IICA

Consultant Final Report
IICA/EMBRAPA-PROCENSUL II

GENETIC RESOURCES AND BIOTECHNOLOGY

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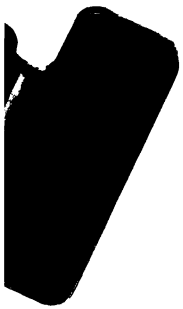

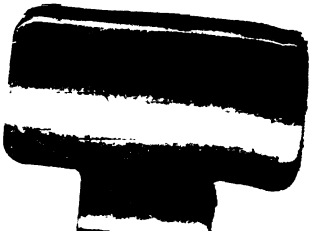

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GENETIC RESOURCES AND BIOTECHNOLOGY

Consultant Final Report
IICA/EMBRAPA-PROCENSUL II

James E. Maruniak

Brasília, agosto de 1989

INSTITUTO INTERAMERICANO DE COOPERAÇÃO PARA A AGRICULTURA
EMPRESA BRASILEIRA DE PESQUISA AGROPECUARIA

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APRESENTAÇÃO

A reprodução e difusão dos Relatórios de Consultores, no âmbito restrito das Diretorias das Unidades do Sistema Nacional de Pesquisa Agropecuária, vinculado à EMBRAPA, tem como objetivo principal o de divulgar as atividades desenvolvidas pelos consultores e as opiniões e recomendações geradas sobre os problemas de interesse para a pesquisa agropecuária.

As atividades de consultoria são realizadas no âmbito do Projeto de Desenvolvimento da Pesquisa Agropecuária e Difusão de Tecnologia na Região Centro-Sul do Brasil - PROCENSUL II, financiado parcialmente pelo Banco Interamericano de Desenvolvimento - BID e a EMBRAPA conforme os contratos de Empréstimo 139/IC-BR e 760/SF-BR, assinados em 14 de março de 1985 entre o Governo Brasileiro e o BID.

As opiniões dos consultores são inteiramente pessoais e não refletem, necessariamente, o ponto de vista do IICA ou da EMBRAPA.

A coordenação dos Contratos IICA/EMBRAPA agradeceria receber comentários sobre estes relatórios.

Horacio H. Stagno
Coordenador Contratos IICA/EMBRAPA

INTER-AMERICAN INSTITUTE FOR COOPERATION ON AGRICULTURE
IICA/EMBRAPA CONTRACT

CONSULTANT FINAL REPORT

1. Consultant's full name: *James E. Varuniak*

2. Specialist in: *Genetic Resources and Biotechnology*

3. Title of IICA Project: *2.SB.3*

4. EMBRAPA Program for which consultancy is provided:

PROGRAMA: PROCENSUL II
SUBPROGRAMA: 05-RECURSOS GENÉTICOS

IICA Project Activity Code: *2,SB.3.05*	Administrative Code: *R 4894 B1B 03 105*
Title of Activity of IICA Project corresponding to this consultancy	*Cooperation with EMBRAPA on research and applications of genetic resources, biotechnology and biologic control of plagues diseases and weeds.*

CONSULTANT CONTRACT PERIOD	DUTY LOCATION (Center)
April 12 to 20, 1989	*CENARGEN/EMBRAPA*
CONTRACT EXTENTION PERIOD (If any)	DUTY LOCATION (Center)

