

BSF Pathogens and Pests

Benedict Ling, Entomology Research Assistant

Potential pathogens and pests are abundant in our surroundings – their intrusion into your Black Soldier Fly (BSF) farm may just spell trouble for your entire colony. Learn more about these pathogens and how to best keep them away from your insect colonies!

Dipteran Fly and BSF Immunology

Just like how humans rely on white blood cells and antibodies to help us combat against illnesses and diseases, dipteran flies too have their own immune system to ward off any ill-intentioned pathogens looking to proliferate at the insects' expense. The first line of defense for insects would be their outer exoskeleton, which serves as a physical barrier to any larger pathogenic threats. Should any get past the outer shell of the insect, they would then have to deal with bodily responses on the cellular and humoral (body fluids) level.

The premise for immunology in insects is somewhat like those in humans – genes that code for antimicrobial peptides (AMPs) are activated by the presence of pathogen-associated molecules, recognized by receptors in the host cell. As a result, AMPs are synthesized and released into the hemolymph (body fluids in insects that function like blood). These AMPs then work to kill off the pathogens synergistically (Takov et al., 2020).

In the case of BSFs, which also fall under the Dipteran order, they are known to have a wide spectrum of AMPs, credited to the high bacterial loads present in their diets. By feeding larvae with protein-supplemented diets, the variety and levels of AMP expressed will be at its highest as well. This in turn results in the BSFs' ability to inhibit the activities of a wide variety of bacteria and microorganisms (Vogel et al., 2018).

Pathogens

Pathogens are organisms that cause and proliferate diseases upon entry to the body. We might already be familiar with the bacteria and viruses that leave us ill with flu and fever, but pathogens that typically cause diseases in insects also include organisms like fungi and protozoans. Since the host range for some of these pathogens are rather narrow and specific, they have also been commercialised as insecticides for pest control in farms.

One piece of good news for BSF farmers would be that these insects are known to be highly resistant against pathogenic infections and are currently not known to be victims of any specific diseases which may manifest in other insects that are currently being produced for food or feed. However, we could also attribute this to the fact that BSF entomopathogens are largely understudied, given that the BSF industry is a relative newcomer to the insect farming and production scene. As such, it is important to note of the pathogens that afflict their close relatives in the dipteran family with deadly diseases and glean some insights on how these pathogens may one day figure out the befuddling puzzle that is the BSF immune system.

Fungi set themselves apart from the other insect pathogens – their infection route starts with their active penetration through the outer exoskeleton and cuticle of the insect body, as compared to oral ingestion (Federici, 2009). The aptly named Entomophthorales group of fungi, which specifically targets insect species for nutrient intake and reproduction, are known

to be a major threat to many dipteran fly species. Their spores, dispersed by fruiting bodies which emerge from the carcasses of their unfortunate insect victims, land on the exoskeleton of insects before germinating, allowing their emerging hyphae structures to penetrate into their bodies (Joosten et al., 2020). In the past, widespread epidemics caused by Entomophthorales species have hit production facilities which rear *Musca domestica* (houseflies), and there had also been records of successful transmission of these fungi to other dipteran fly species (Eilenberg et al., 2015). Thus, this is a cause of concern for BSF facilities as any infected dipteran fly originating from outside of the facility could potentially transmit these pathogens to BSFs.

Viruses, on the other hand, are obligate intracellular parasites which can only reproduce with the help of organelles present in living cells. Their host range tends to be larger than that of fungi, and viral transmission tends to happen via oral ingestion of viral particles found in the excretion of afflicted individuals, or through carcasses (Joosten et al., 2020). Various viral infections have occurred in dipterans like tsetse flies and houseflies, affecting the reproduction and biological fitness of their respective fly colonies (Joosten et al., 2020). Given that BSFs and other closely related dipterans are saprophagous in nature and consume decomposing matter for nutrients, viral pathogens could potentially become a problem in production facilities if feed sources are not managed properly as well.

Protozoans are free-living single-celled organisms. Unlike bacteria, protozoans possess a nucleus and largely resemble the cells found in animals and plants. Nevertheless, they too can transmit diseases like their bacterial counterparts. Microsporidia are protozoans that are widespread, and they are responsible for most of the diseases in insects caused by protozoa (Federici, 2009). Infection also tends to occur via oral ingestion when spores latch on to the insect's midgut and germinate. The protozoa then make their way into the insect's cell via a filament, before undergoing multiple cycles of cell division and subsequent proliferation (Federici, 2009). While these pathogens do not usually cause widespread mortality in insects, they are known to cause chronic infections in dipteran populations, resulting in stunted development, behavioural abnormalities, and reduced biological and reproductive fitness (Joosten et al., 2020)– all which spell trouble for insect farms focused on production.

Bacteria are single-celled organisms that proliferate rapidly due to their ability to thrive by feeding on a plethora of substrates. Given that insects also feed on the very same substrate, it is of no surprise that most entomopathogenic bacteria infect their insect hosts through oral ingestion. The most widely studied entomopathogenic bacteria is *Bacillus thuringiensis*, otherwise known as Bt. These bacteria have a wide variety of strains that are naturally-occurring and traces of them can easily be found in soil, leaf litter, surfaces of leaves, and even insect faeces (Federici, 2009). These bacterial strains contain endotoxins that when activated upon ingestion, will work its way through the intestines by destroying midgut cells, entering the body, and proliferating – this kills infected insects in a matter of days. Due to its effectiveness and insect host specificity, many strains of Bt are being used as environmentally friendly organic insecticides in the agricultural domain. One strain in particular, *B. thuringiensis israelensis*, is known to be toxic to dipteran larvae (Joosten et al., 2020).

