What Biofilms Can Teach Us About Individuality

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1. Introduction

What is a biological individual? At first glance, the answer to this question seems obvious. The co-authors of this chapter are biological individuals. As is Sarah's pet parrot. However, when one turns to the biological and philosophical literature, one finds over a dozen accounts of biological individuality (Clarke 2010). These are not crank accounts, but significant ones in biology and the philosophy of biology. When considering life's diversity, this disagreement over biological individuality should come as no surprise. Run of the mill examples such as mammals and birds are easy cases, but start looking at plants or microbes, then the question of individuality becomes increasingly problematic.

Consider two standard criteria for biological individuality. According to one, the bottleneck view (Maynard Smith and Szathmáry 1995), an individual starts at the beginning of ontogenetic development, when an individual is a single cell or a few cells. Each new bottleneck is the beginning of a new individual. According to another account of individuality, the sexual view (Janzen 1977), a new individual begins when the genes of two parents are combined into a single genotype. Each new zygote, with its distinctive genes, is the beginning of a new individual.

These accounts of biological individuality do not sound so different, but an example shows that they are. Aphids are cyclical parthenogenetic organisms: they reproduce asexually in the summer and reproduce sexually once at the end of the

summer.[1] On the sexual account, an individual only begins after sexual reproduction. Thus the sexual view counts all of the aphids that occur during the summer as one individual. The motivation for doing so is the assumption that all of these aphids are clones of each other. The bottleneck account gives a different answer. Each aphid begins as a single cell and undergoes a bottleneck, so each aphid is a distinct individual. The sexual and bottleneck accounts of individuality provide different answers to the question of how many individuals are present. Notice that this disagreement does not merely affect the bookkeeping of individuals in a situation. The phenomenon of individuality is central in natural selection. Selection selects among variant individuals in a population. The process of natural selection, in other words, distinguishes individuals. Yet, biologists and philosophers lack a well-accepted account of individuality.

Recently, Godfrey-Smith (2009) and Clarke (in press) have offered trenchant reviews of standard definitions of biological individuality. They argue that such definitions are too limiting. They offer their own more liberal approaches to biological individuality. Rather than asserting that one process is central to biological individuality, as the standard definitions do, Godfrey-Smith's and Clarke's accounts allow that different processes give rise to biological individuals. We believe that Godfrey-Smith's and Clarke's theories are important steps in the right direction. Clarke's (in press) account captures the diversity of biological individuals in the organic world. Godfrey-Smith's account, though more liberal than standard accounts, is not liberal enough. Below we use the example of biofilms to test Godfrey-Smith's and Clarke's theories of individuality.

We also offer our own account of biological individuality. We follow in the

[1] We follow Clarke's (in press) description of this example.

tradition of Hull's (1980) interactor account of individuality, but we renovate Hull's theory in several ways. First, we place it in a general sortal framework, along the lines of Wiggins' (2001) sortal account of identity. There are different sorts of individuals in the world, and when asking if an entity is an individual we need to specify the sort of individual under consideration. Second, we dive into the metaphysics of individuals, articulating the different types of processes (internal versus external) that cause entities to be parts of an individual. Third, we drop Hull's use of replicator theory and argue for a liberal account of reproducers. In the end, we offer an interactor account of biological individuality embedded in a more general theory of individuality.

Before getting to these accounts of individuality, two preliminary remarks are in order. There are two main debates over individuality in biology. One concerns the ontological status of species. There the question of individuality turns on whether species are particulars or classes (Hull 1978). We will not discuss this debate. (See Wilson 1999 for papers that review this issue.) Our focus is biological individuals that play an essential role in natural selection.

The other preliminary remark concerns the choice of our example, biofilms. Biofilms are single or multispecies communities of microorganisms. We focus on biofilms in a study of individuality because biofilms display many individual-like characteristics. The microorganisms of a biofilm form and share an extracellular substance. That substance prevents predation and it captures and digests nutrients. It also allows the cells of a biofilm to communicate and share genes. At the same time, biofilms fail standard criteria for biological individuality, such as having bottlenecks or reproducing sexually or being composed of members of one species. Biofilms, in other

words, strain various accounts of biological individuality. But they also provide clues for constructing a more adequate theory of biological individuality. In addition, their nature has implications for a general metaphysics of individuality. The lowly biofilm, we shall see, can teach us a thing or two about the metaphysics of science.

2. Biofilms: A Primer

We begin by explaining the nature of biofilms in more detail. Biofilms are found throughout the environment. They grow on the rocks of rivers, the surfaces of stagnant water, and on our teeth. The bacteria of a biofilm collectively produce, and are embedded in, an extracellular polymeric substance (EPS). EPS matrices hold the cells of a biofilm together. More interestingly, they are digestive systems that trap nutrients in the environment and break those nutrients down with extracellular enzymes (Flemming and Wingender 2010). EPS matrices also protect biofilms with molecules that bind to antimicrobial agents and prevent their access to biofilm cells. In addition, EPS matrices are media for cell communication among the bacteria of a biofilm (see below), and they foster the exchange of genetic material through Lateral Gene Transfer (see below).

