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A Critical Approach to the Definition of Darwinian Units of Selection

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Abstract. What are the biological units of selection? In fact, the notion of "unit of selection" (UOS) is blurred by ambiguity and controversy. To further evaluate the biological entities that are the objects of natural selection, three novel conceptual criteria (holism, minimalism, functionalism) are critically applied; they reveal, in addition to the self-evident case of the "individual," at least six distinct types of UOSs. These UOSs do not always have a defined structural organization; they can be parts of a living organism, a cohesive group of conspecifics, a multiunit entity, a totipotent cell, a DNA fragment, or a whole organism. UOS types diversify by amalgamation or parcelation processes of apparent entities. Therefore, previous attempts to characterize the UOSs solely on some morphological levels (gene, individual, group) without applying stringent criteria have failed to cope with the structural variations of natural phenomena and have led to the ambiguity of terms used.

Introduction

Much of the ambiguity, confusion, and controversy engendered by the concept of the "unit of selection" (UOS) seem to arise from a failure to identify the biological entities upon which natural selection operates (Sober and Wilson, 1994; Mayr, 1997; Gould and Lloyd, 1999, and literature therein). Along with the debates about the three to four possible organizational levels of selection (gene, individual, group, and metapopulation), the objections to the hierarchical theory of selection (Wilson and Sober, 1994; Michod, 1997; Gould, 1998; Gould and Lloyd, 1999), and the distinction between transmitted units and those which transmit (Wynne Edwards, 1962; Lewontin, 1970; Mayr, 1970, 1997; Dawkins, 1976; Hull, 1980; Gliddon and Gouyon, 1989; Sober and Wilson, 1994; Wilson and Sober, 1994; Williams, 1996; Gould, 1998), "metaphors have replaced the empirical world as foci for discussion while precise meanings and derivatives have been forgotten in the process" (Slobodkin, 1986). Even the basic term "unit of selection" is under dispute (Wilson and Sober, 1994), bearing polemic aspects (Mayr, 1997) as do other terms in this discipline (Gould and Lloyd, 1999).

One approach to clarifying such an ambiguous field is a critical evaluation of the arguments and definitions used (Hull, 1980; Sober and Wilson, 1994; Mayr, 1997; Gould and Lloyd, 1999). Such a reevaluation process might germinate a novel idea or might help dispel excessive ambiguity. On the other hand, anathematized concepts could reappear, revealing further ill-considered definitions (Gould and Lloyd, 1999) or adding additional ambiguities. An alternative approach is to envisage the main controversial issues through an untraditional analysis. In this essay, such an untraditional approach is used to examine the biological entities that are the objects of natural selection. By adapting the unbiased principle that any living thing can be all or part of a potential UOS, we can critically evaluate organisms—regardless of their level of morphological organization—on the basis of a few conceptual criteria.

Criteria for Analysis of UOSs

Three conceptual criteria guide this examination:

Holism

Genes and soma are not necessarily independent. The distinction between the terms "interactor" and "vehicle," as opposed to "replicator" and "gene" (Dawkins, 1976; Hull, 1980) is central in the debate over UOSs (Hull, 1980; Sober and Wilson, 1994; Mayr, 1997). The use of these terms to

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identify different units of selection evolved from the *a priori* rationale that living organisms are made of at least two distinct types of evolutionarily selected units. Additionally, the notion of the UOS has become ambiguous because it was used to refer to either replicators or vehicles, depending on the choice of the author (Wilson and Sober, 1994). I suggest that this rationale is false and misleading, that it artificially distinguishes between "genes," "information," and "replication" on the one hand, and "soma," "vehicle," and "interactor" on the other (Lewontin, 1970; Dawkins, 1976; Hull, 1980; Buss, 1982; Gliddon and Gouyon, 1989; Sober and Wilson, 1994; Mayr, 1997). The genes in any organism have a fate in common with their amalgamated soma (Sober and Wilson, 1994, and literature therein). They are part of a whole; they are not completely independent (with the exception of specific cases as outlined in the next section), but rather functionally integrated within the soma. In physics, light and mass are regarded as two facets of energetic matter. Similarly, in biology, genes and soma should be regarded as two facets of an organic entity that constitutes a living organism. Even the term "unit" (Oxford Dictionary) embraces this metaphysical concept of holism. A unit is a thing (individual, person, group, etc.) that is complete or distinctive and that has the characteristics of the complex whole. Following this rationale, the so-called replicators and interactors of each entity are intermingled to form, for each UOS, its idiographic (its own peculiar) entity, which is presented to natural selection as a coherent whole. This is in contrast to the acknowledgement of recent years that interactors, not replicators, constitute the causal unit of selection" (Gould and Lloyd, 1999).