5. Financial support: *PROCENSUL II*

6. ACTIVITIES UNDERTAKEN BY THE CONSULTANT AND RESULTS

6.1 RESEARCH DONE UNDER DIRECT RESPONSIBILITY OF THE CONSULTANT

Research activities developed	Results Achieved
1. A rapid microextraction protocol for baculovirus DNA from either infected cell cultures or infected larvae was demonstrated.	1. A typed protocol was given to CENARGEN scientists and the experiment was performed.
2. Anticarsia baculovirus infected Anticarsia larvae brought from EMBRAPA Soja were tested for the effectiveness of the rapid microextraction protocol in purifying viral DNA.	2. About 20 Anticarsia baculovirus infected larvae were bled of hemolymph to obtain non-occluded virus. The experiment went well because reagents and equipment were readily available. There appeared to be purified DNA.
3. Restriction enzyme analysis of microextracted viral DNA and gel electrophoresis were performed	3. The restriction enzyme analysis and electrophoresis in agarose gels showed that Anticarsia baculovirus DNA could be easily visualized after ethidium bromide staining. Because of this, the need for radioactive phosphate for labeling viral DNA can be avoided in certain situations.
4. Reagents, cell culture supplies and titered Anticarsia baculovirus were brought to perform infected insect cell protein synthesis experiments.	4. These protein synthesis experiments could not be done because the radioactive methionine was stopped at customs in Rio de Janeiro for at least the two weeks from the time it was delivered. Experiments were performed, however, to isolate nuclei and cytoplasm from cell cultures when the isotope arrives.
5. Attempts were made to modify the rapid microextraction protocol to purify fungal mitochondrial DNA by using lysozyme to make fungal protoplasts before separating mitochondrial DNA from chromosomal DNA.	5. Fungal mitochondrial DNA purified by the modified microextraction protocol was visible after ethidium bromide staining. However, a considerable amount of degraded fungal chromosomal DNA was visible. One possibility is that the lysozyme used to make fungal protoplasts had considerable nucleases that degraded the chromosomal DNA. Another one is that the temperature and time of enzymatic digestion by the lysozyme was incorrect and this must be adjusted. A third possibility is that the fungi that were treated with liquid nitrogen already had degraded chromosomal DNA before the extractions procedure was started.

6.3 TRAINING ACTIVITIES DEVELOPED BY THE CONSULTANT

Date	Training subject matter	Type of event*	Number of beneficiaries	
			From EMBRAPA	From other institutions

Not applicable

* Short courses, seminars, conferences, etc.

6.4 IN-SERVICE TRAINING PROVIDED BY THE CONSULTANT

In-service training subject matter	Names of counterparts
1. Extraction methods for insect virus DNA.	Dr. Marlinda Lobo Pinheiro CENARGEN EMBRAPA
2. Isolation of nuclei and cytoplasm from insect cells.	
3. DNA restriction enzyme analysis and gel electrophoresis.	
4. Isolation of infected larvae hemolymph.	

6.5 ACTIVITIES IN SUPPORT OF RESEARCH STRATEGY AND PLANNING

Research subject matter	Research program to which subject matter is concerned
Insect virus research.	The CENARGEN - University of Florida cooperative research agreement was discussed. We determined that CENARGEN will be working on baculovirus protein synthesis in infected cells and amino acid sequencing of polyhedrins. CENARGEN will analyze DNA from Anticarsia baculovirus geographical isolates.

6.6 ACTIVITIES IN SUPPORT OF OTHER CENTERS AND UNIVERSITIES IMPROVING THE RESEARCH CENTERS LINKS WITH ABROAD

Subject matter on which links were recommended	Persons, centers and universities recomended for contact
Not applicable.	

6.7 PUBLICATIONS AND REPORTS UNDERTAKEN WITH THE CONSULTAT'S PARTICIPATION

Author(s)*	Title of publication or Report and other bibliografic identification

Not applicable.

* Personal, institutional, etc.

6.8 SUPPORT PROVIDED TO EMBRAPA RESEARCHERS IN THESIS AND DISSERTATION WORK

Name of the student	Thesis subject matter and sinthesis of advice
Paolo Zanotto	Genetic and molecular analysis of Anticarsia gemmatalis baculovirus polyhedrin gene. Started January 1988 at the University of Florida.

6.9 OTHER ACTIVITIES DEVELOPED BY THE CONSULTANT

Not applicable.