The life of a biofilm proceeds through a series of stages (Hall-Stoodley et al. 2004). For example, a multispecies oral biofilm begins its life cycle with first colonizers, *Streptococcus gordonii*, attaching to tooth surfaces. Then secondary colonizers from the species *Porphyromonas gingivalis* coaggregate with the cells already attached (Kolenbrander et al. 2010). Coaggregation is "a process by which genetically distinct bacteria become attached to one another via specific molecules" (Rickard et al. 2003, 94). It is common for biofilm formation to be a sequential process involving different species

at different stages (Kolenbrander et al. 2010). Once the biofilm is fully colonized it matures. Then dispersal cells are produced and released to the environment. A biofilm life cycle, thus, consists of four stages: planktonic lifestyle (cells live as single unattached cells); attachment; colonization; and dispersal.

The cells of a biofilm interact in numerous ways, giving credence to the claim that biofilms are individuals. Quorum sensing, for example, is a cell-to-cell signalling system that enables bacteria within a biofilm to respond to and regulate cellular density. Quorum sensing occurs through the secretion and detection of molecules called 'autoinducers.' When the concentration of autoinducers reaches a certain threshold, cell differentiation in a biofilm is affected (Davies et al. 1998). Another signalling system, called 'molecular signalling,' affects a biofilm's lifecycle. For example, low concentrations of nitric oxide produced by *P. aeruginosa* trigger biofilm dispersion (Stewart and Franklin 2008).

Another type of biofilm interaction is lateral gene transfer (LGT). LGT is gene transfer among bacterial cells that is not due to reproduction. It occurs among conspecific strains and strains in different species. Biofilms provide favorable conditions for LGT. Consider two LGT mechanisms: transformation and conjugation. Transformation consists of the uptake of free DNA from the environment by a bacterial cell. Transformation requires extracellular DNA. In biofilms, this prerequisite is met because environmental DNA is a major constituent of biofilms. The other mechanism for LGT, conjugation, occurs via cell-to-cell junctions or bridges. Such bridges allow the transfer of mobile genetic elements, usually plasmids (Thomas and Nielsen 2005). The physical stability caused by EPS matrices reduces the chance of conjugal bridges breaking (Ehrlich et al. 2010). In short, lateral gene transfer occurs within biofilms for

several reasons: the occurrence of extracellular DNA, high cell density, and the physical stability EPS matrices provide.

Stepping back from these details, we see that biofilms have repeatable life cycles. Those cycles are caused by various types of interactions within biofilms, such as quorum sensing, molecular signalling, aggregation, and lateral gene transfer. In addition, EPS matrices serve as digestive systems, defence mechanisms, and media for communication. Biofilms, we submit, are good candidates for biological individuality, a point we will argue further below.

3. Godfrey-Smith's Account of Biological Individuality

Godfrey-Smith's (2009, 2011a, 2011b) account of biological individuality starts with Lewontin's (1985) characterization of natural selection. According to Lewontin, natural selection occurs when three necessary conditions are met: there is variation among individuals (the principle of variation); that variation is heritable (the principle of heritability); and that variation results in differential fitness among individuals (the principle of differential fitness). Godfrey-Smith discusses all three of Lewontin's conditions for selection. When it comes to biological individuality he focuses on an aspect of selection that Lewontin leaves unexplored, reproduction. "The link between "individuality" and reproduction is… inevitable. Reproduction involves the creation of a new entity, and this will be a countable individual" (Godfrey-Smith 2009, 86). Among other things, individuals in natural selection must be reproducers: those entities that not only vary and have differential fitness, but also have countable descendants.

Godfrey-Smith's discussion of reproducers focuses on what he calls 'collective

reproducers' –individuals that reproduce using some but not all of their parts.[2] Multicellular organisms are collective reproducers. Godfrey-Smith measures such reproduction using three parameters. The first parameter is reproductive bottleneck. According to Godfrey-Smith, paradigmatic cases of reproduction require a bottleneck, such as when a zygote develops from a small propagule. Human reproduction involves such bottlenecks. On the other hand, no bottleneck occurs when a new structure is formed by the aggregation of cells, for example when free living *Dictyostelium* cells aggregate and form a slime mold (ibid., 95). Godfrey-Smith's second parameter for measuring reproduction is germ/soma distinction. This distinction measures the degree of reproductive division of labour within a reproducer. Humans score high because we have distinct germ and soma lineages, where the first type of lineage is responsible for reproduction. Sponges, on the other hand, score low on this parameter when they reproduce asexually because any fragment of a sponge can start a new sponge (ibid., 92). Godfrey-Smith's third parameter, integration, concerns the boundary between an individual and its environment, and the mutual dependence of its parts with respect to viability. Mammals have high integration, buffalo herds low integration, and sponges somewhere in between.