Minimalism

Ignore complex cases; choose the simplest ones. Additional ambiguity is caused by different hypotheses for the UOS that deliver opposing predictions about the traits that have evolved (Sober and Wilson, 1994; Wilson and Sober, 1994). In such cases, a search for the simplest manifestation of the system, the minimalist approach (Slobodkin, 1986), has been suggested to be the most useful in maintaining clarity. This approach has been characterized as "the process of deliberately choosing to work in the simplest possible mode that is still recognizable as part of an existing professional field." Slobodkin (1986) has also discussed the main objection against this approach as the claim for uncritical acceptance of standards. However, this objection may not be the case in the controversy over the UOS, where metaphors, rather than empirical themes, dominate the scientific discipline (Wilson and Sober, 1994; Gould and Lloyd, 1999). When employing the minimalist approach (Slobodkin, 1986), or the very similar "back to basics" (Sober and Wilson, 1994) treatment, complex cases (such as the situations illustrated in Wynne Edwards, 1962) are left aside for future analyses when the field will presumably be more formally organized. Therefore, we must accept the idea that the UOS theory, almost three decades after it was first elaborated (Lewontin, 1970), should still be conceptualized through the clearest examples.

Functionalism

UOSs function *in vivo*. A unit of biological organization upon which selection might act should be both an autonomous functional entity and physically and structurally coherent, even if it is in the form of a gene. It cannot be in the form of "information" or "avatar" (Gliddon and Gouyon, 1989; Tuomi and Vuorisalo, 1989a) or "anything in the universe of which copies are made" (Dawkins, 1989). A UOS must function, because functionalism is the primary focus of natural selection. Functionalism, therefore, does not rest upon an active maintenance of distinctive properties (Gould, 1998), but evaluates the general sum of independent activities presented by a UOS. At this point, the existence of only a single functional level or of several functional levels (in hierarchical order, Tuomi and Vuorisalo, 1989a; Gould and Lloyd, 1999; or not) will not be discussed. Only a holistic unit (possessing cohesive structural and information properties) may reveal the capacity for functionalism. Therefore, previously distinguished UOSs such as replicators, interactors, vehicles, memes, etc., that are literally not holistic, are excluded from being real UOSs. They remain as highly justified theoretical paradigms that characterize only components of holistic and functional units of selection.

The three conceptual criteria (holism, minimalism, functionalism) provide enormous flexibility for analysis and circumvent the use of ill-defined issues and debatable arguments. These criteria have been used to scrutinize different potential types of UOSs that are presented by a variety of organismal entities. The term "organism" refers here to "any biological entity whose parts have evolved to function in a harmonious and coordinated fashion" (Wilson and Sober, 1994). This analysis has revealed several types of UOSs; of these, one traditional and six new characteristic types (Table 1, Nos. 1–7) are briefly described below.

Seven Types of Units of Selection

I am—and part of me is it

Molecular sequences may themselves be UOSs. "Doctor there is a fly in my genome" was the title chosen by the journal *New Scientist* (Vol. 149, p. 16, 1996) for an article about a tiny fragment of an insect genome (called *mariner,* a jumping gene first discovered in the fruit fly *Drosophila*) that is embedded in human chromosome 17. This location directly coincides with a recombination hot spot and has been associated with distinct hereditary neurological syndromes (Reiter *et al.*, 1996). This is only one of an enor-

Table 1

Who is it that can tell me who I am?

—Shakespeare, *King Lear,* Act 1, Scene 4

No.	Type of unit of selection	Organizational level on which selection acts	Examples
1	I am—and part of me is it	On a molecular level, a piece of DNA, usually not larger than a single chromosome (B-chromosomes, however, can pair among themselves to form a chiasmata)	Symbiotic/parasitic DNA in eukaryotic and prokaryotic genomes
2	I am—and part of me is he	On a whole organismal level	Natural chimerism and mosaicism
3	I am—and this is actually he	On a cellular level	Germ cells parasitism
4	I am—and this is actually we	On groups of conspecifics that intermingled together	Mammalian whole-body chimerism and invertebrate multichimerism
5	I am—and this is actually only part of me	On different ramets of the same genet	Asexually developed organisms, monozygotic twins, polyembryony
6	We are—and this is actually me	On multiunit entities	Rhizocephalen cirripedia
7	I am—that I am (Exodus 3:14)	On the whole organismic level	Many unitary organisms

mous number of documentations that eukaryotic and prokaryotic cells carry foreign DNA molecules of various types (plasmids, B chromosomes, t haplotypes, retroviruses, and more), as well as diverse mobile DNA sequences (such as transposons, retrotransposons, LINEs, SINEs, mobile introns) that are transmitted vertically or horizontally within genomes (Zeyl and Bell, 1996; Flavell, 1999) and may be regarded as real UOSs. These DNA sequences have functional and holistic properties; they are characterized by a discrete organismal realm, function in a coordinated fashion, and are clearly subject to natural selection forces. Many examples now point to real UOSs situated within the genomes of other UOSs. A few will be outlined below.

