



## Article

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### **Transferability of genomes to the next generation: the fundamental criterion for the biological species**

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#### **Abstract**

Biological species as the basic units in biodiversity unify organisms that are similar in structure, development and ecological demands. Since Darwin's work on the origin of species, intensive efforts have been mounted to find a criterion for biological species that is common to all organisms, prokaryotes as well as eukaryotes, making species natural units. This has led to numerous species concepts, but none have met the requirement of universal application. Additionally, many concepts are based on criteria that can be used only for recognizing species (operational criteria), not defining the 'being' or make-up of the species (explanatory criteria). The definition of a species concept proposed herein regards species as a pool of similar genotypes interconnected in successive generations. This pool can be homogeneous or be divided into sub-pools. Interconnectivity within such pools is given by transferability, which means the potential to transfer complete genomes or genome halves to the next generation, perpetuating transferability. A change in genotype frequencies over successive generations is caused by preferred or restricted genome transfer due to intrinsic and/or extrinsic factors. Speciation is defined as splitting up or splitting off a pool of genotypes into pools with differing genotype frequencies, in combination with a definite loss of transferability of genomes or genome halves between these pools.

**Key words:** Biological species, genotypes, genome transferability, speciation

#### **Introduction**

Biological species are taken to be real things in the world. The recognition of such entities and the information transfer of their existence to other persons are fundamental for humans, especially when these differences are important for nutrition (e.g. Wilkins 2010). This is also true for animals, which learn to discriminate between different species and generalize from this experience to all similar forms (Hohenegger 1992). The reality of such entities explains the correspondence between classification of organisms by natives and specialized taxonomists (Diamond 1966). It contradicts the 'nominalistic species concept', which negates such entities as constructs of the human mind (Bessey 1908).

Since the hierarchical classification of Linnaeus (1758), classes of the 'species' category were used to represent the diversity of organisms. Starting with Darwin (1859), the nature of the biological species has been a longstanding and important topic, becoming intensified with the introduction of the 'biological species concept' (Mayr 1942). The biological species concept fails to be universal in two ways. First, it cannot be applied to all organisms because it is restricted to sexual reproduction. Second, 'isolation' as the result of speciation does not explain the factors leading to isolation and can be used only to delimit species. Isolation is thus restricted to recent species, because it can be tested solely on living organisms. Therefore, those factors causing isolation must be regarded as the fundamental criteria for a biological species concept. Paterson (1985) introduced the 'recognition species concept', terming these factors a 'common fertilization system' (Paterson 1993). The recognition criterion is not operational and thus inapplicable for delimiting species. Moreover, it also fails to be universal by excluding all organisms without a mating system.

Several species concepts have been established since the introduction of the biological species concept. All attempt to provide a universal concept that is general, applicable and theoretically significant, but all failed (Hull 1997). Mayden (1997) categorized the 25 concepts developed until 1996 and arranged them in a hierarchical order (Mayden 1997: Fig. 19.1).

The search for a universal species concept, the target of the ‘endless debate’ (de Queiroz 2007), resulted in the acceptance of the ‘evolutionary species concept’ (Simpson 1961, Wiley 1978) as the only one that is consistent with other concepts (Mayden 1997, de Queiroz 1998, 2005, 2007, Hey 2001, 2006). The ‘evolutionary species concept’, however, has the disadvantage of lacking a definition based on universal criteria. This concept only describes lineages as a series of ancestor-descendent populations. All criteria for species definition are regarded as secondary (contingent) properties, because a part of these properties in different combinations has the potential to establish lineages (de Queiroz 2005). This concept was named the ‘unified concept of species’. Accordingly, species defined as evolutionary lines can be regarded as individuals because they have their distinct location and own history with beginning and ending (Ghiselin 1974, Hull 1978, Gayon 1996).

A further but important problem in itemizing species criteria is the confusion between ‘cause and effects’, e.g. between operational and explanatory properties (and processes). For example, the criterion of realizing a sexual partner in the ‘recognition species concept’ is responsible for the genesis of a species thus an explanatory property, but morphology in the ‘phenetic species concept’ or the possession of shared derived character states in the ‘monophyletic species concept’ is not. They are the results of speciation, as is ‘isolation’ in the biological species concept (Paterson 1985). The former criterion is thus a factor giving rise to a species, while the latter are effects that can only be used to recognize species. Only ‘process related’ (Smith 1994) criteria that give rise to species can be used to define species. The static ‘pattern related’ criteria do not give rise to species; thus they can be used merely as operational criteria to identify and delimit species. Wilkins (2010) described the essence of the species groups—beside genetics—by their ‘typical developmental systems and responses to typical environments’. All criteria in Wilkins’ approach can only be used to recognize or delimit species. They do not explain the basic properties that are common to all species, making up species and causing their different developmental systems and response to the environment.

