Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Microbiology

Are you my symbiont? Microbial polymorphic toxins and antimicrobial compounds as honest signals of beneficial symbiotic defensive traits

Kai Hillman and Heidi Goodrich-Blair

In defensive symbioses where microbes benefit their host by killing competitors, predators or parasites, natural selection should favor the transmission of microbes with the most beneficial defensive traits. During the initiation of symbiosis, the host's ability to accurately pre-assess a symbiont's beneficial traits would be a selective advantage. We propose that one mechanism by which a host could recognize and select a beneficial partner would be if the latter displayed an honest signal of its defensive or other symbiotic capabilities. As one example, we suggest that polymorphic toxins and their surface receptors, which are involved in inter-microbial competition and predator killing activities, can be honest signals that facilitate partner choice in defensive symbioses.

Address
Department of Bacteriology, University of Wisconsin-Madison, Madison, WI, USA

Corresponding author: Goodrich-Blair, Heidi (hgblair@bact.wisc.edu)

Current Opinion in Microbiology 2016, **31**:184–190

This review comes from a themed issue on **Environmental microbiology**

Edited by **Monica Vasquez** and **Steven Hallam**

http://dx.doi.org/10.1016/j.mib.2016.04.010

Introduction

Microbes are fundamental components of all life on earth. Their metabolic and chemical diversity affords abundant potential for goods and services that may be exchanged with, or used against, other organisms they encounter [1]. In mutualistic microbial symbioses, microbes associate intimately with hosts (plants, animals, or other microbes) and provide activities that promote host health and reproductive fitness. Experimental evidence in diverse microbial symbioses has revealed that in numerous associations, a particular species or strain of microbe is faithfully transmitted to the next generation, while others are excluded. Here we review findings from recent and past literature, encompassing binary and consortial symbioses and those of invertebrates and vertebrates, to suggest the concept of honest signaling as a unifying model for molecular mechanisms by which partner selection and specificity is achieved.

Main text

Symbiont selectivity and partner choice

In many symbioses, the association between the host and microbe is specific, such that only one strain or species of microbe associates with or is transmitted by a particular species of host [2–4,5••,6••]. Evolutionary theory suggests that in environmentally transmitted symbioses, in which the symbiont is recruited anew each host generation, the host may select best performing symbionts from a population of possible partners, in a process known as partner choice [7]. While myriad data support the concept of partner choice, the mechanisms underlying this process remain obscure for the majority of symbioses [8].

Partner choice in some systems will depend on the host's ability to assess symbiont performance before engaging in the symbiosis. How are beneficial partners distinguished from the many other possible associates present in an environment? How are the distinctive characteristics of a beneficial microbe transduced to promote their exclusive physical association with an as-yet un-colonized host? The answers to these questions will help connect the concept of partner choice to the molecular events occurring during symbiont transmission.

The key characteristic that must be assessed in partner choice is the beneficial function(s) provided by the symbiont, two prominent categories of which are nutritional and defensive [9,10]. In the former, microbial symbionts provide essential nutrients and can enable expansion of plants and animals into diverse nutrient-poor or toxic niches that would otherwise be uninhabitable (Table 1). For instance, hydrothermal vent mussels from the genus *Bathymodiolus* are fed by a horizontally acquired gill-tissue symbiont that uses inorganic oxidation to fix carbon [11•,12••], and land plants acquire essential nutrients, such as nitrogen, through symbiosis with arbuscular mycorrhizal fungi or bacteria [13]. Symbiosis can also contribute to degradation of inaccessible nutrient stores, such as recalcitrant plant material that is made accessible to some types of ants through cultivation of herbivorous fungi [14]. Similarly, nematodes in the genera *Steinernema* and *Heterorhabditis* can kill living insects for use as a food source by virtue of symbiosis with *Xenorhabdus* and *Photorhabdus* bacteria, respectively, which contribute to killing and tissue bioconversion of the insect [15].