One well-studied group is the B-chromosomes, a variety of germ-line parasites described from more than a thousand species of plants and animals. These small chromosomes do not contribute to the regular functions of the host, and their numbers per cell vary even within the same host organism. More important, although they share the same nucleus with regular chromosomes, they have evolved peculiar characteristics of their own. By various non-Mendelian systems of biased transmission and by their ability to move specifically to one of the two products of the first meiosis division (such as by avoiding penetration into the polar body during oogenesis), they increase their representation in the germline nuclei. The B-chromosomes in the wasp *Nasonia,* which are transmitted solely through sperm, are a representative case. The entire parental set of chromosomes in an infected zygote becomes condensed and is lost, leaving a haploidized animal that develops as a male, transmitting the B-chromosome to all its gametes (citations in Bell and Burt, 1990). Such functionalism of the parasitic entity reveals distinct host and parasitic units of selection. Within this context, I am reluctant to consider the B-chromosomes as selfish chromosomes. They are distinct molecular UOSs.

The mouse t haplotypes (each extending over the proximal half of chromosome 17) also have developed the ability to propagate at the expense of the wild-type homolog from heterozygous males. These entities probably evolved from a wild-type form of chromosome 17. Genes that were recruited later on, together with the addition of accompanying inversions, all increased the survival rates of the t haplotypes, until finally these entities started "taking on a life of their own" (reviewed in Silver, 1993).

Not only a whole piece of chromosome may be counted as a UOS; even transposable genetic elements, gene size segments of DNA, may be so considered. This field is too broad to be even partially covered here, so only the most relevant features of these mobile elements will be discussed. Many transposable elements have the ability to jump from place to place on the chromosomes; they can behave like new introns creating novel intron processing patterns; they may spread vertically and horizontally within host organisms; and they can promote their own replication (the functionalism component). With time, the mobile elements become domesticated through full integration into the host's genome. A good example is the *mariner* which, by being functional in both germ lines and somatic cell lines, could infect many organisms, crossing several phyletic borders (arthropods, platyhelminths, nematodes, chordates), probably by splicing into viral or other pathogenic genomes. During each introduction into a new host species, the *mariner* transposon was probably highly mobile and significantly disruptive. With time, more and more defecting transposons with mutations that disabled the cut-and-paste enzyme were accumulated, littering eukaryotic genomes

with foreign elements in varying stages of decay (Zeyl and Bell, 1996; Flavell, 1999), and blurring the boundaries (Dawkins, 1990) between two distinct units of selection. Many of the mobile elements constitute a significant portion of host genomes. The *Alu* elements (the largest family of SINEs in humans) represent in excess of 5×10^6 copies per haploid genome, and constitute about 5% of the human genome. The chloroplast genome of *Euglena gracilis* possesses at least 155 mobile introns, making up 39% of the genome by forming complex nested structures of introns within introns (literature cited in Zeyl and Bell, 1996).

From highly functional to nonfunctional: natural selection has shaped foreign DNA elements between these two extreme levels of activity. With respect to the UOS paradigm, elements with well-distinguished sequences and with high activity levels of their own (even if they are the size of a single gene) can be regarded as units of selection. Natural selection may act on them independent of their host, and may especially act on those elements that move between different organisms (Flavell, 1999). Other elements that are completely integrated in the host's genome, replicating when the entire collective of genes reproduce and contributing to basic functions and processes derived by the host cells (such as the LINE elements that preserve the telomeres of *Drosophila;* literature cited in Flavell, 1999), are clearly not UOSs.

I am—and part of me is he

In chimeras or mosaics, two or more UOSs amalgamated to form a single distinct UOS. Genetically nonhomogenous entities can be established by chimerism (a situation where an organism possesses cells simultaneously derived from at least two genetically distinct conspecifics) or by a mosaicism (production of an organism with genetically different cells that derived from a single zygote lineage). Both phenomena have been widely documented: chimeric entities in nature have been recorded from a variety of protists, plants, and animals, distributed over nine phyla (Buss, 1982); and a number of factors may produce mosaicism in almost any living organism (Benirshke, 1981; Hall, 1988; Gill *et al.*, 1995). Clear distinctions between chimeras and mosaics are often not available in reported cases because an insufficient number of genetic characters were employed (Benirshke, 1981). Although, in many cases, a chimera or mosaic seems to represent a single UOS, this "single organism" actually consists of two or more distinct embedded units of selection upon which natural selection acts. This type of "blurring of the boundaries" between the interacting entities (*sensu* Dawkins, 1990) obliges us to develop epistemological tools with which we may distinguish between false UOSs and real ones whose existence does not depend upon the researcher's perception.