Are biofilms reproducers and individuals on Godfrey-Smith's account? Let's start with the bottleneck parameter. A bottleneck occurs when a new individual develops from a small propagule. A bottleneck does not occur when a new individual is the result

[2] Godfrey-Smith (2009) also discusses two other types of reproduction: simple and scaffolded. A simple reproducer reproduces using all of its parts. Scaffolded reproducers are reproduced by mechanisms external to them. Godfrey-Smith's discussion of reproduction focuses on collective reproduction, which we simply call 'reproduction' in the text.

of the aggregation of numerous cells. Slime molds form by aggregation and lack bottlenecks. Biofilms are in the same boat. As we saw in Section 2, biofilms form by aggregation and lack bottlenecks. Nevertheless, biofilms satisfy Godfrey-Smith's reason for positing bottlenecks as a condition for paradigmatic individuality. Bottlenecks foster biological individuality because when mutations occur in the germ-line of an organism, bottlenecks spread that genetic change to an individual's somatic cells (ibid., 91). Biofilms have an alternative process for doing this, lateral gene transfer. Biofilm evolution is due in no small part to the introduction of new genetic material within a biofilm, and then the transfer of that material to other parts of a biofilm (Ehrlich et al. 2010). LGT, in other words, causes the existence of stable variant biofilms, and thus contributes to the satisfaction of Lewontin's first condition, the principle of variation.

While biofilms score poorly on Godfrey-Smith's bottleneck parameter, they have an intermediate score when it comes to germ/soma distinction. That distinction measures division of reproductive labour. Recall that humans score high on this parameter: few of our parts are passed on, just our gametes. Sponges, when they reproduce asexually, score poorly on reproductive division of labour because any part can start a new sponge. Slime molds score in the middle: they have "some reproductive specialization" yet more of their parts can reproduce than the parts of a mammal (Godfrey-Smith 2009, 95). Biofilms have dispersal cells that are the source of new biofilms, moreover those cells form a significant part of old biofilms (Hall-Stoodley et al. 2004).

Biofilms do well on Godfrey-Smith's third parameter for reproduction, integration. Godfrey-Smith measures integration by how effectively an entity maintains its boundary between itself and the environment, and how much its parts depend on each

other for their viability. Biofilms are distinct from their environments. The cells of a biofilm are molecularly bonded through aggregation and bounded within an EPS matrix. That EPS matrix catches and digests nutrients from the environment and protects a biofilm's cells from predators. Furthermore, the cells of a biofilm share genetic material via LGT, and there is intercellular communication within a biofilm that regulates a biofilm's development. These interactions set a boundary between a biofilm and its environment (more on this below). Biofilms also score high on Godfrey-Smith's other measure of integration, the degree to which the parts of an individual rely on each other for their viability. There are a number of biofilm-level processes that cause bacteria to have a significantly higher survivorship when they are part of a biofilm than when they live on their own (Costerton 2007). For example, a biofilm's EPS matrix contains antibacterial chemicals that protect its component bacteria, and it contains mechanisms for catching and digesting nutrients.

Stepping back from these details, we see that biofilms score poorly on having bottlenecks, middling on division of reproductive labour, and high on integration. For Godfrey-Smith, paradigmatic reproducers, and consequently individuals, need to score high on all three parameters (2009, 94). Biofilms do not –they fail to have bottlenecks and are middling on division of reproductive labour. Nevertheless they score high on integration because they have a number of processes that promote their stability and demarcate them from the environment. Biofilms are biological individuals (a point we argue further below), yet they fail Godfrey-Smith's account of individuality. This shows that Godfrey-Smith's theory of individuals is too restrictive.

Biofilms reveal a further problem with Godfrey-Smith's account, namely his view

of what sort of parent-offspring lineages can be individuals. Some biofilms are multispecies. Godfrey-Smith allows the existence of multispecies individuals so long as the different species lineages within an individual run in tandem (2011b). He cites the case of aphids and their symbiotic bacteria to demonstrate this. These bacteria and their host aphids have the same reproductive cycle: an aphid mother transfers bacteria to its offspring through its ovary. Biofilms, however, do not meet the requirement that the lineages of a multispecies individual run in tandem. The bacteria that form a biofilm are scattered in the environment and they come from different sources. Furthermore, their coaggregation occurs at different stages of biofilm formation. In other words, the different bacterial lineages that comprise a biofilm do not run in tandem, and they fail to form a unified parent-offspring lineage in Godfrey-Smith's sense. As a result, biofilms do not conform to his notion of reproductive lineage and his account of individuality. Yet, we contend, biofilms are individuals.

Before leaving Godfrey-Smith's account of individuality, we should address several possible objections to biofilms being biological individuals. First, one might worry that biofilms are ecological communities and not individuals. We have tried to address that concern above. To emphasize that biofilms are individuals and not merely communities, contrast biofilms with common examples of symbiotic complexes, such as the symbiotic relation between ants and acacias (Godfrey-Smith 2011a). Bacteria in a biofilm exchange genetic content; ant/acacia symbionts do not. Bacteria within a biofilm build and employ EPS matrices. Such matrices, as we have seen, defend a biofilm's bacteria from predators, capture and digest nutrients, and facilitate communication among component bacteria. The sorts of interactions and interpenetrations that occur among the

bacteria of a biofilm far outstrip symbiotic and other kinds of ecological relations.

