Foreign Exploration of the Cereal Leaf Beetle (*Oulema melanopus*) Natural Enemies, Especially Egg Parasitoids, in China for Potential Exportation to the USA

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Summary of Accomplishment (<200 words)

This was the first year of a proposed three-year foreign exploration project to search for cereal leaf beetle natural enemies in China. The US team (Bai & Pike) traveled to China during May 23 to June 9 and collected extensively with Chinese collaborators in two northern provinces. The team traveled through five cities and established contacts in each for future cooperative works. The team also evaluated the climate, environment and crops there compared with the CLB habitat in the western US. The team surveyed fields of spring wheat, rice, corn, beans and some vegetable, and collected about 300 adults and 3000 eggs of *Oulema* species. After the US team left China, Chinese collaborators - coordinated by Dr. Wan of the Biocontrol Institute in Beijing, continued collections on millet and wheat. More than 1000 larvae and some eggs of *Oulema* were collected. Most of this material was reared in Dr. Wan’s lab in Beijing. Some larval parasitoids (identification not confirmed) were reared from CLB larvae from Xinjiang but no egg parasitoids were reared out. While in China, the US team observed a small parasitic fly ovipositing in eggs of CLB in Xinjiang. Effort will continue in 2003 to search for potential natural enemies with Chinese colleagues.
**Project Objectives** (to be carried out over a three-year period):
1. Conduct foreign exploration for CLB natural enemies, especially egg parasitoids, in China.
2. Receive and rear natural enemies in quarantine facilities at Washington State Univ. (WSU).
3. Assess the ecological and physiological host range of selected parasitoids in China, and of those imported, assess their acceptance of Northwest CLB.
4. Propagate large numbers of suitable parasitoids and release them against CLB in the western US.

**Achievement in 2002**

This is the first year of the proposed three-year foreign exploration project. We noted in the original proposal that the overall work may take 3 years or longer to achieve. During the first and second year, we will concentrate on collecting parasitoids from China; during the second and third year, on establishing parasitoids in quarantine for host specificity evaluation, and possibly subsequent field releases.

The foreign exploration included two components: 1) field visits and collections by Bai and Pike from the US in China and 2) collection of *Oulema* and parasitoids by Chinese collaborators.

1) **Fields visits and collections by the US team in China**

During May 23 to June 9, 2002, Drs. Bai from Oregon Department of Agriculture and Pike from Washington State University, traveled to China and met with Dr. Wan and his group at the Biological Control Institute of Chinese Academy of Agricultural Sciences in Beijing. After making necessary arrangements for various local areas, Dr. Wan’s graduate student (Mr. Shize Zhang) accompanied the US team to two northwestern areas in China for *Oulema* and parasitoid collections. The team spent the first week travelling to Yulin, a city in northern Shaanxi Province, to collect *Oulema tristis* and parasitoids. Yulin is about 700 km west of Beijing. The city sits near the latitude of 38ºN and longitude of 110ºE, which geographically resemble the western states of Utah, Wyoming and Colorado where cereal leaf beetle infestation occurs. Around the Yulin area, the general landscape is desert-like, dry, and the soil is sandy. Elevation is about 900 –1200 meters above sea level. This area is part of the ‘yellow soil’ high plateau in northern China. The agriculture is mainly based around the Yulin River valley, which runs from north to south through the city, and beyond.

The main crops grown in the area include spring wheat, millet, rice, corn, beans, potatoes and other vegetables. Apple orchards are also seen in the area. Our team spent about three days in the Yulin area and collected in fields of spring wheat, rice, corn, soybean, and in grasses along field edges and irrigation ditches. We also visited a commercial vegetable company where squash, cucumber, tomato and peppers were grown in a primitive green house. An apple orchard was visited where collection effort was directed mostly toward the ground cover grasses. At the time of these field visits, the temperature was sunny and warm ranging from 15 – 28ºC. The spring wheat was about 50-60 cm in height. Rice was recently planted, about 10-16 cm tall. Corn was about 20-30 cm in height.

The team traveled to fields about 30 km north of Yulin near the town of Jinjizhi to check spring wheat for CLB, and to fields about 60 km south of Yulin to collect in rice and corn fields (near the town of Zhenchuan). We also stopped in various places in between to search for the beetle,
especially in roadside grasses. Insect sweep netting was the main method of detection together with visual inspection.

We did not find any species or life stages of *Oulema* in any fields in the Yulin area. There were aphids in the wheat fields. Generally, almost all the fields were well farmed and well taken care off. Weeds were rare in any of the visited fields. Crops looked healthy and were well fertilized and irrigated. *Oulema tristis* was reported to occur on millet around Yulin, but our visit was too early as millet was not yet planted. Local contacts indicated *Oulema tristis* would appear in millet fields around late June to early July (see below for collection results by local Chinese colleagues at later dates).