7. OTHER NATIONAL SYSTEM CENTERS, APART FROM DUTY STATION CENTER, ASSISTED BY THE CONSULTANT

Research center	Area of assistance provided by the consultant
EMBRAPA Soja, Londrina	The cooperative research program involving the DNA analysis of the *Anticarsia* baculovirus applied since 1979 was discussed and bioassays of plaque purified isolates were initiated in *Anticarsia* larvae.

8. CONSULTANT'S SUGGESTIONS AND TECHNICAL OR INSTITUTIONAL RECOMMENDATIONS FOR THE IMPROVEMENT OF THE RESEARCH SERVICE

8.1 The virology group at CENARGEN including Dr. Pinheiro, Dr. Sampaio and William Sihler are progressing well in their research on the Anticarsia gemmatalis baculovirus.

8.2 The structural protein analysis of Anticarsia and Spodoptera baculoviruses is complete. The infected cell protein synthesis of these viruses is planned and should be completed soon.

8.3 With the newly described rapid microextraction protocol developed at the University of Florida, the analysis of geographical isolates of baculoviruses will be greatly facilitated because the researchers will not be dependent on the delivery of radioactive phosphate from other countries.

8.4 Geographical isolates with genetic changes can either be tested in insect cell culture at CENARGEN or in larvae at CENARGEN or EMBRAPA Soja.

8.5 Plans are being formulated to have Dr. Marlinda Pinheiro present her research on Anticarsia baculovirus proteins at the Society for Invertebrate Pathology meetings August 20-24, 1989 in College Park, Maryland, U.S. Along with this we are planning for her to visit and do experiments on baculovirus at the University of Florida in Dr. Maruniak's lab.

8.6 Research should continue on fungal pathogen mitochondrial DNA and restriction fragment length polymorphism (RFLP). Dr. Maruniak is trying to coordinate a research visit for Dr. Myrian Tigano to the University of Florida and to determine if RELP cloned DNAs are available for her research.

8.7 Companies which send isotopes to CENARGEN should send a telex to a responsible person at CENARGEN and to a specific person at the custom's office in Rio de Janeiro. The telex should tell which flight the isotope is on and when it will be delivered.

9. AGREEMENTS OR COMMITMENTS ESTABLISHED WITH EMBRAPA RESEARCHERS IN-SERVICE OF THE FUTURE DEVELOPMENT OF RESEARCH IN THE CONSULTANT'S FIELD OF SPECIALIZATION

a. Plans are being made to have Dr. Marlinda Pinheiro visit the University for one month in August 1989.

b. If CNPq gives a very high priority to the CNPq-NSF cooperative research grant and if someone from EMBRAPA CENARGEN and Soja push hard, our grant has a possibility of being funded. Otherwise, the proposal will have to be submitted a third time which means that future visits and cooperation will be delayed.

10. CONSULTANT'S COMMENTS ON CIRCUMSTANCES WHICH AFFECTED THE CONSULTANCY WORK

As usual the cooperation of the different research groups (plant molecular biology and biological control) made the research easy and enjoyable.

The lack of radioactive methionine did not permit us to do the main purpose of this research visit. As mentioned earlier (see 8.7), this isotope purchased in the U.S. by Dr. Maruniak from New England Nuclear was sent on Varig but was stopped at customs in Rio de Janeiro. There are a number of reasons (some of which I do not know) why this happened. First, when radioisotopes are sent to Brazil they should never arrive on Friday; this happened with the radioactive methionine. Secondly, the container and the paper work for customs should clearly be labeled perishable not just rush. Thirdly, if there are problems finding the isotope, Luis Travelli, customer services manager at Varig cargo terminal at the Rio de Janeiro international airport phone number 393-8744, should be contacted. The flight number and airbill number must be known and given to him.