Mosaic, sectorial, and cytomictial (mixed-cell) chimeras often occur after allogeneic encounters in a variety of colonial marine invertebrates (Rinkevich, 1996a). Participants in such chimeras are sometimes so intermingled that the death of one of them (*e.g.*, from senescence) results in chimeric death (Rinkevich and Weissman, 1989; Rinkevich *et al.*, 1992). The evolutionary significance of chimerism has been evaluated by comparing (Buss, 1982; Grosberg and Quinn, 1986; Rinkevich and Weissman, 1987a; Rinkevich, 1996a) the fitness cost-benefit ratio of the chimera with that of the genetically homogenous UOS. Several classes of benefits, including the increase of genetic variability, improvements in growth rates, reproduction or survivorship, and developmental synergism (citations in Buss, 1982; Rinkevich and Weissman, 1987a; Rinkevich, 1996a), have been attributed to chimeric states. Costs are the threats of somatic and germ-cell parasitism (next section) and, within chimerical selection, towards the more heterogeneous partner (Rinkevich, 1996b). If the outcome is a state of chimerical improvement, each UOS participating in it gains. Biological and environmental factors may directly affect just one UOS within a chimerical entity or may influence the chimera as a whole.

Vertebrates also exhibit a variety of naturally occurring chimeras, mostly in the form of dizygotic twin bone-marrow transplantation and as diseases like choriocarcinomas (Benirshke, 1981; Tippet, 1984; Benirshke and Kaufman, 1990). These and other types (whole body and germ cell chimerism, which will be discussed in the next two sections) are much more common than is usually believed.

Studies on cattle, sheep, goats, pigs, horses, humans, rodents, deer, mink, birds, and other vertebrates (Benirshke, 1981; Benirshke and Kaufman, 1990) have unequivocally established the occurrence of placental (when applicable) or vascular anastomoses between dizygotic twins. Hematopoietic precursor cells are then frequently exchanged during early embryonic periods; and by virtue of acquired tolerance, they may continue to propagate throughout life in the new host. The new UOS, thus formed at one higher level entity, also bears evolutionary relevance in at least two types of phenomena. The first type comprises resultant costs such as freemartinism (masculinization of the female twin, resulting in sexual reproductive sterility; Benirshke, 1981) and a high frequency of malignancy (Picus *et al.*, 1985). The second type—more interesting, but sporadically observed—is gonadal chimerism (literature cited in Benirshke, 1981). In this phenomenon, primordial germ cells may reach the gonads of the other partner through early vascular anastomoses. Since a mammalian XY germ cell, for example, has the capacity to develop into an oocyte (Evans *et al.*, 1977), it is possible that even in twins of different sexes moving germ cells may become functional, giving an evolutionary significance to both genotypes in the chimera.

Genetic heterogeneities are also frequently developed as single-gene, chromosomal, and germ-cell mosaicisms (Be-

nirshke, 1981; Hall, 1988; Gill *et al.,* 1995), and are also recorded in human monozygotic twins (Ford, 1969). This scientific field is too broad to be even partly covered here. As UOSs, however, many mosaic cases have evolutionary relevance because they are hereditarily transmitted and may manifest a variety of costs (Benirshke, 1981; Hall, 1988; Gill *et al.,* 1995; Rinkevich, 1996a). Studies of human syndromes in offspring have shown that somatic mutations of the germ line may occur in phenotypically normal parents (Hall, 1988). These mutations probably evolved from a germ-line cell or its precursors, before the meiotic event. The same holds for plants and for at least nine different animal phyla in which a variety of organisms develop by somatic embryogenesis (where at least one cell lineage remains totipotent throughout the whole life cycle) or epigenetic development (where sequestration of germ cells is made late in the life span; refs. in Buss, 1982; Gill *et al.,* 1995). Somatic mutations in those organisms not only provide the variation necessary to deal with fluctuating environments (Gill *et al.,* 1995), they also create new entities that may maintain and inherit the genetic heterogeneities through the colonial expansion of viable mutated cells.

I am—and this is actually he

Phenotypically expressed entities can serve as "incubators" for the germ line of other conspecific entities. For example, a detailed and very thorough study (Mayr *et al.,* 1979) reported the case of a human female chimera detectable only by investigation of her progeny. None of the four children fitted genetically with their mother, and none of the 21 unique genetic markers found in the children could be detected in the woman. The possibility of any type of somatic mutation was ruled out, as was the least probable hypothesis that all four children had been interchanged. The conclusion of this study was that this female possessed two populations of allogeneic cells, one in the soma and the second in her gonads. An extreme somatic clearance process was suggested for this case, occurring either in a dispermic chimera or after the fusion of two embryos into one entity (see next section), with only the germ line to be left from one partner.

