

# Human Monoclonal Antibodies for Neutralization and Diagnosis of H5N1

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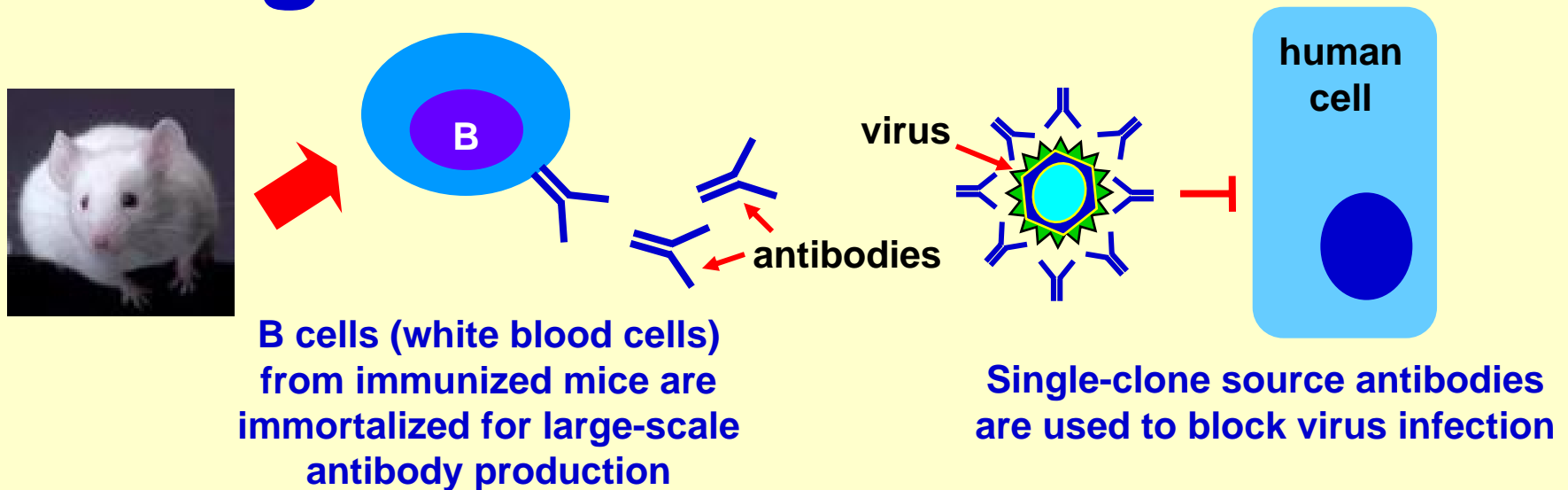
MITRE Sponsored Research



# Problem

- **Can B cells of human origin be manipulated to produce antibodies able to bind to bio-targets?**
  - **Many biological organisms and proteins remain a threat (BW or natural)**
  - **Immunizations have covered only a small fraction of the spectrum**
  - **Antibody therapy is a potential alternative**

# Background



- Vaccination, drug and antibody therapies are the best available interventions against infectious diseases and toxin exposure
- Where no vaccine or drugs are available antibody therapy is an option
- Antibody therapies continue to rely on animal or “humanized” sources
- Technologies for producing antibodies of human origin will drastically expand our control of disease and improve diagnostic tools

# Objective

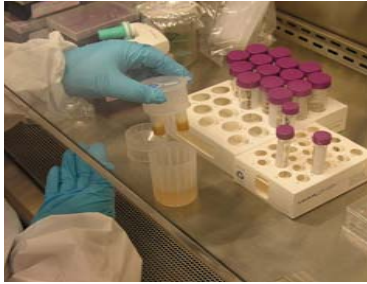
- A new method for production of human monoclonal antibodies will be optimized for treatment of infectious diseases
- The resulting antibodies will be tested for therapeutic blocking of avian influenza viruses
- Ability to identify virus also will be tested
- The “contact point” between target viral proteins and antibodies will be mapped

