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Theronts of *Ichthyophthirius multifiliis* find their Fish Hosts with Complex Behavior Patterns and in Response to Different Chemical Signals

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How parasites find and recognize fish hosts is best known for trematode cercariae. These multicellular organisms respond with complex behavior patterns to sequences of different chemical host signals, although these invasive stages are produced in large numbers. However, little is known on the host-finding of protozoan parasites. We therefore studied how the infective theronts of *I. multifiliis* find their fish hosts by offering them various fractions and chemical modifications of fish skin surface.

Free swimming theronts showed at least 6 different behavior patterns, 4 of them were stimulated by fish skin components, e.g. by amino acids. Theronts approached towards solid substrates ("stop and go ahead") in response to macromolecular glycoproteins of fish skin, and they were repelled ("stop and turn away") by other macromolecular glycoconjugates. After contact with solid surfaces, they repeatedly dipped on the surface ("dipping contact") in response to glycoconjugates of fish skin and they swam along the surface ("body parallel") when it contained proteins of fish skin. The results demonstrate that also a protozoan fish-parasite without nervous system and cellular chemoreceptors may show very diverse host-finding behavior patterns (at least 11) and may respond to different chemical host cues (at least 5).

Key Words : *Ichthyophthirius multifiliis*, Theront, Host-finding, Fish-host, Behavior

INTRODUCTION

Many fish-parasites infect their hosts via actively penetrating larval stages. The invasive stages are normally produced in very large numbers and the question is, whether they must specifically recognize the fish-hosts for successful transmission. Most data on the finding and recognition of fish-hosts are available for trematode cercariae [reviews: 2-4]. These multicellular organisms show sequences of complex behavior patterns when approaching and invading their hosts and they respond in each of their behavior patterns to independent host cues such as water turbulence, shadow stimuli and in particular very different chemical signals. This sophisticated host-recognition seems to play a key role in the transmission of these fish-parasites despite the mass production of the invasive cercariae. In this context it was interesting how protozoan fish-parasites without cellular receptors and nervous systems find and invade their hosts. Therefore we studied the host-finding and host-recognition of the invasive stages (theronts) of the ciliate *Ichthyophthirius multifiliis*, one of the most pathogenic protozoan parasites of fish [1, 7]. Much is known on the behavior of non-parasitic ciliates, in particular on *paramecium* and *Tetrahymena* species. They show few swimming movement patterns such as forward and backward swimming with various speeds and direction changes and the swimming patterns are correlated with changes in the membrane potentials [8]. Would the *I. multifiliis* theronts rely on such simple behavior patterns when they search their fish hosts ?

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RESULTS

We studied the behavior of *I. multifiliis* theronts when they encounter fish substrates. As substrates we offered living fish, dead fish, but also fish skin surface extracts and their isolated and chemically modified compounds integrated in agar or presented via membrane filters. The theronts showed a surprising diversity of responses during swimming, in particular when they approached the substrates and after they had encountered the substrates (Fig. 1).

(1) Free swimming theronts responded in 4 of their behavior patterns to fish skin surface compounds: "Burst" responses were stimulated by unknown factors in the presence of living fish, but not of dead fish, "wobbling" by components of fish (not frog and snail) skin, M_r< and > 1 kDa. The responses "stop and orientation" and "change of direction" were stimulated mainly by amino acids of the fish skin surface and in addition by unknown macromolecular fish skin compounds.

(2) When the theronts approached toward the fish substrates they orientated in direction to the fish host by responding with "stop and go ahead" to macromolecular glycoproteins of the fish skin surface. However, they obviously also were repelled by compounds of the fish skin surface : glycoconjugates of fish skin surface stimulated the behavior "stop and turn away" .

