DNA IN THE UK

By Erica Van Coverden

Dr. Kelly Goodwin, a microbiologist at AOML, finished up the year 2000 working on a small grant for research of "Molecular Biological Techniques for Environmental Microbiology." Dr. Goodwin spent five weeks in Oct.-Nov. 2000 working with Dr. Ian McDonald and Dr. Colin Murrell at the University of Warwick, Coventry, UK.

Dr. Goodwin decided to use a molecular approach to study *Leisingera methylohalidivorans*, a marine microorganism that can consume near-ambient concentrations of methyl bromide (~ 2 pM), but it also can grow on up to 500 µM of this toxic substrate. Methyl bromide is a fumigant that contributes to ozone-depletion, and there is much interest in quantifying its global budget. Consumption of both high and low concentrations of methyl bromide are of distinct environmental significance. For example, the ability to consume toxic levels of methyl bromide may find industrial applications because of environmental engineering efforts to limit the amount of methyl bromide escaping to the atmosphere during fumigation procedures.

Several approaches were used simultaneously to maximize the chance of identifying the gene during Dr. Goodwin's relatively short stay in the UK. In one approach, total protein was extracted from cultures grown under various conditions. The protein was run on gels, stained, and the gels were scrutinized for proteins induced during degradation of methyl bromide. Those proteins then would be sequenced and the amino acid sequence would be used to backtrack to the DNA sequence that codes for that protein. In another approach, total DNA was extracted, digested with restriction enzymes, run on a gel, and fixed on blotting paper. The DNA was then hybridized with radiolabeled probes designed from DNA sequences of the (hopefully homologous) genes from terrestrial methyl-bromide degrading bacteria. The last approach used PCR to amplify specific DNA sequences from the total DNA extracted from the bacterial cultures. The DNA was amplified using primers based, again, on the genes sequenced from the terrestrial methyl bromide degraders.

The work proceeded efficiently using a three-person team consisting of Dr. Goodwin, Dr. McDonald, and graduate student, Karen Warner. Indeed, the team obtained initial sequence for a gene involved in methyl bromide degradation (called the *cmu*A gene) by the end of Dr. Goodwin's stay. Significant progress was made in a short amount of time, and Dr. Goodwin returned with knowledge and skills that will allow her to explore genomes here at AOML.