

OCT 13 1989

COVER SHEET  
FOR TRIP REPORTS SUBMITTED TO THE  
OFFICE OF ENERGY RESEARCH

Destination(s) and Dates for Vienna, Austria-September 13-16, 1989  
Which Trip Report Being Submitted: Birmingham, England-September 18-23, 1989

Name of Traveler: Jonathan Woodward

Joint Trip Report  Yes  
 No

If so, Name of Other Traveler(s): \_\_\_\_\_

MASTER

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ORNL/FTR--3390

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**ORNL**

**FOREIGN TRIP REPORT**

ORNL/FTR-3390

DATE: September 29, 1989

SUBJECT: Report of Foreign Travel of Jonathan Woodward, Development Staff Member II,  
Chemical Technology Division

TO: Alvin W. Trivelpiece

FROM: Jonathan Woodward

PURPOSE: To attend and participate in TRICEL 89, an international symposium on  
*Trichoderma* cellulases, Vienna, Austria, September 14-16, 1989, and to visit  
and to present a seminar at Aston University, Birmingham, England, on  
September 20, 1989.

**SITES**

VISITED: 9/14-16/89 TRICEL 89, Vienna, Austria

9/20/89 Aston University, Birmingham, England P. E. Barker

ABSTRACT:

The traveler attended TRICEL 89, the first international symposium on  
*Trichoderma* cellulases. He presented a paper summarizing his work at ORNL.  
He also visited Aston University in Birmingham, England, and gave a lecture  
describing his work at Oak Ridge National Laboratory (ORNL). In Vienna, the  
latest knowledge concerning *Trichoderma* cellulases was obtained, and two  
contacts were established that will lead to unclassified and nonproprietary  
cooperative research efforts between ORNL and these European colleagues.  
In Birmingham, unclassified and nonproprietary discussions were held with  
Professor P. E. Barker and his staff in the Department of Chemical Engineering  
and Applied Chemistry. As a result of these discussions, areas of research  
cooperation between ORNL and Aston University were identified, and it is  
hoped that this cooperation will attract funding from the National Science  
Foundation (U.S.) and the European Economic Community (EEC).

## REPORT OF TRAVEL TO AUSTRIA AND ENGLAND

September 13-23, 1989

Jonathan Woodward

The traveler attended TRICEL 89, the first international symposium on *Trichoderma* cellulases, held at the Technical University, Vienna, Austria, on September 14-16, 1989. This conference was particularly relevant to the traveler's responsibilities at ORNL because he has been involved in cellulase research since joining ORNL in 1980. The fact that an entire conference was devoted to the enzyme cellulase produced by a single fungus indicates the importance of this enzyme in relation to its function, namely, the conversion of cellulosics into fermentable sugar. A total of 95 participants, representing 24 different countries, attended the conference.

There were six technical sessions: (1) cellulase protein structures; (2) identification, mechanism of action, and activities of *Trichoderma* cellulases and xylanases; (3) regulatory aspects of cellulase secretion and location; (4) biochemical aspects of cellulase formation; (5) cellulase gene technology; and (6) production and application of cellulases. There was also a poster session, with the nature of the posters fitting into all the technical sessions.

The traveler made an oral presentation of his work in the first technical session on cellulase protein structures entitled "Denaturation and Renaturation of *Trichoderma reesei* C30 Cellobiohydrolase I." The talk was well received and of particular interest to Dr. Tuula Teeri, of the Biotechnical Laboratory of the Technical Research Centre of Finland, Espoo, Finland. Dr. Teeri has cloned *T. reesei* cellobiohydrolase I (CBH I) in yeast and, thus far, has found it very difficult to refold the nascent recombinant protein into an active enzyme. Generally, the protein aggregates upon folding and becomes inactive. In a cooperative research effort, Dr. Teeri will send the traveler some of the inactive recombinant CBH I so that he can attempt to correctly refold and, hence, activate this recombinant enzyme using techniques developed in his laboratory at ORNL.

At ORNL, work funded by the U.S. Department of Energy entitled "Kinetics of Enzyme-Catalyzed Reactions" involves the purification of cellulase enzyme components. Recently, Dr. Marc Claeysens, of the State University at Gent, Belgium, and a participant at TRICEL 89, developed an affinity gel for the purification of CBH I. Dr. Claeysens will send a sample of the affinity material,  $\rho$ -aminobenzylthiocellobioside, to the traveler so that he can determine its usefulness in the purification of CBH I at ORNL. Thus, the contacts made with Drs. Teeri and Claeysens may be of importance to DOE programs that are funding basic research at ORNL in the cellulase area.