Colonies of the cosmopolitan urochordate *Botryllus schlosseri* may undergo natural transplantation reactions upon allogenic contacts between their peripheral blood vessels. They may develop cytotoxic lesions in contact zones or form vascular parabionts (review in Weissman *et al.,* 1990; Rinkevich, 1992). This histocompatibility discrimination resides in a single highly polymorphic fusibility-histocompatibility (Fu/HC) locus (Weissman *et al.,* 1990). Allogeneic fusions occur between colonies that share at least one Fu/HC allele; rejecting partners share no Fu/HC allele. After fusion, all modular units (zooids) from one partner in the chimera are resorbed by massive phagocytosis, leaving the zooid of the other colony intact, a phenomenon called colony resorption (Rinkevich and Weissman, 1987b). In three clear, independent studies (Pancer *et al.,* 1995; Stoner and Weissman, 1996; Stoner *et al.,* 1999), polymorphic molecular markers were used to demonstrate somatic and germ-cell parasitism of the inferior partners in the resorption phenomenon. Of special interest are the cases where the soma were cleared of foreign cells, but the only foreign partner's cells were found in the gonads. This unilateral germ-cell parasitism (Pancer *et al.,* 1995; Stoner and Weissman, 1996; Stoner *et al.,* 1999) documents another example of an incubator that carries and successfully delivers the genetic material of an allogeneic partner to the next generation (Stoner *et al.,* 1999).

Incubated entities, as in the above cases, are the evolutionarily successful UOSs, whereas the *incubator* entities are those with the role of directly interacting with the environment. In such unique cases, natural selection therefore operates with consequences that do not fit the accepted dogma (Lewontin, 1970), because the positively selected organisms inherit different, nonrelated sets of genetic material. The intimate relationships between the incubator entities (which cannot be regarded as valid UOSs and better fit the notion of the "extended phenotype"; Dawkins, 1989) and the incubated UOSs are still unknown. Moreover, without discussing, at this point, the conflicts of interests between the genes of the incubated and the incubator entities, it is evident that the physically blended incubated entities blur the conventional practical divisions between one organism and the other. The perception of a UOS as a group of dispersed stem cells raises the conceptual dilemma of a physically noncoherent UOS.

I am—and this is actually we

Whole body chimerism—a complete integration of two or more genetically different conspecifics into a single unified entity, with a shared participation in the soma and the germ line—creates another type of self-maintaining UOS. Such a new form may bear specific properties, different from those expressed by each of the components. Natural selection may act simultaneously on each component and on each of the chimeric entities as a whole. In some chimeric entities, the physical boundaries between the different units are so blurred that a morphological separation between the components is not possible. The literature reveals instances where such a blending is beneficial to the original components, and others that are characterized by malformations or a variety of costs (such as higher rates of malignancy and other pernicious phenomena). Both situations will be discussed here, since successful sexual reproduction has been recorded even by malformed entities.

Colonies of *Botryllus schlosseri* may also form natural multichimeras (multiple partners; more than two fused ge-

notypes) that result from an aggregated co-settlement of Fu/HC compatible colonies (Rinkevich, 1996b). When compared with bichimeras, multichimeras grow faster; reach larger sizes; do not fragment; have lower frequencies of colony resorption cases; and like more equilibrated entities, show other features that increase robustness (Rinkevich and Shapira, 1999). In these "monsters," the various costly intraspecific conflicts between the participant genotypes neutralize each other, generating an improved entity. In such an instance, natural selection may act on the "group" level (the chimera as a whole; Rinkevich, 1996b; Rinkevich and Shapira, 1999). The increase in fitness of the multichimeric entity, a new higher level of UOS, eventually increases the individual fitness of each UOS within this chimeric alliance. Therefore, even less adapted genotypes may survive and propagate.

