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## VIRUS-LIKE SYMPTOMS IN A TERMITE (ISOPTERA: KALOTERMITIDAE) FIELD COLONY

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Researchers in the field of termite biological control have tried for decades to isolate and formulate microbial agents that could spread within termite (Isoptera) groups so as to create an epizootic event and achieve successful biological control (Culliney & Grace 2000). The incentive to develop a so-called “environmentally friendly” technology as an alternative to chemical treatment resulted into unrealistic optimism and despite all the promising laboratory tests, no termite biological control technology was ever achieved (Chouvenc et al. 2011). Although the original idea was to use self-replicating entomopathogens that could be spread by social contact among individuals of the colony, recent studies have shown the importance of defense mechanisms within termite colonies that reduce the chance for entomopathogens from spreading throughout the nest (Chouvenc et al. 2008; Chouvenc & Su 2010; Hamilton et al. 2011). Because termites can prevent pathogens from completing their life cycle within the group, it was suggested that the traditional concept of an epizootic (Steinhaus 1949) could not be applied to termites (Chouvenc & Su 2012).

Paradoxically, while this field of study was based on the assumption that an epizootic could naturally occur in termites (Schmid-Hempel 1998), the occurrence of disease in termite groups was only observed under laboratory conditions in highly artificial conditions or in the field using inundative control methods, which bypass the need for an epizootic (Chouvenc et al. 2011). Therefore, to our knowledge, there are no official reports about the occurrence of any natural epizootic event in a termite colony under field conditions. The absence of such observations may be explained by the cryptic life style of termites, as termite colonies are usually contained within a closed gallery system, and it can be difficult, if not impossible, for any observer to have access to the colony at the precise moment it is undergoing an epizootic event. Alternatively, natural epizootics in termites may be so rare that they simply never have been observed. The fact is, no natural epizootic in a termite colony has ever been reported.

On 11 Jun 2011, while prospecting for termite samples in forested areas of Broward County (Florida, USA), we encountered a small colony of *Neotermes jouteli* (Banks) (Kalotermitidae)

within a single branch of a small dead tree (unidentified). All individuals ( $\approx 30$ ) were found dead within the gallery but their level of decomposition was not advanced, suggesting that all individuals died in a short time frame within the past 24 h or so. Most individuals showed a yellow milky color, and the head capsule and parts of the thorax were unusually dark brown or black (Fig. 1). At the time of collection, the rainy season had just started, with severe lightning storms, a characteristic of Florida summer weather (Hodanish et al. 1997). Hence, we first hypothesized that the tree or its surrounding may have been hit by a lightning, killing the termite colony in the process, but no obvious signs of such event (burn marks) were found in the area. Secondly, because some termite cadavers were showing signs of bacterial growth, we hypothesized that the colony may have succumbed to a bacterial epizootic. We isolated 4 strains of bacteria (Gram-rods, Gram + *Coccolobacillus*, Gram + rods, Gram + *Bacillus*, identification according to Holt et al. 1994) from the cadavers, using a sterile cotton swab streaked on 1/5<sup>th</sup> strength potato dextrose agar. Pure isolates were cultured in potato dextrose broth. In absence of laboratory colonies of *N. jouteli*, we exposed groups of 20 termites (*Coptotermes formosanus* Shiraki) in a 45-mm petri dish with a wet cellulose pad on the bottom to a range of concentrations of the microorganisms ( $10^6$  to  $10^9$  cells applied on the cellulose pad), and termite groups were maintained for 15 d in laboratory conditions. None of the groups exposed to the bacteria showed significant mortality when compared to control groups (Cox proportional hazard regression analysis), and out of the few individuals that died within the time frame, none showed signs of their head capsule turning black. We suggest that all 4 bacteria were present in the termites' body as saprophytes as previously observed (Chouvenc et al. 2012). Although our bioassay was performed on *C. formosanus* only and a species-specific virulence cannot be fully excluded, it is unlikely that such bacteria were responsible for the sudden death of the *N. jouteli* colony.

As we failed to show a case of bacterial epizootic within termite groups, and because of the apparent absence of mycosis in any of the cadavers, we did not pursue the study further.