One might grant that biofilms are more organized than ecological units, but nevertheless maintain that biofilms are not individuals in natural selection, they are something in between. For example, Godfrey-Smith (2011b) discusses metabolic organisms. A metabolic organism is a system of entities that collectively work together using environmental resources to maintain that system. For Godfrey-Smith, such organisms may fail to be individuals (in natural selection) because they do not form reproductive lineages. Godfrey-Smith explains his individual/organism distinction using the example of squid-bacteria symbiotic complexes. Godfrey-Smith argues that such complexes are not individuals in natural selection because each complex is "a metabolic knotting of reproductive lineages that remain distinct" (ibid.). Using this concept of organism, one might object that biofilms are not individuals (in natural selection) but merely organisms as Godfrey-Smith defines them. We respond by pointing out that biofilms are not simply organisms *sensu* Godfrey-Smith. Bacterial lineages within a biofilm do not remain distinct, as do the lineages of squids and their symbiotic bacteria. Lateral gene transfer genetically blends the different species lineages of a biofilm. Then there are the other interactive processes among the bacteria of a biofilm that we have discussed. A biofilm is not a mere metabolic knotting of bacteria.

Finally, one might worry that if biofilms are not reproducers given Godfrey-Smith's multifaceted account of reproduction, then perhaps biofilms are not reproducers. And if biofilms are not reproducers, then they are not individuals in natural selection. We address this concern in Section 5. We suggest that biofilms are indeed reproducers using Griesemer's (2000a) theory of reproduction.

4. Clarke's Account of Biological Individuality

Like Godfrey-Smith, Clarke (in press) ties her account of individuality to Lewontin's conditions for natural selection. Clarke focuses on the mechanisms that underwrite the satisfaction of Lewontin's first condition, the principle of variability. She suggests that two types of mechanisms underlie the existence and maintenance of variation: policing and demarcating mechanisms. A policing mechanism is "any mechanism that limits the capacity of an object to undergo within-object selection" (ibid.). Policing mechanisms reduce conflict within an individual and prevent the break-up of an individual. Bottlenecks and germ/soma separation are examples of policing mechanisms. Bottlenecks reduce conflict by increasing the degree of genetic relatedness among subsequent cell lineages (Maynard Smith and Szathmáry 1995). Germ/soma division suppresses conflict among somatic cells within an individual by limiting reproduction to an individual's germ line (Buss 1987).

A demarcation mechanism is "any mechanism that increases or maintains the capacity of an object to undergo between-object selection" (Clarke, in press). Such mechanisms cause and maintain variants among the individuals of a population. Recombination, mutation, and polyploidy, according to Clarke, are demarcation mechanisms. They produce variation among individuals. Then there are demarcation mechanisms that preserve variant individuals, such as immune systems and mechanisms that construct and maintain an individual's physical boundaries.

Turning to individuality, Clarke (in press) maintains that biological individuals are those entities that possess policing or demarcation mechanisms. In other words,

individuals are entities that have mechanisms that cause and maintain stable variants for selection to act on. Are biofilms individuals on Clarke's account? As we shall see, biofilms fulfill Clarke's requirements for individuality: biofilms have both policing and demarcating mechanisms.

Biofilms have at least several policing mechanisms that diminish competition among component bacteria. The literature on cooperation among the cells of a biofilm centers on the notion of public goods. Public goods are costly products manufactured by some cells that benefit other cells in a biofilm, such as signaling molecules (for quorum sensing) and EPS compounds. The existence of public goods in biofilms poses the problem of what prevents the spread of cheats within a biofilm –those cells that benefit from the products of other cells but do not themselves produce public goods, or produce them to a lesser extent. As we shall see, three policing mechanisms foster cooperation among the cells of a biofilm: character displacement, ecological disturbance, and LGT.

One set of experiments exploring the causes of cooperation within biofilms focuses on the bacterial species *Pseudomonas fluorescens* (Rainey and Rainey 2003). One strain of *P. fluorescens*, the wrinkly spreaders, produces a public good, cellulosic polymer, which improves access to oxygen by enabling the construction of biofilms at the surface of liquids. However, biofilms with the wrinkly spreader strain are susceptible to invasion by another strain of *P. fluorescens*, the smooth spreaders: they reap the benefits of being part of a biofilm without paying the cost of building the biofilm. Brockhurst et al. (2006) investigate the susceptibility of biofilms with cooperating cells (wrinkly spreaders) to invasion by cheats (smooth spreaders). Their investigation has two results. First, division of labor among non-cheats increases the productivity of a biofilm because

a wider range of resources can be exploited. Second, the proportion of cheats is lower in communities with a diversity of wrinkly spreaders (non-cheats) than in communities with the same type of wrinkly spreaders. Brockhurst et al. (2006) show that character displacement among the wrinkly spreaders increases the productivity of biofilms and keeps the number of cheats in check.