During the second week, the team traveled to Altay, a city in northern Xinjiang Province, to collect *Oulema melanopus* and parasitoids. Altay is about 3000 km northwest of Beijing, and is probably one of the most northwestern cities in China bordering Kazakhstan, Russia and Mongolia. The city sits near the latitude of 48°N and longitude of 87°E, which geographically resemble the western states of Montana, Idaho and Washington. This is a mountainous region. The Altai Mountains run to the northeast side of the city of Altay along the borders of the four countries – China, Mongolia, Russia and Kazakhstan. The general landscape in the area is rugged mountain with desert in between. The Gobi Desert extends west from Mongolia to as far as this region. The soil is dry and sandy. Elevation is about 1200 –1400 meters where crops are grown. The agriculture is mainly along rivers where irrigation systems are in place.

The team rented a car in Altay and drove to Qinghe County which is about 230 km southeast of Altay near the Mongolia border (about 50 km away from the border). This is an area where large acreages of spring wheat are grown. The cereal leaf beetle has been documented in this area over the last several years. After six hours of driving on the primitive, trail type roads across parts of the Gobi desert, we reached Qinghe. The weather was warm, comfortable, breezy, about 11-27 °C. We collected in three sites around Qinghe County. One site was near Buhaba Village in Baixing, about 10 km north of Qinghe; elevation about 1340 m. Here, about 50 hectares of spring wheat (variety Ningchun 16) was grown in a river valley surrounded by foothills. Plants were about 15-20 cm tall, lush and green, well irrigated and fertilized. Field edges had some trees and shrubs, with riparian and barren mountain nearby. The general environment in the area resembled that of parts of Montana, Wyoming, Colorado, Utah, and eastern Oregon. The high and dry environment made agriculture possible only along rivers where irrigation was available. We found plenty of cereal leaf beetle adults, eggs and some early instar larvae at this site. Sweep net collections yielded about 10-16 adults per 120 sweeps. It was easy to find eggs on the leaves. In a few cases we saw up to 16 eggs on one leaf. Average egg density was about 3-5 eggs/leaf. Lower leaves had somewhat more eggs than upper leaves, probably because they were available for oviposition by CLB adults for a longer period of time than the new upper growth.

The second site in Qinghe was about 2 km away from the first. Here the plants were a little shorter, about 10-15 cm. The site consisted of about 67 hectares of spring wheat (variety Xinchun 8). A hill separated this field from the first site. Cereal leaf beetle adult densities were a little higher than the first site, averaging with 20-80 adults per 120 sweeps. Adults and eggs were the predominant life stages along with a few early instar larvae.

The third site in Qinghe was about 30 km south of the county seat, in the Saerzhaile Village of Arele,
elevation about 1260 m, crop spring wheat (variety Ningchun 16). A river, adjacent to the wheat, was used to flood irrigate the crop. Here, CLB densities were the highest encountered, about 74-90 beetles per 120 sweeps. Some farmers in neighboring fields had sprayed for the beetle.

From the three sites in Qinghe County, we collected about 3,000 cereal leaf beetle eggs and 300 adults. There were two forms of adults, possibly two different species of *Oulema* present in Qinghe. One form manifested the typical CLB coloration of an orange thorax and legs, while the other form was a uniform blue. Both forms occurred in the spring wheat field at the same time. The typically colored CLB was predominant comprising 80-90% of the collected beetles. We also found a small fly ovipositing into CLB eggs in the field. This was seen in two of the three sites visited. Several parasitic wasps (ichneumonids, braconids, chalcids, and others) were taken by sweep net sampling and preserved in alcohol. Some of these parasitoids may have come from the CLB. All of the CLB eggs were taken to Beijing to the Biological Control Institute, Chinese Academy of Agricultural Sciences and held for potential egg parasitoid development, emergence, and recovery.

2) **Collection of *Oulema* and parasitoids by Chinese collaborators**

The Oregon Department of Agriculture entered into a cooperative agreement with the Biological Control Institute of the Chinese Academy of Agricultural Sciences in Beijing. A portion of the NBCI fund was used to pay for Chinese collaborators to host, accompany, and assist the US team at the various field locations to collect *Oulema* and its potential parasitoids. The agreement also stipulated that the Chinese collaborators would make all necessary arrangements with local contacts, and collect more *Oulema* and potential parasitoids in these areas after the US team returned to the US.

Dr. Wan’s lab at the Biological Control Institute was our primary cooperator. The lab made all initial contacts and arrangements with local people in Shaanxi and Xinjiang. Dr. Wan’s graduate student – Mr. Shize Zhang, accompanied the US team for the entire trip. After the US team left China, Dr. Wan and his colleagues in China continued collections of *Oulema* in an attempt to find potential parasitoids.

