

# Termite gut bacteria as "Trojan Horses" — a new approach to termite control

*Dr. Claudia Husseneder  
(Louisiana State University)  
and*

*Dr. J. Kenneth Grace  
(University of Hawaii)*

It struck us that *Clean Asia* Magazine readers might be interested in learning how researchers go about developing brand new pest control methods and products. Not just new twists on old ideas, or minor improvements, but really new approaches that have not been tried before. So, here is a view into some new developments in our laboratories.

According to Greek mythology, the Trojan Horse was used to secretly shuttle enemy soldiers into the city of Troy and thus bring the city and its people to defeat. Taking this same approach to insect control, we are working with genetically engineered microorganisms as "Trojan horses" to deliver foreign genes into an insect host or an insect population. In social insects like termites, these microbial shuttle systems really appear to be promising tools for achieving colony suppression and elimination.

Subterranean termites depend on microbial symbionts (partners in their gut) for wood-digestion and for survival. Each worker termite has a stable microbial community in its gut (or intestinal tract), which is naturally exchanged between colony members through social interactions, such as grooming and sharing food. Therefore, we felt that natural gut microbes would be excellent tools for termite control.

The goal with our microbial shuttles would be to introduce and spread microbes expressing detrimental genes in a termite colony. As a first step



*Dr. Kenneth Grace*

towards this goal, we developed a functional shuttle system using genetically engineered termite gut bacteria to deliver and express marker genes in the termite gut.

We used Green Fluorescent Protein (GFP), which was originally obtained from jelly fish and causes bacteria to glow under ultraviolet (UV) light, as a marker gene. We were able to monitor ingestion of the transformed bacteria by termites, determine their long-term stability in the termite gut and trace the path and speed of bacterial transfer between termites.

We demonstrated ingestion and long-term stability of our modified bacteria in termites by feeding workers from three different colonies with GFP bacteria on filter paper for a



*Dr. Claudia Husseneder*

day. Then, we pulled the guts of a subsample of termites each day, cultured the gut contents and screened for GFP bacteria under UV light. To monitor bacterial transfer between termites, we dyed the donor termites or the recipients, we fed the donors with GFP bacteria, combined them with the recipients (fed with water only), and screened the guts of the recipients each day for GFP bacteria.

Workers ingested genetically modified bacteria rapidly by feeding on the treated filter paper. After just one day of feeding, almost all termites contained GFP bacteria. The bacteria established a stable population in the termite gut for several months.

Workers transferred bacteria efficiently throughout the

laboratory colonies. One day after combining donors with recipients, over half of the recipients had GFP bacteria in their guts. After one week, this proportion reached 80-100 per cent. Effective transfer and spread of GFP bacteria among workers occurred even with a ratio as low as one donor per 25 recipients. In addition, we found that not only termite workers, but also soldiers receive and transfer bacteria.

So, we have created a functional shuttle system for delivering, spreading and expressing genes throughout a termite colony. At this point, we used this shuttle system only as a monitoring system. The goal in developing a "Trojan horse," however, is to find a gene product that is actually detrimental to termites. In collaboration with scientists from Veterinary Sciences and Entomology at Louisiana State University, we identified gene products (proteins) that destroy the three species of protozoa on which the termites depend for wood-digestion.

We are now in the process of measuring the activity of these protozoa-killing proteins by injecting them directly into the termite hindguts. Next, we will transform bacteria with gene constructs expressing the most efficient termite-killing proteins. To reduce the risk of environmental contamination, we will use specific bacteria that are not able to survive outside the termite gut as our shuttles. Ultimately, we hope to develop a self-replicating, self-perpetuating product that will kill termites rapidly and be cost effective and target specific at the same time. Thus, our "Trojan horse" will soon be approaching the gates of the termites' city of Troy!



*Next step in study is to develop proteins that will be detrimental to termites' ability to digest wood and administered by a 'Trojan Horse'. Photo courtesy of University of Hawaii.*