



**CALIFORNIA STATE SCIENCE FAIR
2003 PROJECT SUMMARY**

Name(s) Darcy E. Bradley	Project Number S1304
Project Title Folate-Yeast Project: A Novel Solution to the World's Malnutrition Problem	
Objectives/Goals The objective is to develop a strain of yeast high in folate through a process of selective mutation.	
Abstract Methods/Materials SAF yeast was streaked onto PDA media. A folate analog, methotrexate, was obtained and analog disks were prepared. A single yeast colony was collected and spread onto a PDA plate containing analog disks. The methotrexate inhibited growth in the regions surrounding the disks; however, a few mutant colonies were observed growing. Mutant colonies were collected and plated on a PDA media with three methotrexate disks in the center of the plate. This process was repeated several times to obtain pure mutant colonies. Next, a folate-dependent lactobacillus, 7469, was obtained to be used as an indicator bacteria. Colonies of 7469 were added to a solution of Folic Acid Minus media. Single mutant yeast colonies were collected from the plate containing the analog disks and re-plated on Folic Acid Minus media with 7469. Excretion growth surrounding the colonies was observed as an indication of which yeast colonies were over producing folate. Four colonies with most excretion plus a wild type colony were collected and placed in a solution of Folic AOAC Medium plus varying levels of methotrexate (0-12 drops, respectively) in order to eradicate any non-over producing folate colonies.	
Results Growth from methotrexate solutions will be collected and assayed by placing mutant colonies in a serial dilution array to eliminate the toxin. The analog-free mutant yeast colonies will then be re-plated and grown. Colonies will then be added to a sourdough starter, which will, in turn, be used to make bread. The amount of folate will be quantified using a mass spectrometer. Folate content of the bread produced from this experiment will optimally contain 400-800 micrograms in two slices (thus providing the U.S. Public Health Service recommended daily allowance of folate).	
Conclusions/Discussion The process of selective mutation effectively created a strain of yeast containing elevated levels of folate. Isolation and verification of a high folate producing strain of yeast that could be used in many food products would profoundly improve the human condition. Such a folate supplement would greatly reduce the prevalence of neural tube defects among infants and lower the occurrence of diseases related to elevated homocysteine levels including heart disease and cancer.	
Summary Statement An experiment was developed to create a yeast strain high in folate through a process of selective mutation.	
Help Received Research was performed in AgBioSciences Bldg. at Montana State University, supervised by Dr. David Sands, and at Cate School, supervised by Ms. Cheryl Powers.	