

Crayfish plaque – Aphanomyces astaci A widespread threat to German crayfish populations

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Introduction + Objectives

Crayfish plague is one of the most important diseases of crayfish and is widespread worldwide. The causative pathogen Aphanomyces (A.) astaci belongs to the oomycetes. These filamentous, fungus-like eukaryotes are classified within the class Stramenopiles with about 600 species.

In the mid-19th century A. astaci was introduced from North America to Europe with North American crayfish species such as Pacifastacus leniusculus, Orconectes *limosus* or *Procambarus clarkii*. Although these species can be infected, they rarely show clinical symptoms. However, resistant crayfish are carriers and responsible for further pathogen spread. Spores can also be disseminated via inanimate vectors. A. astaci reproduces asexually in the aquatic environment. Outside the host they survive for up to 14 days. Highly susceptible native crustaceans such as Astacus astacus, Austropotamobius torrentium or Astacus leptodactylus are encountered through filament penetration of the cuticle and deeper organs. Susceptible crayfish develop clinical symptoms after an incubation period of about 10 days with high mortality rates. In the event of mortality or as part of preventive measures in crayfish populations, samples from various German federal states are regularly sent to the Food and Veterinary Institute Braunschweig/Hannover, Germany, for examination. In addition, a project analysing the pathogen's spread in Lower Saxony, Germany, local waters has been initiated.

Why are European Crayfish harmed by this pathogen?

The reason is a different immune response compared to resistant species. Innate immune mechanisms are initiated by the prophenol oxidase activating system (proPO-AS) which leads to an activation of the enzyme prophenole oxidase (ProPO). The result of this reaction is **melanisation**. For activation of this proteolytic cascade, the recognition of the pathogen is necessary. If activated, the proPO-system cascade leads to "melanisation, parasite enclosure and production of cytotoxic and lytic" substances (Martínez-Ríos et al. 2023).

Resistant species have adapted to the presence of this pathogen and managed to establish a "balance between host defence and parasite attack" which is never established by susceptible species like Astacus astacus, probably due to the lack of exposure before introduction of the parasite (Cerenius et al. 2003).





Fig. 1 Sampling from a *A. astaci* positive Austropotamobius torrentium Source: Dr. Dirk Willem Kleingeld, LAVES

Fig. 2 Light microscopic micrograph of hyphae-like filaments of *A. astaci* on the cuticle of a crayfish; bar: 100 µm.





Fig. 3 Evaluation of a real-time PCR for the detection of *A. astaci;* TaqMan PCR with a minor groove binder probe is applied, targeting a 59-bp sequence of ITS1 from the nuclear ribosomal gene cluster (Vralstad et al. 2009); Green: Negative control; Red: Positive control; Grey and blue: samples of a crayfish tested positive





Fig. 4 A+B A. astaci infected crayfish with typical signs of disease; Source: Bavarian Environment Agency Healthy *Pacifastacus leniusculus* Source: Dr. Dirk Willem Kleingeld, LAVES Healthy Astacus astacus; Source: Dr. Dirk Willem Kleingeld, LAVES

Results

A. astaci has been recently detected several times in crayfish taken from waters located in Lower Saxony as well as other German federal states. In the last 10 years we have examined about 250 German samples of different crayfish species. Affected federal states repeatedly initiated disease control measures due to mortality events in crayfish caused by the crayfish plague (For instance: Alatsee, Bavaria, Germany).

Conclusions

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Fig. 5 German Federal States with positive detection of *Aphanomyces astaci* by LAVES laboratory over the last 10 years in different crayfish species; Green: open freshwater habitats; Red: ornamental husbandry; (n): number of detections; Map created with TSN Kartenexplorer

References

- The crayfish plague is spread through invasive species and poses an enormous ecological threat for indigenous crayfish.
- Regular detections of the pathogen A. astaci in both susceptible and vector species confirm its widespread distribution in Germany. Numerous European countries are fighting the crayfish plague as well.
- To prevent further disease spreading, biosecurity and disease control measures are necessary as well as the collection of epidemiological data to document the status quo. Furthermore, stocking with disease-free animals is recommended. In this regard, national and pan-European cooperation is of great importance.
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