Other experiments with *P. fluorescens* show that forms of ecological disturbance maintain biofilms with cooperating cells. Brockhurst, Buckling, and Gardner (2007) varied the degree of ecological disturbance affecting the biofilms they studied. They found that under frequent disturbance, the density of cells is below which biofilm formation is beneficial. Under intermediate ecological disturbance, the proportion of cooperators (wrinkly spreaders) peaks. When there is infrequent disturbance, the number of cheaters increases significantly and biofilms produce fewer public goods. Hence in cases of intermediate ecological disturbance, selection favors biofilms with higher proportions of cooperators. Brockhurst, Buckling, and Gardner's (2007) work shows that ecological disturbance can be a policing mechanism (*sensu* Clarke) against cheaters in biofilms.

Lateral gene transfer is another policing mechanism in biofilms. Mobile genetic elements (MGEs) are genes that can move among prokaryotic genomes via LGT. MGEs are akin to infectious agents, capable of benefiting or harming their bacterial hosts. Smith (2001) hypothesizes that if cooperation is coded in MGEs, then the lateral transfer of these mobile elements may infect non-cooperative bacteria and cause them to become cooperative and produce a public good. Nogueira et al. (2009) provide empirical evidence for Smith's hypothesis by studying the genes that code for the protein

secretome. Such proteins are costly to produce yet they benefit neighboring bacteria. Nogueira et al. (2009) found that the genes coding for the secretome are overrepresented in MGEs and are laterally transferred, thus keeping the number of cheats in check.

Let's turn to demarcation mechanisms. Recall that there are two types of demarcation mechanisms: those that cause variation among individuals and those that maintain that variation. Both of types of mechanisms occur in biofilms. One mechanism that causes the existence of diverse biofilms is mutation within the bacteria of biofilms. Another mechanism that promotes biofilm variation is LGT within a biofilm. It causes the existence of new strains of bacteria within a biofilm (Ehrlich et al. 2010). A third source of biofilm variation, aggregation, causes biofilm variation by bringing together different combinations of bacterial strains during biofilm formation. As we saw in Section 2, the strains that come together to form a biofilm can vary from biofilm to biofilm.

Then there are demarcation mechanisms that maintain variant biofilms. Clarke (in press) discusses the "[s]patial boundaries or barriers around a collection of objects" that maintain an individual. She offers cell walls as an example –they are boundaries around cells that keep their parts together. EPS matrices perform the same function for biofilms. They are central in establishing the boundary between biofilms and their environments (Stewart and Franklin 2008). For example, EPS matrices provide a barrier to antibacterial agents and they capture nutrients from a biofilm's environment. Furthermore, they maintain a biofilm's internal chemical composition, which is different from the chemical composition in its surrounding environment. Coaggregation is another mechanism that maintains variant biofilms. As we saw in Section 2, some species of

bacteria but not others can coaggregate to form particular types of biofilm. So not only does coaggregation bind the parts of a biofilm, it prevents bacteria of the wrong species from being part of a biofilm.

Stepping back from these details, we see that both policing and demarcation mechanisms promote biofilm individuality. Subversion within a biofilm is controlled by such policing mechanisms as lateral gene transfer, character displacement, and ecological disturbance. The existence and maintenance of biofilm diversity is caused by such demarcation mechanisms as mutation, lateral gene transfer, EPS matrices, and coaggregation. Recall that for Clarke an entity is an individual in natural selection if it contains policing or demarcation mechanisms. Biofilms have both types of mechanisms, so they are individuals on Clarke's theory of individuality. Clarke's account captures our test case for biological individuality.

5. Interactor Accounts of Individuality

In this section we develop an interactor account of biological individuality. Our account overlaps with Clarke's approach, but it explores different aspects of individuality and subsequently offers a different theory of individuality. Our presentation in this section starts with a general framework for individuality –general in the sense that it applies to biological and non-biological individuals. Then we focus on biological individuals in natural selection.

The general interactor framework we adopt has two components. First there is the sortal component: when asking if X is an individual we need to ask if X is an individual of sort S. Here we follow in the footsteps of Wiggins' (2001) sortal account of identity.

When asking if two entities are the same entity, we need to place that entity under a sortal and enquire about the identity conditions for the sort of entity in question. The guiding idea is that different sorts of entities have different identity conditions. We follow a similar route when it comes to individuals. When asking whether something is an individual we need to specify the sort of individual under consideration. Evidence for this sortal approach to individuality is found in the different sorts of individuals in biology and their varying identity conditions. There are individuals in natural selection (the focus of this chapter), individuals in systematics (species and other taxa, Hull 1978), metabolic individuals (Godfrey-Smith 2013), immune system individuals (Pradeu 2010), and undoubtedly other types of biological individuals.