The following proposal is designed to find properties of a biological species that fully explain its origin, progress and termination making the biological species comparable for all organisms.

## **Fundamental Properties in a Classification System**

In classification theory, a set of objects *O* is grouped into classes *K* according to a set of characters or attributes *A*. Natural classifications are given when the distribution of character states within the classes is homogeneous compared to the distribution between classes, where it is heterogeneous; otherwise, class systems are artificial or arbitrary (Hohenegger 1989). An object is determined by possessing at least a single attribute (character, property). Thus, an object does not always have to be substantial but could be also conceptual (like words in languages). Classifying objects means the allocation of an object to a given class within a class system according to the attribute states.

Attributes or properties in a classification system can be differentiated into primary (or fundamental) and secondary properties. Fundamental properties characterize the objects of the classification system; therefore, they are general because they determine the objects. In constructing a system of classes, such fundamental properties can be the sole classification criteria or provide the basis of a classification system based on secondary properties. For example, “citizens of USA” as the fundamental property can be classified according to sex, age, language, income etc., all representing secondary classification characters.

Because species can be classified and compared based on their different genetic composition, development and ecological demands (all belonging to secondary classification characters), there must be fundamental properties that are common to all organisms within the species class, making them comparable. For example, making living things—organisms—comparable, they must possess a single or a set of fundamental properties represented in all subjects. In the case of organisms, this common property is “life”, which is defined as a set of properties such as the capabilities “response to stimuli”, “reproduction”, “growth”, “development” and the maintenance of homeostasis. Therefore, life defines organisms and makes them comparable. If such common properties do not exist for the species, comparison between species becomes impossible because it would involve comparing apples and oranges. In the following, an attempt is made to find fundamental properties of species that are valid for all organisms.

## The Fundamental Properties of the Species

Focusing on factors that are responsible for giving rise to a species must reveal fundamental properties of the species members. Every organism is represented by a genotype, thus species can be regarded as pools or clusters of similar genotypes (Mallet 1995). This reduction is legitimate, because both additional criteria of the biological species in Wilkins' (2010) discussion about the essence of species, 'typical developmental systems' and 'responses to typical environments', depend on genetics. All genes of an organism are summarized in the genotype, thus the latter can be used as a fundamental property of the species. It is operational, because homogeneity in the pools of genotypes can be measured. This reduces species definition to the problem of which explanatory properties, common to all genotypes, cause pools or clusters of similar genotypes, i.e. species and are responsible for cohesion in Templeton's (1989) species concept. They must explain

- 1) interconnectivity of succeeding generations within the pool of genotypes
- 2) changes within the pool of genotypes in successive generations, and
- 3) separating pools of genotypes.

Fulfilling the above requirements needs process related instead of pattern related fundamental properties. Indeed, transferability of genomes, either as complete genomes or by the combination of genome halves, allows defining species and can be used as the main fundamental property for a universal species concept. Transferability within the genotype pool explains the interconnectivity between succeeding generations, whereas different intensities of exchangeability between genomes mark changes in genotype frequencies during successive generations. Ultimately, differentiation within a genotype pool and the loss of transferability between the sub-pools leads to separate species.

A universal species concept can thus be determined by three propositions:

### Proposition 1

Species are groups of organisms sharing a pool of genotypes with the potential to transfer genomes (= transferability) to the succeeding generation, perpetuating transferability. This genome transfer can be achieved through strict partitioning (asexual reproduction) or a combination of genome halves originating from the parental generation (sexual reproduction). Groups belonging to a single species are thus interconnected by the potential to transfer half or complete genomes, thus preserving demographic exchangeability and mixing.

### Proposition 2

Abiotic and biotic extrinsic and intrinsic factors can promote or impede reproduction, thus altering genotype frequencies of the successive generations, especially when reproduction in new genotypes is unimpeded.

### Proposition 3

Splitting up/off the pool of genotypes without further transfer potential of genomes or genome halves and the loss of demographic exchangeability gives rise to different species. An abrupt major change in genotype frequencies altering a monotonous sequence of genotype frequencies is a special case of splitting off. It involves the synchronous extinction of the older sequence.

In the first proposition, the transition of the genome to the succeeding generation is explained in those cases where strict partition of the genome is either the only reproductive strategy or can be preceded by a combination of alleles from parental genomes. An unbalanced genome transfer through plasmid-mediated conjugation is typical for prokaryotes (horizontal genome transfer), whereas the combination of half genomes after meiosis, which is restricted to germ cells in multicellular organisms or to generative nuclei in multinucleate single-celled organisms, characterizes sexual reproduction and the exchange of genomes in eukaryotes (vertical genome transfer). A species is thus characterized by genomes that are transferable within and between sub-pools of genotypes based on asexual or sexual reproduction.