From the standpoint of learning up-to-date discoveries concerning cellulases, as well as the representation by ORNL, the conference was a resounding success. Scientifically, several significant findings are worth mentioning. Cellulase enzymes are composed of two domains: (1) catalytic, and (2) cellulose-binding. It has been shown that tyrosine molecules are essential for the binding of the CBH I to cellulose (see below). More specifically, glutamic acid residue 126 of CBH I and glutamic acid residue 127 of endoglucanase I are present in the catalytic sites (domains) of these enzymes. Using site-directed mutagenesis, aspartic residue 173 has been shown to be essential for the catalytic activity of CBH II since its substitution by an asparagine residue abolishes activity. In the traveler's opinion, the most exciting new information concerning cellulases came when Dr. Teeri described work on the three-dimensional structure of the catalytic and binding regions of CBH I. The catalytic binding site, or core protein, of CBH I has been crystallized, and x-ray diffraction patterns suggest that the active site of this enzyme is similar to a tunnel lined with tyrosine and negatively charged groups through which an individual cellulose chain would pass and be hydrolyzed to glucose. Evidence was also presented suggesting that the cellulose-binding site was similar to a wedge which could penetrate cellulose fibers and strip off individual cellulase chains that would then enter the catalytic "tunnel" of the enzyme.

In conclusion, the conference provided exciting, new knowledge about the cellulase family of enzymes. It was also highly relevant to DOE's program at ORNL on the kinetics of enzyme-catalyzed reactions and should provide some fruitful collaboration between ORNL and other laboratories in Europe studying cellulase. It is highly recommended that the United States participate in the next TRICEL conference, which has been tentatively scheduled for 1992 in Finland.

After the conclusion of TRICEL 89, the traveler went to England, where he presented an invited lecture at Aston University, Birmingham, entitled "Cellulase Biochemistry and Biotechnology," on September 20, 1989. The traveler was hosted by Professor Phil Barker of the Department of Chemical Engineering and Applied Chemistry, and it was to members of this department that the lecture was given. After the lecture, the traveler met with several members of the department for technical discussions concerning their work. Of particular note was the discussion held with Dr. Martin Beevers, a physical chemist who is interested in the study of the conformation of polymers using nuclear magnetic resonance (NMR) spectroscopy. It is only very recently that NMR has been used as a tool to probe the structure of proteins and act to complement x-ray crystallography in the study of protein structure. Dr. Beevers and the traveler agreed to explore the possibility of establishing a U.S.-Western Europe Cooperative Science Program with sponsorship from the U.S. National Science Foundation (NSF) and the EEC. ORNL and Aston University have particular expertise that would complement each other in a cooperative program. The traveler and Dr. Beevers agreed that CBH I would be a good model to use in studying conformations of denatured and renatured forms of this enzyme compared with those of the native form. ORNL has expertise in the protein chemistry of CBH I, whereas Aston University has expertise in the use of NMR to study the relative mobility bond angles and degrees of freedom of chain polymers. Attempts to establish this collaboration will begin soon.

The traveler also visited the laboratories of Dr. George Ganetsos, who is studying large-scale carbohydrate processing and separation, and Dr. David Earp, who is carrying out the pyrolysis and gasification of biomass systems in molten salts.

The traveler returned to the United States on September 23, 1989.

## APPENDIX A. ITINERARY

September 10, 1989 Depart Knoxville for Miami

September 10-12, 1989 Participate in American Chemical Society National Meeting, Miami Beach

September 13, 1989 Depart Miami for Vienna, Austria

September 14-16, 1989 Participate in TRICEL 89, the first international conference on *Trichoderma cellulases*

September 16, 1989 Depart Vienna for London, England

September 17-18, 1989 Weekend and travel to Birmingham, England

September 19, 1989 Vacation

September 20, 1989 Present an invited lecture at Aston University, Birmingham, England

September 21, 1989 Vacation

September 22, 1989 Depart Birmingham for London, England

September 23, 1989 Depart London for Knoxville

## APPENDIX B. PERSONS CONTACTED

Professor Phil Barker      Aston University  
Birmingham, England

## Discussions on future collaboration between ORNL and Aston University.

Dr. Marc Claeysens Gent University  
Belgium

Dr. Tuula Terri      Biotechnical Laboratory  
                            Technical Research Centre  
                            Espoo, Finland

## Discussions on the cooperation in cellulase studies between ORNL and Gent State Technical Research Centre.

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