The second component of this interactor framework concerns the interactions necessary for an entity to be a certain sort of individual. Individuals of different sorts have different outcomes (functions, states, products). Consider two examples: individuals in natural selection require processes that allow them to vary and pass on that variance; individuals in biological systematics require processes that cause them to be distinct lineages. Once we determine the sort of individual under investigation and the outcomes necessary to be that sort of individual, our focus turns to the types of interactions required of individuals of that sort. We need to ask if the parts of an entity appropriately interact among themselves or with their external environment to form the sort of individual in question. This framework for individuality is quite general. We see that as a virtue: it applies to various kinds of individuals, both in and outside of biology.

Turning to biology, numerous philosophers of biology adopt an interactor account of individuals in natural selection (Hull 1980, Sober and Wilson 1998, and Dupré and

O'Malley 2009). We offer such an interactor account as well. Our starting point is Hull's (1980) notion of interactor.

Hull offers an interactor-replicator framework for natural selection. In that framework, both interactors and replicators are necessary for natural selection. Replicators, according to Hull, "pass on their structure largely intact from generation to generation" (1980, 315). Genes and asexual organisms count as replicators for Hull. Though some asexual parents and offspring may not be genetically identical they are similar enough to pass Hull's standard. Sexual organisms, colonies, and more inclusive units are not replicators. Recombination in sexual reproduction, for instance, reshuffles the genetic contributions of parents so that sexual offspring fail Hull's standard for replicators. Turning to interactors, an interactor is "an entity that directly interacts as a cohesive whole with its environment in such a way that replication is differential" (*ibid*., 318). Hull suggests that organisms and perhaps colonies are interactors, but he is suspicious of more inclusive entities being interactors. In Hull's theory of natural selection, both replicators and interactors must be present for selection to occur, but they need not be the same entities. In fact, very few entities are both replicators and interactors. Hull suggests that genes fulfill both roles (*ibid*., 320). In the majority of cases, interactors and replicators occur at different hierarchical levels, for example, the organisms of a population are interactors and their genes are replicators.

Are biofilms replicators or interactors on Hull's account? Biofilms are not replicators. As we just saw, Hull believes that recombination prevents sexual organisms from being replicators. Biofilms are in the same boat as sexual organisms. Just as not all the genes of a sexual parent make it into an offspring, not all of the strains of a biofilm

make it into its descendent biofilms (Kolenbrander et al. 2010, 478). Furthermore, according to Ehrlich et al. (2010, 270), lateral gene transfer "among the component strains (and species) [of a biofilm] leads to the continuous generation of a cloud of new strains with a novel combination of genes." In other words, LGT can cause a biofilm to genetically change over time. Biofilms vary too much to be replicators.

Are biofilms interactors on Hull's account? That boils down to the question of whether biofilms interact with the environment as 'cohesive wholes.' Hull clarifies what he means by telling us that populations are interactors that have "populational adaptations, properties characteristic of the population as a whole that allow it to interact with the environment as a whole" (1980, 325). Hull contrasts a whole from a mere group of organisms that is "selected only incidentally –e.g., because all of its members happen to be in close proximity of each other" (1980, 314). Which side of this divide do biofilms fall? As we have seen, biofilms have numerous biofilm-level interactive processes that give their constituent cells an evolutionary leg up. From constructing EPSs to quorum sensing; from coordinating biofilm growth to shared defensive and nutrient gathering mechanisms. Biofilms are not mere groups of bacteria in close proximity to one another, but entities that contain numerous biofilm-wide processes that benefit the biofilm as a whole.

Given that biofilms are interactors and thus individuals on Hull's account, we think Hull's interactor theory of individuality is on the right track. However, we are not completely satisfied with that account. We have two major concerns. First, the notion of interaction needs further precision: do the parts of interactor need to interact, or can they merely interact with their environment? Second, Hull's account of interactor is part of

his interactor-replicator theory of natural selection. The replicator part has been challenged (Griesemer 2000a, Godfrey-Smith 2009). Our concern is that if biofilms are individuals in natural selection, they need to be reproducers. Biofilms are not replicators, so a different account of reproducer is needed. We will look at both of these concerns with Hull's account in more detail. Along the way we will develop an alternative interactor theory of individuals.

Like Godfrey-Smith and Clarke, our starting point is Lewontin's (1985) account of natural selection. As we saw earlier, it contains three necessary and jointly sufficient conditions for natural selection:

1. The individuals of a population vary (the principle of variation).[3]
2. That variation is heritable (the principle of heritability).
3. That variation results in differential fitness among those individuals (the principle of differential fitness).

Hull's interactors capture (1) and (3): they vary, and vary in fitness because of their interaction with the environment. Hull's replicators fulfill the role of (2): they pass on the variation among interactors.