A whole-body chimerism in mammals is a state in which the entire body consists of cells with at least two genetic lineages that are derived from separate fertilization products (Benirshke and Kaufman, 1990). Two types of genetic chimerism are of interest here: the early fusion of two embryos into one entity and the case of dispermic-chimerism, simultaneous fertilization of an ovum and the polar body by two spermatoza (Bernishke, 1981; Tippet, 1984; Bernishke and Kaufman, 1990). Both conditions are characterized by uniform dissemination, throughout the chimeral body, of the different cell lineages in the admixture, and they are found frequently in a variety of animals (Benirshke, 1981), including humans (Tippet, 1984; Benirshke and Kaufman, 1990). In some cases, due either to limited background information or complexity, the two conditions cannot be easily distinguished. One such example (summarized in Tippet, 1984) is a case of a monozygous pair of male twins identical in chromosome markers, HLA, isozymes and serum proteins, both XX/XY in the blood, but differing in other organs sampled such as skin and secretory tissues. One explanation for the unusual chimerism was that two embryos started to develop as XY monozygotic twins. One continued in the normal way, whereas the second fused with a dead XX triplet embryo which was completely adsorbed. In humans, many of such whole-body chimerisms are characterized by sexual reproductive sterility and a variety of tumors, but some of them are fertile (Tippet, 1984). One of the most interesting examples is a report (Talerman *et al.*, 1990) of a 29-year-old phenotypic female, a true hermaphrodite with bilateral ovotestes, a 46XX/46XY karyotype, and a successful pregnancy (before the development of a dysgerminoma, a germ-cell tumor). Several XX/XY male phenotypic dispermic chimeras have been recorded as sexually normal by having children (Tippet, 1984), but there is yet no study that analyzes the possible activation of both germ lines in the gonads, or the genetic constituents of the offspring in fertile cases.

Whole-body chimerism, and even true hermaphroditism, were recorded in a variety of vertebrates, most commonly in cats, but also in dogs, mink, horses, pigs, cattle, sheep, goats, deer, rabbits, rodents, chickens, and primates. In humans, as in other animals, XX/XY dispermic chimeras tend to be phenotypically males (Tippet, 1984), a phenomenon which further simplifies sexual reproduction. It is also possible that many cases of dispermic chimeras, even XX/XY ones, may remain undisclosed (Tippet, 1984) as long as they are healthy and remain fit.

I am—and this is actually only part of me

The same UOS may replicate endlessly to produce multiple identical copies. When addressing the issue of the unit at which selection acts, most biologists take into consideration only a simple list of basic biological organizations (*e.g.*, genes, cells, organisms, group). Most discussions (but see Tuomi and Vuorisale, 1989a, b) eschew conceptually challenged phenomena, such as modular organisms (which consist of repeated morphological units) and organisms that propagate similar, but morphologically independent, structures through a variety of processes, wrongly subsumed under the title of "asexual reproduction." The fuzzy boundaries of terms like "individual," "colony," and "clonal organism" (Michod, 1997) become even more apparent when they emerge in evolutionary concepts, in our case the concept UOS. For example, what are the levels of structural organization and what is the UOS of a stand of 47,000 aspen trees covering 100 ha of land, all produced from a single founder tree by "asexual" reproduction process (Gill *et al.*, 1995)? Or of a large branching coral colony that, during an episodic storm, is broken into fragments which are "replanted" and grow separately in different microhabitats? Or where a larva of an ophiuroid echinoderm produces secondary larval clones (Balser, 1998)? Numerous sessile marine organisms can generate detached fragments that by different mechanisms are dispersed before establishing themselves as independent colonies (Highsmith, 1982; Wulff, 1991). Following that, even the analysis for fragment size may reveal a whole range of controversial aspects, since a variety of life history patterns—such as growth rates, partial or whole fragment mortalities, and fecundity—are directly correlated with size rather than, for example, with the classical evaluated trait of "age" in unitary organisms (Hughes and Connell, 1987).

For this consideration of the UOS issue and evaluation of organismal body constructions, we shall deliberately treat "asexual reproduction" and "modularity" in the wider sense. No consideration will be given to the order of integration in modular organisms, to the physiological or morphological aspects, or to life history parameters. Consequently, it is not important for this discussion whether modules emerge spontaneously by self-organization, are developmentally controlled by a genetic mechanism, or are the products of

environmental or biological causes that affect different conspecifics at random. All that matters is that when separation occurs, the original organism and the fragments continue to survive.

Three classes of "modularity," in which independent separated units (Harper, 1977) are produced, if taken together, may characterize another UOS prototype: a single entity that occurs simultaneously in several places, all distant from each other. The first class includes numerous colonial and clonal organisms (such as plants or marine invertebrates) that divide by fission (spontaneously, or under genetic control) to produce autonomous ramets. The second class includes unitary and clonal organisms (invertebrates, plants) that can, by budding, produce many similar modules that separate from their point of origin upon morphological completion. A well-known example is the freshwater hydra, a small carnivorous organism that, under normal conditions, shows no evidence of aging and continuously buds off unlimited numbers of "copies" of entirely comparable units (Slobodkin, 1986). Bosch *et al.* (1989) further described a dramatic mode of cloning by fission in the planktotrophic larvae of a sea star. The great multiplicative potential of this species prolongs the pelagic life of a genet and enhances its chances for recruitment into benthic adult populations. The third class includes mammalian monozygotic twins (two normally developed organisms that share the same genetic constituents) and polyembryony, in which the division of a single fertilized egg produces several to hundreds of similar genetic larvae. Polyembryony occurs in invertebrates and vertebrates and appears to be a paradox of evolution because it clones more of an unproven genotype at the expense of genetic diversity in a clutch of eggs (Craig *et al.*, 1997).