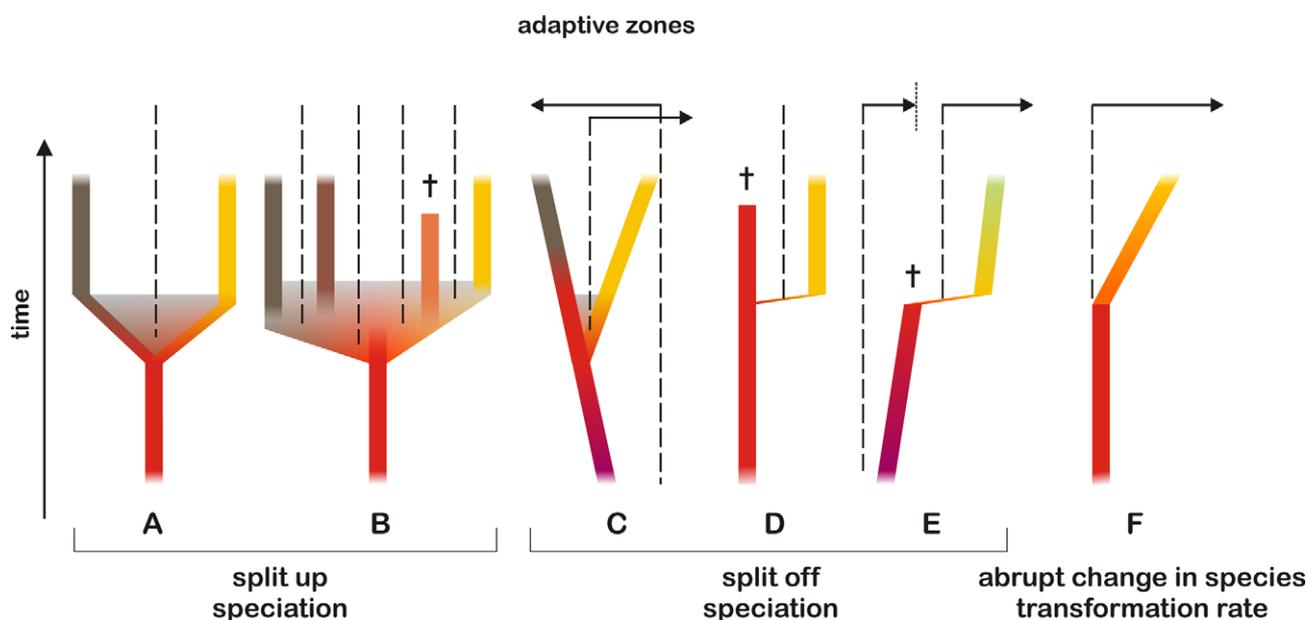
Evolutionary factors, such as mutation (including interspecific lateral gene transfer), selection, genetic drift and gene flow are incorporated in the second proposition. This proposition explains the change in genotype frequencies and incorporates the main criteria of the evolutionary species concept (Simpson 1961, Wiley 1978) and the general lineage concept (de Queiroz 1998, 2005). Furthermore, it explains adaptation to ecological niches and the diversification of developmental systems. A change in genotype frequencies can be directional via selection, which is termed 'gradualism' in evolution theory (Simpson 1944), but random walks may also cause lineages (Hunt 2010). A continuous sequence of genotype frequencies must not be artificially cut into segments as done by the chronospecies concept (George 1956) or the paleospecies concept (Simpson 1961).

Separating the pools of genotypes without further possibilities to transfer genomes is the main criterion for speciation as postulated in proposition 3. In prokaryotes this process can be instantaneous by mutation or lateral gene transfer, in which the new species adapts to a habitat through rapid asexual reproduction, thus becoming an ecotypic species (Cohan 2001, 2002). Otherwise, without changing the habitat and with no competition to the unaltered genotypes, it enlarges the pool of genotypes within the habitat.

In eukaryotes, instantaneous speciation occurs after lateral gene transfer in combination with asexual reproduction. After lateral gene transfer, new genotypes of eukaryotes can be established in a population when they are not impeded by sexual reproduction.

Natural speciation in sexually reproducing organisms is not instantaneous and requires much longer, from tens to ten-thousands of years depending on generation time. This is the 'gray zone' of the unified species concept (de Queiroz 2007), where separation between the newly developing species is blurred. Furthermore, the duration of the speciation process depends on the complexity of the organisms, generation time and reproductive tactics (semelparity versus iteroparity). Therefore, natural speciation in eukaryotes can only rarely be studied during a few human generations; most speciation events must be inferred from the geo-historical record (Smith 1994, Benton and Pearson 2001).