In Shaanxi Province, the local contacts – Mr. Zhenrong Dan and Ms. Fang Yang, collected around Yulin and five counties in the area during June 23 though August 3. The local team visited fields at 15 different dates and collected in millet fields throughout the growing season in plants from 15-70+cm in height. In total, 14 adults, 10 eggs and 124 larvae were found. The eggs were held in a laboratory at the Yulin Agricultural School, but no parasitoids emerged. Adults and larvae were packaged and sent to Dr. Wan’s lab in Beijing. Unfortunately, many larvae died because it took four days for the package to reach Beijing. The *Oulema* found in Shaanxi Province is believed to be *O. tristis*. We will receive the adult specimen of *O. tristis* from Dr. Wan.

In Xinjiang province, the local contact – Ms. Jingfei Yang, collected in four sites during two separate trips between June 26 and July 4. Three counties were visited including Qinghe, Fuyun and Hongdun in Altay area. All collections were made in fields of spring wheat. One site where the US team collected, was revisited in Qinghe County (Arele town). No eggs were found but about 1500 cereal leaf beetle larvae were collected, mostly from Qinghe County. About 2/3 of the collected larvae were sent to Dr. Wan’s lab in Beijing and the rest were reared locally by Ms. Yang. The material reared in Beijing yielded no parasitoids, but of larvae held in Altay, two were parasitized. After four days, seven white parasitoid larvae came loose from the broken skin of the two parasitized CLB larvae.
Whether these parasitoids belong to Diptera or Hymenoptera is yet to be determined. The biology of the parasitoids indicates that they are likely Hymenoptera.

Discussion of actual accomplishment, compared to the objectives established for the project during the 12-month funding period

Work to be accomplished in the 12-month funding period:

**China:** 1. Conduct an in-depth literature review of dominant egg/larval parasitoid species of *Oulema* and their distribution in China.

This was achieved by the Chinese collaborators. Their report is on file with the principal investigator. The species and distribution of Chinese *Oulema* and their parasitoids, and the up-to-date literature citation are compiled in the report. Seven species of *Oulema* including the cereal leaf beetle (*Oulema melanopus*) were recorded along with more than 25 species of natural enemies in China.

2. Survey and collect *Oulema* species and their parasitoids on different crops in regions with similar climate and latitude to the western US, e.g., northern high plateau – Xinjiang, Shaanxi, or Beijing (crops: wheat, barley, millet or broomcorn).

Survey and collections were made in Xinjiang and Shaanxi on wheat, millet, corn, rice etc. See the specific achievement session above.

3. Identify *Oulema* species collected on different crops from different provinces.

The *Oulema* species collected in Shaanxi was identified as *Oulema tristis*. Those collected in Xinjiang were identified as *Oulema melanopus*. Another questionable *Oulema* species from Xinjiang is being identified. All specimens were identified by a leaf beetle specialist, Dr. Hongbin Liang of the Institute of Zoology, Chinese Academy of Sciences, Beijing.

4. Collect, then rear parasitized *Oulema* eggs and larvae for natural enemies in the laboratory for exportation to USA.

Collections were carried out in Shaanxi and Xinjiang provinces. Rearing was done mostly in Dr. Wan’s lab in Beijing. However, some preliminary rearing of eggs or larvae was also conducted in Shaanxi and Xinjiang (see above). Shipping collected material from Shaanxi or Xinjiang to Beijing took about four days. The transport period or temperatures were unfavorable for the *Oulema* and led to high mortality. The improvement next year will be either increasing the shipping speed (e.g., use an overnight delivery service) or doing more local rearing.

5. Develop rearing methods for selected potential biocontrol agents and maintain colonies in the laboratory.

No progress was made on this because of the scarcity of parasitoids in the first year.

6. Help to collect and ship *Oulema* to USA for genetic studies to identify differences among global
geographical strains, biotypes of *O. melanopa*.

This was accomplished while the US team was visiting China. We brought back over three hundred beetles of *Oulema melanopus* from Xinjiang. Two other species: *Oulema tristis* and *Oulema oryzae* will be shipped to the US from Dr. Wan’s lab.

**USA. 1.** Bai and Pike will travel to China at the end of May for two weeks to join Wan and his crew to collect CLB and its parasitoids extensively in the field, and to make comparative evaluations of the climate and environment with that of CLB habitat in the western US.

Bai and Pike traveled to China and visited with Dr. Wan’s group in Beijing and then collected together with Wan’s crew for over two weeks in China. The team traveled to Beijing, Shaanxi and Xinjiang during May 23 to June 9. We covered a large area in northern China. The farthest distance was between Beijing and Altay (close to 3000 km). In each place, we took notes about the climate, geography, elevation, landscape and crops grown. We found that the climate and environment were similar between northern China and the western US. We felt that any parasitoids found in Shaanxi or Xinjiang would likely survive and do well in the western US where CLB is a problem. In every city and area visited in China, we established local contacts and laid a foundation for future cooperative work.

2. **WSU quarantine facilities receive, quarantine, and rear shipped material from China.**

No progress was made during the first year because: 1) no import permit was issued by USDA and 2) not enough parasitoids were obtained from China.

3. **WSU quarantine facilities maintain parasitoid colonies and conduct host specificity testing.**

No progress was made during the first year (see the reasons above).