The above three classes of modular organisms share one basic life history trait, the production and dispersal of somatic individuals, the ramets. Each single genotype is therefore represented by more than one ramet. In ecological terms, each ramet could be regarded as an individual (Harper, 1977); from the perspective of the UOS, the whole genet constitutes a single unit of selection (assuming that no somatic mutation or any other type of somatic mosaicism is taking place). Among modular organisms, each unit of selection may be found simultaneously under different environmental conditions and exposed to a variety of selection pressures that sometimes oppose each other. Under these conditions, some ramets will die, while others will survive, which provides the option for each specific genet to "exercise" its phenotypic potentiality.

We are—and this is actually me

Several genets may form one coherent whole. The situation wherein several conspecific UOSs combine to form a morphologically new structure is best represented by certain primitive crustaceans (order Rhizocephala in the subclass Cirripedia, the barnacles). The rhizocephalans are mostly known for the genera *Sacculina* and *Peltogaster* (Hoeg and Rybakov, 1992; Glenner and Hoeg, 1995), which are parasitic, almost exclusively on decapod crustaceans, and are structurally unique. The "adults" have neither appendages nor segmentation, in contrast to all other arthropods, and their massive body is fastened to the host by a stalk from which "roots" proceed into the host tissues. These creatures also have neither an alimentary canal nor a mouth.

The life history of these parasitic crustaceans (Hoeg and Rybakov, 1992; Glenner and Hoeg, 1995) reveals a unique type of UOS. The cypris larva develops from a nauplius stage (both larval types are characteristic of primitive crustaceans). When the cypris is attached to the host crab, remarkable changes occur: the whole trunk of the parasite is discarded and a hollow, dart-like organ is formed. This organ is thrust into the crab's body cavity and the remnant of the cypris, a mass of undifferentiated cells enclosed within a thin ectodermal layer, is injected. The cell mass travels through the host's body cavity, attaches itself to the intestine, and anchors there by rootlets. Recent studies (Glenner and Hoeg, 1995) have further documented that the injected parasite has the form of a motile vermiform body that splits up into a number of naked, motile amoeboid cells. Each cell has the potential to develop into an adult parasite. A globular mass begins to develop. This structure will develop only the female gonads. Meanwhile, other cypris larvae attach themselves to the body of the juvenile parasite and inject their cellular contents into its mantle cavity. Only the first two will be successful in this enterprise. The cells from each such larva migrate and eventually enter one of the two "testes" (a better term would be spermatheca); there they develop into spermatozoa. Additional larvae attached to the parasite will be rejected. Reproduction is internal and within each parasitic unit.

Each single rhizocephalen organism is therefore an amalgamated structure, consisting of three distinct conspecific UOSs (two form only spermatozoa, one the soma and eggs). Together they participate in forming a different adult structural organism and a new unit of selection at a higher level. Selection acts only on this adult structure.

Epilogue

Thompsonia, another rhizocephalan parasite, is an extreme case; this crustacean has degenerated to the level of a fungus with rootlets that diffuse throughout the host crab. The rootlets branch off numerous sacs on small stalks, each sac contains one ovum per sac. The structureless parasite has no testes, ganglia, alimentary canal, or appendages, and there is no evidence of segmentation. It is believed that ova develop into cypris larvae by parthenogenesis, escaping the sacs through small openings (Lützen, 1992), although recent studies have challenged this hypothesis.

What is the unit of selection in this example? ("selection of?" *sensu* Sober, 1984). It is only one out of many cases where the data are insufficient for such analysis. However, the six types of UOSs characterized in this essay, in addition to the whole organismic level as a UOS (No. 7 in Table 1; not discussed here), indicate that a multiplicity of patterns are shaped by selective forces. The examples raised here symbolize the failure of many biologists and theoreticians to grasp the rich diversity of UOSs imposed upon the endless variety of adaptive structures found among living organisms. That many of the UOSs described in this essay are unconventional was therefore to have been expected, when the three novel conceptual criteria were applied to the analysis.