Fecha: April 28, 1989

James E. Blumenthal
Firma del Consultor

Programa II. Geração e Transferência de Tecnologia

O Programa de Geração e Transferência de Tecnologia é a resposta do IICA a dois aspectos fundamentais: (i) o reconhecimento, por parte dos países e da comunidade técnico-financeira internacional, da importância da tecnologia para o desenvolvimento produtivo do setor agropecuário; (ii) a convicção generalizada de que, para aproveitar plenamente o potencial da ciência e da tecnologia, é necessário que existam infra-estruturas institucionais capazes de desenvolver as respostas tecnológicas adequadas às condições específicas de cada país, bem como um lineamento de políticas que promova e possibilite que tais infra-estruturas sejam incorporadas aos processos produtivos.

Nesse contexto, o Programa II visa a promover e apoiar as ações dos Estados membros destinadas a aprimorar a configuração de suas políticas tecnológicas, fortalecer a organização e administração de seus sistemas de geração e transferência de tecnologia e facilitar a transferência tecnológica internacional. Desse modo será possível fazer melhor aproveitamento de todos os recursos disponíveis e uma contribuição mais eficiente e efetiva para a solução dos problemas tecnológicos da produção agropecuária, num âmbito de igualdade na distribuição dos benefícios e de conservação dos recursos naturais.

INSTITUTO INTERAMERICANO DE COOPERAÇÃO PARA A AGRICULTURA

O Instituto Interamericano de Cooperação para a Agricultura (IICA) é o organismo especializado em agricultura do Sistema Interamericano. Suas origens datam de 7 outubro de 1942, quando o Conselho Diretor da União Pan-Americana aprovou a criação do Instituto Interamericano de Ciências Agrícolas.

Fundado como uma instituição de pesquisa agronômica e de ensino, de pós-graduação para os trópicos, o IICA, respondendo às mudanças e novas necessidades do Hemisfério, converteu-se progressivamente em um organismo de cooperação técnica e fortalecimento institucional no campo da agropecuária. Essas transformações foram reconhecidas oficialmente com a ratificação, em 8 de dezembro de 1980, de uma nova convenção, que estabeleceu como fins do IICA estimular, promover e apoiar os laços de cooperação entre seus 31 Estados membros para a obtenção do desenvolvimento agrícola e do bem-estar rural.

Com um mandato amplo e flexível e com uma estrutura que permite a participação direta dos Estados membros na Junta Interamericana de Agricultura e em seu Comitê Executivo, o IICA conta com ampla presença geográfica em todos os países membros para responder a suas necessidades de cooperação técnica.

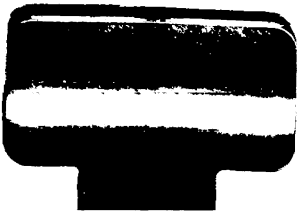
As contribuições dos Estados membros e as relações que o IICA mantém com 12 Países Observadores, e com vários organismos internacionais, lhe permitem canalizar importantes recursos humanos e financeiros em prol do desenvolvimento agrícola do Hemisfério.

O Plano de Médio Prazo 1987-1991, documento normativo que assinala as prioridades do Instituto, enfatiza ações voltadas para a reativação do setor agropecuário como elemento central do crescimento econômico. Em vista disso, o Instituto atribui especial importância ao apoio e promoção de ações tendentes à modernização tecnológica do campo e ao fortalecimento dos processos de integração regional e sub-regional.

Para alcançar tais objetivos o IICA concentra suas atividades em cinco áreas fundamentais, a saber: Análise e Planejamento da Política Agrária; Geração e Transferência de Tecnologia; Organização e Administração para o Desenvolvimento Rural; Comercialização e Agroindústria, e Saúde Animal e Sanidade Vegetal.

Essas áreas de ação expressam, simultaneamente, as necessidades e prioridades determinadas pelos própios Estados membros e o âmbito de trabalho em que o IICA concentra seus esforços e sua capacidade técnica, tanto sob o ponto de vista de seus recursos humanos e financeiros, como de sua relação com outros organismos internacionais.

Esta publicação foi reproduzida, em agosto de 1989, numa tiragem de 100 exemplares.



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