# Activities

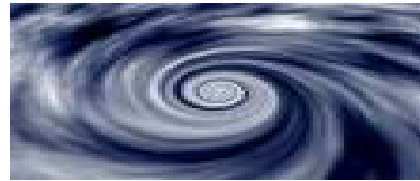
- **Human B cells are isolated from tonsils obtained after tonsillectomy**
- **B cells are manipulated to generate antibody producing clones with indefinite lifespan**
- **The generated antibodies are tested for binding to target proteins on H5N1 avian influenza**
- **Mapping technologies will define antibody target contact points, identifying antibodies useful for diagnostics and therapeutics**

# Highlight

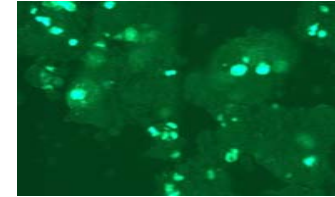
## 1. Efficient Immortalization of Human B cells



Epstein-Barr Virus concentration (EBV)

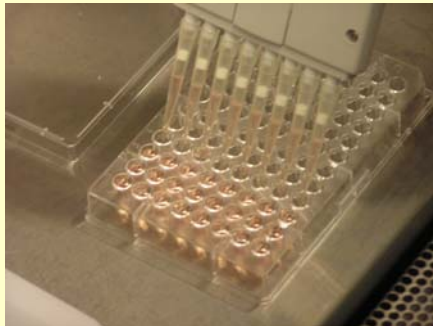


Tonsil extracted B cells are "spinfectd" with EBV

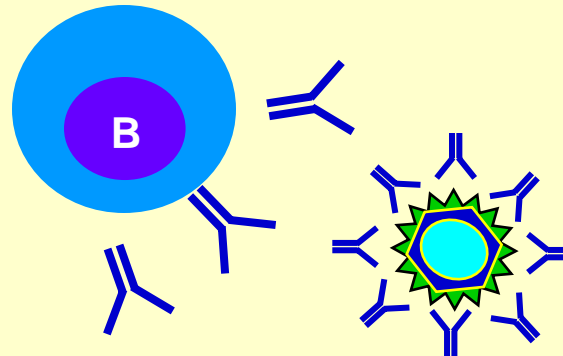


"Immortalized" B cells shown with fluorescent label

## 2. *In Vitro* Antibody Production

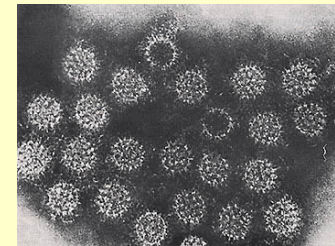


Newly immortalized B cells are further treated to induce antibody secretion *in vitro*