Our first suggested refinement of Hull's notion of interactor concerns Lewontin's first condition, the principle of variation. That condition requires mechanisms that cause and maintain variation among individuals. Here we can disambiguate the metaphysics of individuals. The mechanisms that cause and maintain variation can work among the parts of an individual, or they can be environmental mechanisms that act on the parts of an

[3] Lewontin's (1985, 76) characterization of this condition refers to species rather than populations. We have replaced 'species' with 'population' to allow multispecies individuals.

individual. One might think that interactors are individuals in virtue of their parts causally interacting. But nothing in Lewontin's framework requires that the mechanisms that cause and maintain variant individuals must be mechanisms internal to individuals. They could, in principle, be external mechanisms outside an individual that act on the parts of an individual.

When we turn to biofilms, we see that they have both internal and external mechanisms that cause and maintain variable individuals. Lateral gene transfer is an example of a mechanism that causes and maintains variants through the interaction of an individual's parts. As we saw in Section 4, the transmission of genes within a biofilm promotes the existence of non-cheats and keeps internal competition in check. Ecological disturbance, on the other hand, is an example of an external mechanism that promotes the stability of variant individuals. In Section 4 we saw that some forms of ecological disturbance select biofilms with a higher percentage of non-cheating *P. fluorescens*. Ecological disturbance keeps the number of cheaters in check and prevents subversion within those biofilms. The relevant point for us is that ecological disturbance, an external force, promotes the existence of stable variant biofilms.

Returning to an interactor account of biological individuality, such an account should recognize both types of mechanisms –those that work internally and those that work externally to cause and maintain variable individuals.[4] Accordingly, we recast Lewontin's first requirement as:

1´. The interaction of the parts of an individual, internally or externally, causes the

[4] There is a parallel here to Hull's (1978) account of species as individuals. For Hull, the mechanisms that cause species to be individuals can be internal or external. Gene flow among the members of a species is an internal mechanism. Stabilizing selection that acts separately on the organisms of a species is an external mechanism.

existence and maintenance of variation among individuals in a population. There is a general lesson here. The case of biofilms raises the question of whether the maintenance of individuals could be due to external forces acting on the parts of an individual rather than internal causal interaction. What we know to date about biofilms indicates that both external forces and internal interaction cause and maintain stable variant biofilms. The point we would like to highlight is that an individual (biological or otherwise) could be the result of just external forces affecting its parts. This goes against the common intuition that individuals must be composed of internally interacting parts. Here is an example from science reminding us that we should not assume that the world conforms to our intuitions. Whether an individual must be maintained by interaction among its parts depends on the empirics of the individual in question, not on *a priori* metaphysics.

We have discussed Lewontin's first condition, the principle of variation, and framed it in interactionist terms. Let's turn to his second condition, the principle of heredity. Throughout this chapter we have maintained that biofilms are individuals. One might worry about this assumption by questioning whether biofilms are reproducers. Recall that if natural selection is to occur, the individuals in that process must reproduce.[5] If biofilms are individuals in natural selection, then they must reproduce. Consequently, we should say why biofilms are reproducers. As we shall see, explaining why biofilms are reproducers will help us fill out Lewontin's second requirement for natural selection.

Thinking of biofilm reproduction as replication won't work. Recall that

[5] Bouchard (2010) questions this assumption. He suggests that in some cases we should count the fitness of an individual in terms of differential growth rather than differential reproduction.

replicators "pass on their structure largely intact from generation to generation" (Hull 1980, 315). Hull does not think that sexual organisms pass on enough of their structure to be replicators because parents and offspring contain different combinations of genes. Similarly, the bacterial strains that form biofilms vary (Ehrlich et al. 2010). By Hull's standards, biofilm structures do not remain sufficiently intact to be replicators. Notice that biofilms are not an outlier counterexample to a replicator theory of reproduction. If reproduction boils down to replication, then sexual organisms do not reproduce either.

Alternatively, perhaps we should adopt Godfrey-Smith's notion of reproducer (2009). However, as we argued in Section 2, biofilms fail to satisfy Godfrey-Smith's account of reproduction. For Godfrey-Smith, paradigmatic individuals have bottlenecks. We have seen that biofilms do not have bottlenecks, yet they perform the desiderata for bottlenecks –the spreading of genetic novelty within an individual. Furthermore (and discussed in Section 2), biofilms do not form the sort of parent-offspring lineages Godfrey-Smith attributes to individuals. For Godfrey-Smith, multispecies individuals must have lineages that run in tandem (2011b). As we saw earlier, the bacteria that form a multispecies biofilm come from different sources and coaggregate at different stages of biofilm formation. Given these considerations, Godfrey-Smith's account of reproducer does not apply to biofilms and a different account of reproducer is needed.

Griesemer (2000a, 2000b) offers an account of reproduction that captures biofilm reproduction. According to Griesemer, reproduction is the "multiplication with material overlap of mechanisms conferring the capacity to develop" (2000a, 361). There are two parts to this account. First, parents and offspring must have a genealogical relationship caused by material overlap. Second, entities capable of reproducing must develop or

have life-cycles. Griesemer describes development as the acquisition of the capacity to reproduce (2000a, 360). For Griesemer, "[t]he realization of a reproduction process entails the realization of a developmental process. The realization of development entails reproduction" (2000b, 74). This interdependence between reproduction and development forms a hierarchical structure. The reproduction of a multicellular organism requires an organism to develop from cells; cell reproduction requires cells to develop from organelles and chromosomes; and so on. This hierarchy bottoms out at the level of "null development," which is a case of reproduction in which offspring lack the capacity to develop (Griesemer, 2000a, 362). DNA, for instance, reproduce but don't develop.