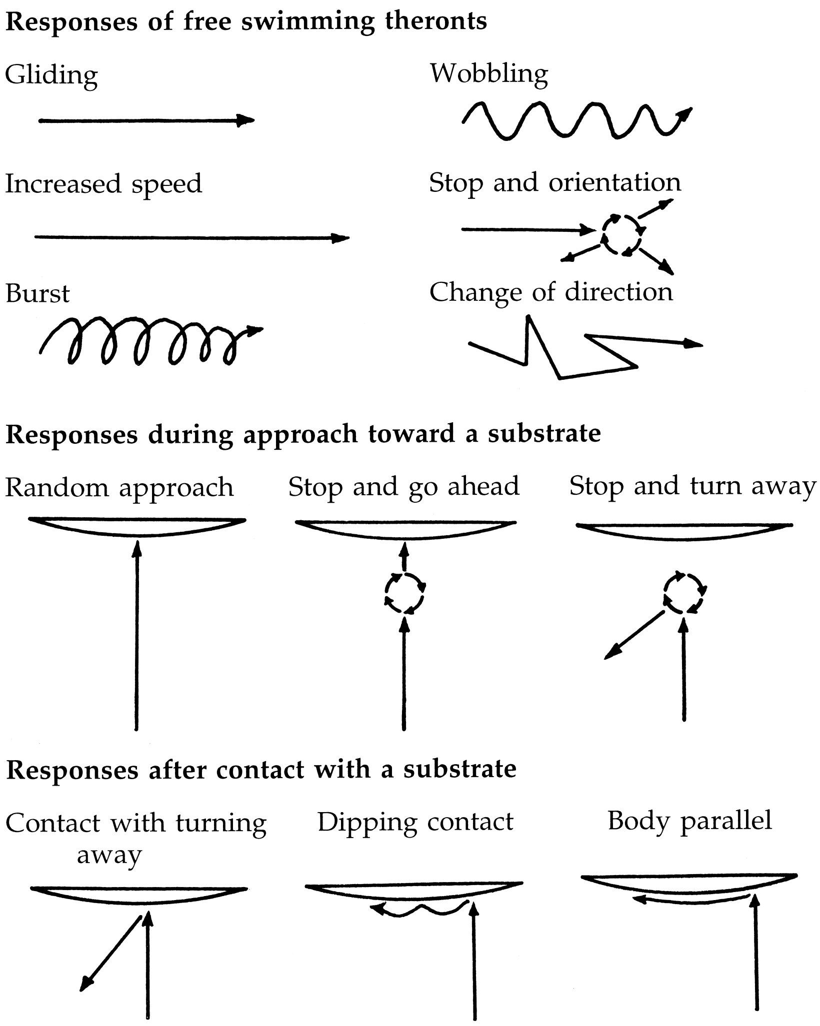

Fig. 1 Types of swimming behavior of the invasive stages (theronts) of the ciliate fish-parasite *Ichthyophthirius multifiliis*.

(3) After contact with the substrate's surface, the theronts obviously recognized the fish hosts. They responded with "dipping contact" to certain glycoconjugates of the fish skin surface extracts and with "body parallel" to macromolecular fish skin surface proteins.

DISCUSSION

Observations of Lom and Cercasovova [5] suggest, that theronts of *I. multifiliis* do not orientate towards their fish hosts over long distances and the concentration of theronts in water around living fish may even be lower than in control water samples [9]. Also fish invading cercariae seem not to be capable for a long-range chemo-orientation towards their hosts [2-4]. In fact, it is not probable that a stable chemical concentration gradient will establish over long distances around fish as fast-moving hosts. However, fish parasites such as trematode cercariae increase the probability of encounters with the fish host by responding to signals in the close vicinity of the fish. This seems to happen also in *I. multifiliis* theronts. The free swimming larvae responded with at least 4 of their 6 swimming behaviors to fish skin surface components. The movement patterns of the responses may considerably increase the chances for a contact with a nearby fish. During their approach toward a substrate the organisms obviously can decide, whether they will contact the subject ("stop and go ahead") or whether a contact should be avoided ("stop and turn away"). After contact with substrates the theronts responded with two behavior patterns ("dipping contact" and "body parallel") which resemble those seen in the ciliated trematode miracidia [6] encountering their snail-hosts, and which obviously serve host-identification.

The parasites showed at least 11 different movement patterns and they responded to at least 4 different fish-emitted chemical signals. The swimming behavior of *I. multifiliis* theronts is obviously more diverse and more complex than that described for free living ciliates. These results support the view, that actively fish invading parasites need complex host-finding and host-recognition mechanisms despite their mass production.

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