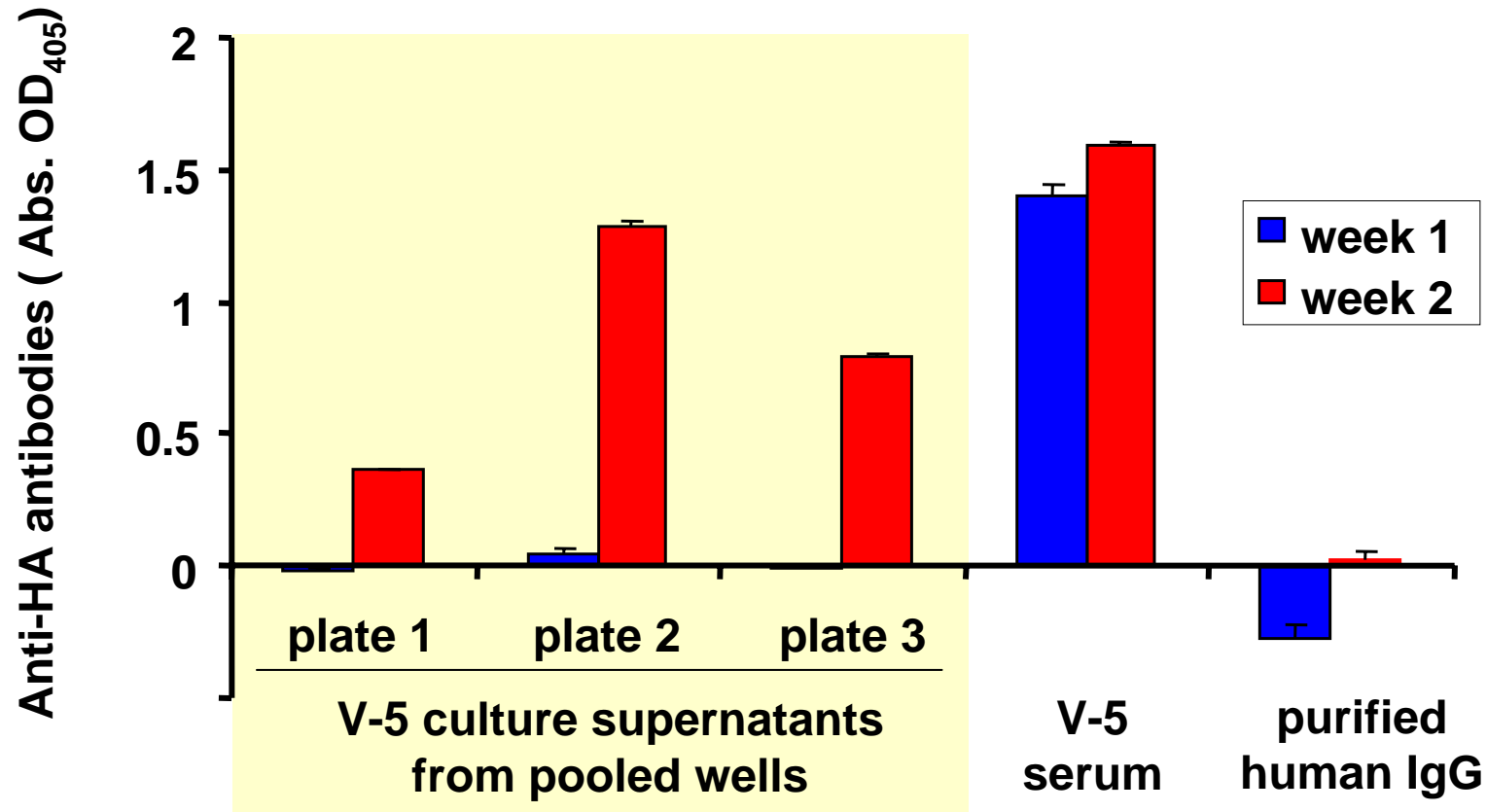
## 3. Selection of Virus-specific B cells

Antibody coated flu virus



B cells secreting antibodies against avian influenza H5N1 virus are isolated and expanded

# Highlight



Immortalized pools of B cells from human volunteer V-5 were induced; by 2 weeks they produced detectable anti-H5N1 hemagglutinin (HA) antibodies (IgG). Graph shows B cell supernatants probed by ELISA to detect anti-HA IgG antibodies. Samples shown are pooled supernatants from three 96-well plates. IgG levels shown as absorbance at OD<sub>405</sub>

# Impacts

- **Immortalized B cell clones are treated to produce large quantities of monoclonal antibodies**
- **H5N1 virus neutralizing antibodies can be selected and purified for use as therapy**
- **The antibodies will be diagnostic tools**
- **FDA guidelines are available for large-scale manufacture of antibodies from immortalized B cell lines in bioreactors**

# Future Plans

- Testing monoclonal antibodies for universal neutralization of influenza viruses
- Application of the technology to other emerging biothreats
- Applying the technology to stockpile therapies against BW agents (bacteria, viruses and toxins)