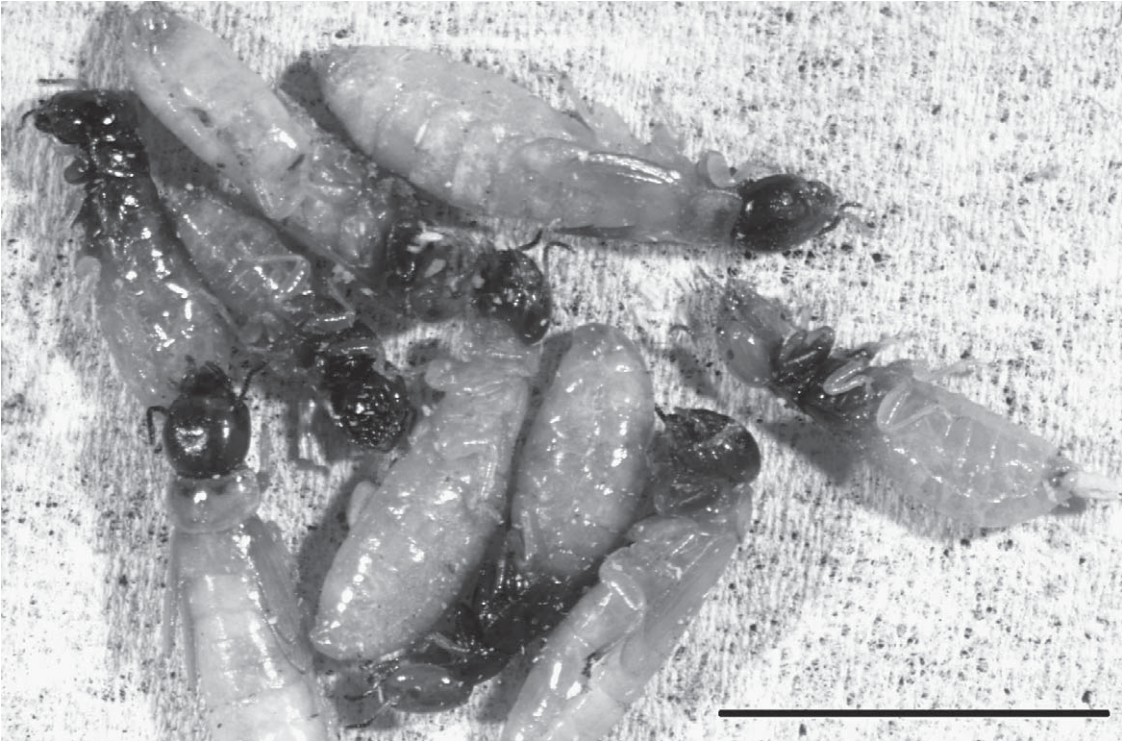


Fig. 1. *Neotermes jouteli* cadavers showing a dark-head syndrome. All individuals ( $\approx 30$ ) from a single field colony were found dead, displaying this syndrome. Scale bar = 8 mm.

It is only recently that we considered that the termites may have been infected by a viral agent. Levin et al. (1993) described the symptoms of *Reticulitermes flavipes* (Kollar) exposed to an entomopox virus, which was previously isolated from a grasshopper (*Melanoplus sanguinipes* Stål; Orthoptera: Acrididae) in laboratory conditions. The photograph and the description provided by Levin et al. (1993) match our own observations and suggest that the colony of *N. jouteli* we discovered may have been killed by a viral epizootic. Unfortunately, and to our biggest disappointment, we realized that the cadaver samples had not been stored properly, which prevented us from testing for the presence of such virus in our samples, and ultimately prove this hypothesis. Although we could not prove that the death of the *Neotermes* colony was caused by a viral agent, the symptoms strongly indicated the presence of a virus in these samples. The purpose of this study was, first, to document the occurrence of a virus-like symptom in termites under field conditions, as such observation was previously never reported, and, second, to provide an incentive for future research if such symptoms are observed again to test for the presence of viruses in termites.

#### SUMMARY

The concept behind the use of biological control agents against termites was based on the assumption that epizootic events can occur naturally in a termite colony. However, no reports ever mentioned the occurrence of epizootic in natural field conditions in any termite species. We here describe what could be the first report of a viral epizootic in a field colony of *Neotermes jouteli*.

Key Words: bacteria, entomopathogen, Koch's postulates, *Melanoplus sanguinipes*, *Reticulitermes flavipes*

#### RESUMEN

El concepto detrás de la utilización de agentes de control biológico contra las termitas se basa en la suposición de que eventos de epizooticas pueden ocurrir de forma natural en una colonia de termitas. Sin embargo, no se conocen informes que hayan mencionado la aparición de epizooticas en condiciones naturales de campo en especies de termitas. Se describen lo que podría ser el primer informe de una epizootia viral en el campo de una colonia de *Neotermes jouteli*.

Palabras Clave: bacterias, entomopatógenos, postulados de Koch, *Melanoplus sanguinipes*, *Reticulitermes flavipes*

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