeba* to switch between different life forms.
- How mannose-binding protein enables adhesion to the cornea.



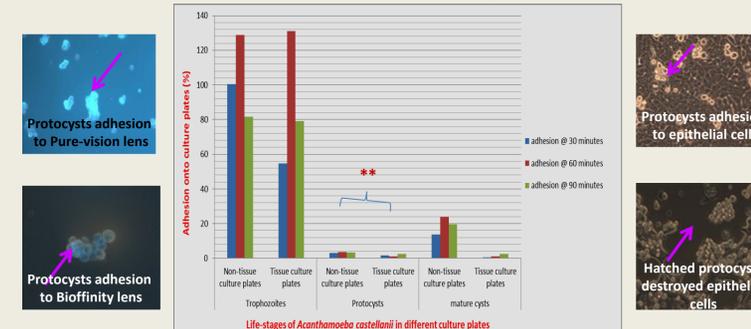
METHODS

- Study of changes of *Acanthamoeba* life forms by cell culture & microscopy.
- Attachment of *Acanthamoeba* and cell killing.
- Cloning and production of recombinant *Acanthamoeba* MBP.
- Generation of antibodies against purified MBP to block *Acanthamoeba* adhesion.

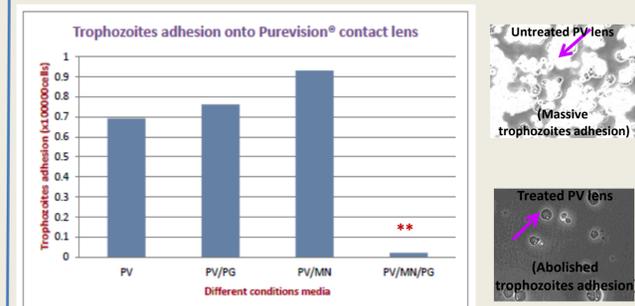
RESULTS

1. *Acanthamoeba* adhesion & differentiation

- Three distinct life stages of *Acanthamoeba* spp have now been established.
- The different life forms have different adhesion properties in the order : trophozoites > mature cysts > protocysts
- All life stages sticks to:
 - *contact lenses e.g. Pure-vision® (left, above) and Biofinity® (left, below);
 - *Polystyrene plates (not shown); and
 - *Epithelial cells: untreated (right, above); destroyed by *Acanthamoeba* (right, below).



- Protocysts rapidly & reversibly convert to trophozoites.
- However, protocysts can NOT convert directly into mature cysts.



2. Role of mannose

- Preliminary data suggest that a combination of propylene-glycol (PG) and mannose (MN) abolish *Acanthamoeba* adhesion to pure-vision (PV) contact lenses.