The concept of UOS is variously defined by different authors. Former attempts to identify the particular entities that are the targets of natural selection (Wynne-Edwards, 1962; Lewontin, 1970; Mayr, 1970, 1997; Dawkins, 1976, 1989; Hull, 1980; Buss, 1982; Gliddon and Gouyon, 1989; Sober and Wilson, 1994; Wilson and Sober, 1994; Williams, 1996; Michod, 1997; Gould, 1998; Gould and Lloyd, 1999) have suggested three or four potentially "structural" UOSs—the gene, the individual, the group, and the metapopulation—but there has been no consensus. Some (Kitcher *et al.*, 1990) have even argued that there are no "things" like UOSs, stating that "asking about the real unit of selection is an exercise in muddled metaphysics." However, I completely agree with the notion that "if selection is real, then so are units of selection" (Shanahan, 1997).

Kitcher *et al.* (1990), on the other hand, have correctly pointed to a major pitfall in the concept of the UOS by advocating that biologists "assume that for each selection episode, there is a unique account that will identify the level of selection." When the descriptions of UOSs in the literature are aligned with the organizational levels, they fail, in many cases, to grasp the structural comprehensiveness of other UOSs and no consistency emerges (Hull, 1980; Kitcher *et al.*, 1990; Sober and Wilson, 1994; Mayr, 1997; Shanahan, 1997). For example, the argument for the "gene," allegedly the most appropriate UOS (the reductionist approach), does not hold if we consider the changes that genes may go through during development (structurally and functionally). One such change is gene methylation. A methylated gene must be demethylated before it can be transcribed (Cedar, 1988). Another example is the changes that occur in the maturation of the mammalian immune system: the T and B cell genome rearrangement, the reshuffling of DNA fragments like a kaleidoscope to generate enormous genetic recombination patterns. Within a single individual, no two B cells, of more than 10^8 produced, are alike. In such situations, a single gene on its own may be regarded as only a tiny information fragment, a fraction within the organismal machinery that cannot produce anything unless it is in the right internal environment. With all its biological importance, a single gene cannot be termed a UOS (except in UOS type 1; Table 1).

In this essay, I have focused on the argument that real UOSs should evince a kind of holism and should possess the properties of independent functionalism. I have also eliminated cases that fail to comply with Slobodkin's (1986) minimalistic approach; thus I have omitted symbiosis (Nardon, 1999) and complicated cases such as symbiotic-parasitic relationships between a virus, an algal chloroplast, and a sea slug (Pierce *et al.*, 1999). Following from this analysis, six new types of UOSs were discussed (Table 1, Nos. 1–6), in addition to the self-evident case (Table 1, no. 7) of the "individual" (but see the search for several kinds of individuals based on characterizations of genetic uniqueness, genetic homogeneity, and autonomy; Santelices, 1999), which was not discussed here.

All UOSs differ from each other in substantial ways, and the characteristic properties of any one of them cannot be imposed on others. The analysis further revealed that neither the morphology nor the structural organization of a UOS is always orthodox. UOSs can also be blended morphologically into the somatic background of other conspecifics or different organisms. The blurred boundaries between organisms and colonies may raise a new theoretical question about the definition of "an organism." We find here that UOSs are associated with a variety of structural organizations, ranging from a DNA fragment (No. 1 in Table 1), to cells (No. 3), part of an organism (No. 5), whole organisms (Nos. 2, 7; that differ in the contents of the entity), a group of conspecifics (No. 4), and finally to a multiunit level entity (No. 6). The UOSs discussed here are, variously, based on one or a mixture of conspecific entities (Nos. 2–7), or on an association between several biological species (No. 1). There are probably other UOSs belonging to other biological organizations, even where the dividing line between components is not blurred; one good example is the existence of symbiotic unicellular algae within animal cells. Since these types of UOSs are more complicated, they were not analyzed here. In any event, all of the above UOSs bear in common their holistic character and their functionalism. All multiply through a variety of reproductive activities.

This essay reveals that a unit of selection can be a part of a biological organization, or can be an integration of several such organizations. It is not necessarily related to any conventional biological organization. Different selective forces operating on different levels of biological organizations may account for the diversification of UOSs by processes of integration (Nos. 1–4, 6; Table 1) or parcelation (No. 5). The simple characterization of the UOS on the basis of pure morphological level (gene, individual, group) may lead to unsatisfactory results. An entity like a single "individual" organism may represent a group of conspecifics that are intermingled (No. 4 in Table 1), only a part of a larger UOS (No. 5), an entity that possesses other types of UOSs (No.

1), another conspecific UOS (No. 3), a conglomerate of two units (No. 2), more than the sum of several conspecific UOSs (No. 6), or simply the traditional "individual" as the unit of selection (No. 7). Using an unprejudiced analysis on biological phenomena, we seem able to slip from the biased thinking of UOSs as being fixed entities, into an understanding that a UOS is the existence at a specific time point of a holistic and functional entity. Points of disagreement with traditional opinions always arise from the plurality in nature.

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