Through DNA sequencing, the range of bacteria associated with BSFs have also been uncovered, and the list includes Enterobacteriales strains which are potentially pathogenic to humans (Zheng et al., 2013). As such, this might also be a cause of concern for food safety in the case where BSF larvae are used as feedstock for other animals in agri and aquaculture.

However, this health risk to humans can be prevented through the cooking process of the food, as these bacterial strains are relatively heat-sensitive (Baylis, 2006).

Pests

Insect pests like roaches and houseflies are a menace to us humans when it comes to health and safety, just like how aphids and whiteflies are detrimental to agricultural crops. Likewise, BSF production facilities also have their own set of insect pest woes.

These insect pests invade BSF colonies and affect production yields through competition, predation, and parasitism. Entire colonies can be thrown into disarray should they be allowed to reproduce and manifest in production facilities. One known mite species by the name of *Caloglyphus berlesei* is commonly known to be a predator of nematodes (otherwise known as roundworms). These parasites have been found to infest BSF larval colonies that had less than ideal conditions for development (Reguzzi et al., 2021). Meanwhile, parasitoid wasps tend to invade colonies by laying their eggs within the insect host, which then hatch and feed on the host, killing them. Infestations by the parasitoid wasp *Dirhinus giffardi* had been recorded in BSFs over in West Africa, whereby non-emerged pupae were found to have these parasitoids within them, resulting in only a 26.9% emergence rate (Devic & Maquart, 2015). Infestations by fellow dipteran species under the Phoridae and Muscidae family can also occur due to the use of organic food waste and strong-smelling attractants in facilities, which draw them in. While these flies are not predatory, they compete with the BSF larvae for resources like food and space. Eventually, this translates to less-than-desirable growth rates and development. Infestations from common indoor pests like the Pharaoh ant (*Monomorium pharonis*) can also cause serious damage to BSF colonies, as they have been observed to prey on eggs and newly-hatched neonates (Reguzzi et al., 2021). The vertebrate predators that we are all familiar with, in the form of spiders, mice and house geckos, are also potential predators.

Discussion and Preventive Measures

From what we have gathered so far, two main media are effective at transmitting diseases and pests: insect feed, as well as insects outside of the production facility. Existing insect pests and fungal spores could already be manifesting within the feed when they arrive at the facility, and the insect pests themselves could have already been afflicted by infectious pathogens from viral, bacterial and protozoic sources, giving them the potential to spread them throughout the facility.

Manure and carcasses, as mentioned, is a direct source of viral and bacterial pathogens, especially if the excretion or carcasses belong to infected individuals. If these are used as insect feed, the risk of feed contamination is high. Even if alternative feed sources like organic food waste are used, we cannot rule out the presence of insects or fungal spores, and measures must be taken to ensure that these do not manifest in the facility. A one-size-fits-all approach would be to heat the raw feed materials upon arrival at the facility to kill off any traces of fungi, bacteria and insects. However, heating dry feed materials itself is highly energy intensive given a large volume of feed. Alternative methods will also include further grinding or milling, which mechanically rids of potential insect pests (Joosten et al., 2020).

In terms of the facility, it is key to prevent as many mites and pests from getting within grasp of the colony or feed by putting up as many barriers to entry as possible. An example would be to install multiple layers of physical barriers to seal off any rooms that may contain the

colonies or feed, for example using doors that are well-sealed using rubber flaps. It is also essential to ensure that internal conditions are kept sterile, such that they do not promote unwanted bacterial or fungal growth. This is relevant in terms of air temperature and humidity – while insects like BSFs require high humidity to thrive at different life stages, high humidity also encourages fungal growth. As such, employees will need to be vigilant and do visual checks for signs of fungal growth within the feedstock and colony on a regular basis. Employees and workers should also be mindful of what they bring into the facility – whether they are consciously aware of it or not. Food items brought in may attract unwanted pests, so those should strictly be prohibited in the facility. However, potential pests and pathogens can also tag along on clothing – these include bacteria like *Staphylococcus aureus*, commonly found on human skin, which could be transmitted to the insects (Veldkamp et al., 2021). As such, proper personal protective equipment (PPE) like coveralls, hair nets and masks are also important to prevent transmission.

Finally, the brood stock of insects obtained by the facility should come from a pathogen-free source (Joosten et al., 2020), as unverified or wild BSFs might already have been afflicted by pathogens that could be passed on to subsequent generations or end up infecting other existing colonies that are already within the facility. It is also advisable to introduce genetic diversity by having insects from a variety of such pathogen-free sources, as this makes the colony more resistant to the spread of diseases (King & Lively, 2012).

Conclusion

Today there is a shortage of information with regards to pathogens that afflict BSFs. Thus, from the study of other closely related dipteran flies and insects reared in production facilities, we can glean some information about how these pathogens and pests could potentially be detrimental to BSFs and take preventive measures accordingly. It is therefore key to ensure that the barriers to entry are sufficiently secure, be it in the form of physical barriers and PPE or preventive processes such as feed cooking. With the growth of the insect protein industry, more studies will be done on BSF immunology and disease prevention in the future, and perhaps more specialized measures can be taken to ensure that larvae and flies remain in the pink of health.

Similarly, pests can reduce the production output of insect products, be it through outcompeting the BSFs for food and other resources, or by means of BSF predation. It is thus important to ensure that conditions within the facility remain clean and sterile to prevent the infestation of such pests.

By taking the joint implementation of these preventive measures into account, we as BSF farms can minimize external risks as much as possible. Ultimately, these form the traits of a well-run farm, as unknown factors such as BSF pathogens can be left out of the equation. These, in combination with new studies of BSF immunology and health, the occurrence of disease and pests in insect facilities can be further reduced.

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