

MIT OpenCourseWare  
<http://ocw.mit.edu>

7.344 Directed Evolution: Engineering Biocatalysts  
Spring 2008

For information about citing these materials or our Terms of Use, visit: <http://ocw.mit.edu/terms>.

# Enzyme evolution using yeast surface display

Boder, E.T.; Wittrup, K.D. Yeast surface display for screening combinatorial polypeptide libraries. *Nat. Biotechnol.* **1997**, *15*, 553-557.

Shiraga, S.; Kawakami, M.; Ishiguro, M.; Ueda, M. Enhanced reactivity of *Rhizopus oryzae* lipase displayed on yeast cell surfaces in organic solvents: potential as a whole-cell biocatalyst in organic solvents. *App. Environ. Microbiol.* **2005**, *71*(8), 4335-4338.

# Wittrup Lab yeast surface display

- What are the limitations the authors identify with previous display methods that make their system attractive? What kinds of proteins are hard to express?
- Explain the display scaffold. How are proteins displayed? (Figure 1)
- How is the surface expression of scFvs verified? (Figure 2) How is the number of fusions quantified?
- How does the enrichment factor compare to previously discussed methods?
- Explain their library generation and selection strategy. What are the results?
- What are the benefits/downsides to using this method?

# Figure 1- Aga-based protein display

Image removed due to copyright restrictions.  
Please see Fig. 1 in Boder, E. T., and K. D. Wittrup. "Yeast surface display for screening combinatorial polypeptide libraries." *Nat. Biotechnol.* 15(1997): 553-557.

# Figure 2 – characterization of yeast surface fusions

Image removed due to copyright restrictions.  
Please see Fig. 2 in Boder, E. T., and K. D. Wittrup. “Yeast surface display for screening combinatorial polypeptide libraries.” *Nat. Biotechnol.* 15(1997): 553-557.

# *Rhizopus oryzae* lipase display

- What are the authors trying to do and why is this significant?
- What is the strategy used? How is the enzyme library made and screened?
- What are the results? (Table 2)
- What are the pros/cons of the method?