The Great Escape: How Urea Amidolyase Allows A Pathogenic Fungus To Escape The Immune System

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Abstract:
According to Rice University, 70% of people are affected by the infectious fungus Candida albicans. The immune system uses T and B cells to stop pathogens. People with suppressed immune systems, such as transplant patients, and AIDS or cancer patients, lack functional T and B cells, and rely on macrophages to destroy Candida. Candida can kill and exit macrophages due to an enzyme: urea amidolyase (UAL). While in the macrophage, an environmental change causes Candida to morphologically switch from a sphere to a structure with hyphae. UAL converts urea to ammonia (NH₃) and CO₂, creating an environment hazardous for the macrophage. The Greenfield SMART (Students Modeling A Research Topic) team used 3D printing technology to model UAL. The bistin carboxylase (BC) domain uses energy from ATP to cleave CO₂ to the swinging arm portion, or biotin carboxyl carrier protein (BCCP) domain. The BCCP domain swings across UAL, attaching CO₂ to urea forming allophanate in the carbonyl transferase (CT) domain. Allophanate moves to the allophane hydrolyase (AH) domain, hydrolysing the allophanate into CO₂ and NH₃. Increases in CO₂ and NH₃ cause the macrophage to form, destroying macrophages and allowing Candida to spread. Researchers could block UAL’s active sites to prevent Candida’s macrophage-killing shape change, preventing systemic candidiasis without damaging human cells. Supported by a grant from NIH-CTSA.

A. Systemic Candidiasis Affects People With Weakened Immune Systems:

Normally, the human immune system uses white blood cells called T and B cells to fight pathogens. These cells do not function as well in people with weakened immune systems, such as children who have recently had a transplant, people with AIDS, and cancer patients. People with weakened immune systems must then rely on macrophages, another type of white blood cell, to fight pathogens. A common fungus, Candida albicans, can cause systemic candidiasis, a potentially lethal infection that spreads throughout the body and is difficult to treat.

B. How Candida Escapes From Macrophages:

Step 1: When any foreign agent, such as Candida albicans, infects an organism, macrophages engulf and break down the pathogen.

Step 2: Once inside the macrophage, Candida adjusts its metabolism to produce large amounts of urea. An enzyme called Urea Amidolyase (UAL) converts this urea into ammonia (NH₃) and carbon dioxide (CO₂). The increased concentrations of NH₃ and CO₂ change the internal macrophage environment surrounding the engulfed Candida.

Step 3: Increases in NH₃ and CO₂ signal the creation of DAG-grider-like projections called hyphae. These projections kill the macrophage and allow the Candida to escape from inside.

Step 4: The Candida is now free to continue colonizing the host organism’s body. Ultimately this could lead to fatal systemic candidiasis.

C. How Do We Know UAL Makes A Difference?

In an experiment, mice were injected with wild type Candida albicans in image to the right13. Over time, survival rate of the infected mice was calculated. Similarly, mice were injected with Candida lacking the gene for UAL, called Dur 1, 2. This null mutant form is referred to as KWN6 (+). Candida lacking Dur 1, 2 cannot make UAL. The survival rate of the mice infected with KWN6 was also calculated. The mice infected with KWN6 had the highest survival rate (about 50%) because these Candida could not make hyphae to break out of the macrophages.

D. The Four Domains of Urea Amidolyase (UAL)

1. The BC domain utilizes energy released from splitting ATP to attach CO₂ to the BCCP swinging arm component.

2. The BCCP domain swings across UAL to attach the CO₂ to urea in the CT domain.

3. The CT domain forms allophanate, which then travels to the AH domain.

4. The AH domain has an active site that hydrolyses allophanate into two CO₂ molecules and two NH₃ molecules, creating an environment for the Candida to form hyphae.

E. How UAL Functions to Increase CO₂ and NH₃ Inside the Macrophage

F. The Future of Urea Amidolyase in Preventing Systemic Candidiasis

In the future, targeting urea amidolyase could decrease occurrences of systemic candidiasis and related casualties. Future research could possibly find a method of preventing hyphae from being produced and thus not allowing the fungus to escape the macrophages. A method of inhibiting one of UAL’s domains without interfering with human active sites could cause UAL to no longer function. Thus, people with compromised immune systems would be less susceptible to developing systemic candidiasis.

SMART Teams are supported by the National Center for Advancing Translational Sciences, National Institutes of Health, through Grant Number 8UL1TR000055. Its contents are solely the responsibility of the authors and do not necessarily represent the official views of the NIH.

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