Table 1

Symbiotic systems: host, microbial partners, and their associated interactions

Host organism	Microbial symbiont	Nutritional or defensive symbiont	PTS/antibiotic effectors	Reference
Bathymodiolus azoricus	Sulfur-oxidizing (SOX) symbiont	Both	YD-repeats RTX	[12••]
Bugula neritina	*Candidatus* Endobugula sertula	Defensive	Bryostatin	[43,54]
Acromyrmex sp.	*Pseudonocardia* sp.	Defensive	Antimicrobials	[22•,55]
Philanthus sp. (beewolf)	Candidatus 'Streptomyces philanthi' (CaSP)	Defensive	Antimicrobials	[6••,56]
Acyrthosiphon pisum (pea aphid)	*Hamiltonella defensa*	Defensive	Shiga-like toxin, YD-repeats	[30,57]
Drosophila neotestacea	*Spiroplasma*	Defensive	Shiga-like toxin	[37•]
Steinernema carpocapsae (nematode)	*Xenorhabdus nematophila*	Both	YD-repeats, bacteriocin, lysine rich peptide	[2,23,24]
Heterorhabditis sp.	*Photorhabdus*	Both	Tc toxins	[29,58]
Steinernema feltiae	*Xenorhabdus bovienii*	Both	Predicted Shiga-toxin	[25••]
Danio rerio (Zebrafish)	*Vibrio* (ZWU0020)	Likely defensive	YD-repeats RTX	[49•]
Germ-free Swiss Webster Mice	*Lactobacillus reuteri*	Likely defensive	Bacteriocin	[4,44]

In defensive or protective mutualisms, the microbial symbionts help defend the host organism or its progeny from predators and pathogens [9], including by producing chemically diverse small molecules or protein toxins that can kill aggressor or competitor organisms [16] (Table 1). As just one example of many, *Altermonas* bacteria on the surface of shrimp embryos produce istatin, which protects the embryo from the fungal pathogen *Lagenidium callinectes* [17]. Also, *Hamiltonella defensa* provides protection to its host pea aphid *Acyrthosiphon pisum* by producing a phage-encoded toxin effective against parasitoid wasps [18]. Inter-microbial competition can also be a defensive symbiotic trait, since it can protect niche resources from opportunistic invaders. For instance, arthropods associate with antimicrobial-producing Actinobacteria that help protect food or progeny from pathogens [19–21,22•] and the nematode symbiont *Xenorhabdus nematophila* provides an advantage to its entomopathogenic nematode host by producing an R-type phage tail bacteriocin that kills the obligate symbiont of a competitor nematode [23].

In systems where antimicrobial or anti-predator activities are an essential component of the mutualism, natural selection will favor the inter-generational transmission of microbes with the strongest toxicity toward the target. Recent work in beewolf predatory wasps indicates partner choice is occurring in the transmission of the symbiont from the female antennae to the brood cell in which progeny cocoons develop. In experimental crosses, the antennae of female beewolves could be colonized by the symbiont of another beewolf. However, those females did not deposit antennal gland secretions into the brood cells, effectively blocking transmission of the non-native symbiont to progeny [6••]. Similarly, fungus-cultivating *Acromyrmex* ants exhibit self-grooming behavior, which may function to maintain the symbiont on the cuticle surface, more frequently when colonized by their native strain of antibiotic-producing defensive symbiont, relative to strains isolated from other ant populations [22•]. Such crossing experiments also demonstrated *Steinernema* spp. nematode adaptations and specificity for native species or strains of their *Xenorhabdus* bacterial symbionts [24,25••]. Similarly, strains of the dominant gut symbionts of honeybees and bumblebees show specificity for their respective hosts and can outcompete non-native strains even when numerically disadvantaged [5••].

Honest signaling

To provide a framework to investigate mechanisms of partner choice, we propose a model in which assessment occurs through honest signaling, wherein the symbiont, at some cost to itself, displays or sends a signal to the host of its ability to cooperate (e.g. provide nutrients or defensive functions). Here, we define a signal as a product of the symbiont that has evolved to be recognized by the host, and to which the host has evolved to respond. In our model, the honest signal presented to the host directly and accurately (i.e. non-deceptively) represents the beneficial activity the symbiont provides [26]. For example, honest signaling for nutritional and defensive symbioses might occur through the evolution of processes that lead to specialized accumulation and secretion of a metabolite characteristic of a nutritional pathway and an antimicrobial compound/toxin, respectively. The model requires that the host has evolved mechanisms to recognize the honest signal and to respond by promoting association with, or transmission of, the appropriate signal-producing symbiont. Below we expand on this model by describing how symbiont-produced toxins and antibiotics could serve as honest signals, functioning both as defensive factors that benefit their host and as distinctive traits enabling recognition and physical association during partner choice (Figure 1).

Defensive polymorphic toxins of microbial symbionts

Microbes produce a dizzying array of bioactive secondary metabolites and proteins, many of which have functions