Biofilms satisfy Griesemer's account of reproduction. Once a biofilm matures, it releases cells to the environment, either as individual cells or as clumps of cells. For example, *P. aeruginosa* biofilms produce motile cells that swim out of a biofilm, and *S. aureus* biofilms shed clumps of hundreds of non-motile cells (Hall-Stoodley, Costerton, and Stoodley, 2004). The released cells, or their descendants,[6] aggregate with other cells and form new biofilms. New biofilms, thus, are built using material contributed by old biofilms. Furthermore, that material provides new biofilms with the capacity to develop. First note that biofilms have developmental life-cycles. As we saw in Section 2, biofilm formation involves a life-cycle of four stages: planktonic lifestyle, attachment, colonization, and dispersal. Biofilm formation also has the reproduction-developmental hierarchy that Griesemer proposes. The reproduction of a biofilm requires a biofilm to develop from cells; and cell reproduction requires cells to develop from organelles and chromosomes.

[6] Some bacterial cells in their planktonic stage multiply through binary fission.

Biofilms reproduce according to Griesemer's account. Neither replicator theory nor Godfrey-Smith's account of reproduction captures biofilm reproduction. Griesemer's account does, and therefore it is an appropriately inclusive account of reproduction. We should hasten to add, however, that we are not suggesting that Griesemer's account should be the universal theory for all cases of reproduction. As inclusive as Griesemer's account is, it may leave out some reproducers. Godfrey-Smith (2009, 83-84) argues that retroviruses fail Griesemer's requirement of material overlap. We are sympathetic to thinking of 'reproducer' in a disjunctive fashion: some reproducers satisfy a bottleneck account like Godfrey-Smith's and others fall under Griesemer's theory. Arguably, the way to define 'reproducer' should be akin to the way that Clarke (in press) defines 'biological individual': reproducers are multiply realized. One reason to adopt such an approach to reproducers is that new types of reproducers may evolve which are not captured by our current theories of reproduction. In light of these considerations we recast Lewontin's second condition as follows.

2´. Variation is inherited through the reproduction of individuals.

'Reproduction' here refers to a disjunction of legitimate accounts of reproduction, where no one account is seen as the universal definition of 'reproduction.' The qualifier 'legitimate' is inserted to guard against the adoption of any proposed account of reproduction.

It is now time to step back and provide a summary of the interactor account of biological individuality on offer.

X is an individual in natural selection if:

i. The interaction of the parts of X, internally or externally, causes the existence and maintenance of variation among Xs in a population.

ii. That variation is inherited through the reproduction of Xs.

iii. That variation results in differential fitness among Xs.

This account is obviously framed in terms of Lewontin's often-cited conditions for natural selection. We have recast (i) and (ii), but not (iii). For (i), we explored the different ways that individuals can be stable variants. For (ii), we discussed the notion of heredity in terms of reproducers to highlight the reproductive aspect of individuality. Throughout this discussion we have used biofilms as our test case for formulating (i) and (ii).

6. Conclusion

This chapter offers an interactor account of biological individuality embedded in a more general theory of individuality. The biological account takes its lead from Hull's interactor notion of individuality but improves on Hull's account. It decouples an interactor theory of biological individuality from replicator theory. It explicitly places such an account within the context of Lewontin's requirements for natural selection. And it explores the nature of interaction by highlighting the difference between internal and external factors that maintain variant individuals. Most importantly, the suggested theory of biological individuality is more inclusive than traditional accounts and Godfrey-Smith's approach, thus it better captures the diversity of individuals in the biological world.

The more general approach to individuality outlined here employs a sortal framework. That framework captures the idea that there are different sorts of individuals in the world. When asking if an entity is an individual, we need to consider the sort of individual under study and ask if the parts of that entity interact (internally or externally) such that it produces the type of outcome that individuals of that sort produce. The inclusiveness of this sortal framework is a virtue –it allows for the production of different theories of individuality corresponding to the different kinds of individuals in the world.

Finally, biofilms can teach us a thing or two about individuality. Within biology, the nature of biofilms teaches us that standard ideas about individuals in natural selection should be abandoned. Individuals in natural selection need not have bottlenecks or a high division of reproductive labor. Such individuals can be composed of lineages from different species and those lineages need not run in tandem. Biofilms also teach us that a proper theory of reproduction should be more inclusive than commonly conceived. When it comes to individuality more generally, biofilms teach us that common intuitions about the types of relations required among the parts of an individual could be wrong. Biofilms may not be the most exciting individuals in biology, but studying them improves our understanding of individuality.

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