Speciation is caused by opening new adaptive zones (Schemske 2010). This opening could be continuous or instantaneous within geological time-scales. An instantaneous opening is mostly caused by rapid environmental changes, sometimes catastrophic events. Splitting up the pool of genotypes (Fig. 1 a, b) is continuous in respect to geological time intervals, and not restricted to two daughter species as proposed by proponents of the phylogenetic species concepts (Willmann 1985, Meier and Willmann 2000). It ranges from a split into two species (Fig. 1 a) when the adaptive zone is divided into two new zones, to several species when more zones open. This leads to a reticulate speciation pattern (Silvester-Bradley 1977, Veron 1995), typically lasting up to several ten-thousands of years depending on generation time (Fig. 1 b). During reticulate speciation, a temporary loss of transferability between sub-pools of genotypes is caused by geographic separation. These sub-pools can be taxonomically treated as subspecies (Mayr and Ashlock 1991).



**FIGURE 1.** Types of speciation. A, gradual speciation of two daughter species from a mother species; B, reticulate speciation; C, gradual speciation of a daughter species from a surviving mother species; D, punctuated equilibrium of speciation; E, punctuated gradualism (quantum evolution) of speciation; F, species transformation into a following subspecies by an abrupt change in the transformation rate. Dashed lines show the onset of new adaptive zones. Stasis in adaptive zones is shown as constant colours in vertical evolutionary lines. Horizontal arrows indicate monotonously changing adaptive zones. The gray zones show restricted or temporary loss of genome transferability between different subgroups (e.g. subspecies).

It is very difficult to determine species in a reticulate speciation event when observation is possible only during a short time interval, as is the case for biologists examining living organisms or palaeobiologists investigating a

single geological horizon. This explains the difficulties in determining species evolving today in a reticulate pattern: this 'gray zone' allows observation of a restricted genome transfer between distinct genotype pools due to sympatric hybridization, as often found in plants (Raven 1976, Futuyma 1998), or due to the circular overlap (ring species) in the geographic variation of animals (Mayr 1963).

In splitting off (Fig. 1 c–e), a daughter species separates from the mother species by either adapting continuously (Fig. 1 c) or discontinuously to the new adaptive zone (Fig. 1 d). At the same time, the mother species in the first case maintains its evolutionary tendency due to a continuously changing adaptive zone (Wiley 1981) or, in the second case, remains unaltered, in equilibrium with a stable adaptive zone (Eldredge and Gould 1972). Speciation by splitting off is analogous to budding, which also does not change the individuality of the mother organism.

An abrupt ending of the adaptive zone, often caused by environmental turnover, leads to extinction of the mother species. When a few genotypes have the potential to adapt to the new zone, the change in genotype composition is extremely fast, geologically speaking. After the sudden change in genotype composition, the daughter species either remains in stasis (in a stable environment) or shows evolutionary tendencies (in a monotonously changing new adaptive zone). This type of speciation, expressed in a discontinuity within the evolutionary line, is known as 'quantum evolution' (Simpson 1944) or 'punctuated gradualism' (Malmgren et al. 1983) (Fig. 1 e).

Quantum evolution must not be confused with an instantaneous change in transformation rates within a species (Fig. 1 f). In contrast to quantum evolution, genotype frequencies are the same before and after starting a different transformation rate. Often, an adaptive zone that is stable within a geological time interval suddenly starts to change monotonously due to beginning environmental changes, for example a constant temperature decrease after a stable period (Hohenegger 2007). In this case, a species can be divided into subspecies marking the different transformation rates.

## Recognition of the Biological Species

This proposal discusses 'species conceptualization', in which transferability of genomes acts as the criterion and must not be confused with 'species delimitation', which attempts to infer the boundaries and numbers of species (de Queiroz 2007). As explained above, the transferability criterion cannot be used as an operational criterion to delimitate species. For this purpose, mostly phenetic criteria are in use. Operational methods for species delimitation in prokaryotes have formerly been based on morphological criteria (Goodfellow 1997), creating distinct clusters (Sneath 1985); today, artificially fixed limits based on genomic similarities, which belong to phenetic criteria, are preferred (Stackebrandt and Goebel 1994, Roselló-Mora and Amann 2001). In eukaryotes, most methods for species delimitation are based on molecular and population genetics, sometimes in combination with geographical distances and ecology (Sites and Marshall 2003, Rissler and Apocada 2007). In the fossil record (e.g. older than 200,000 years), only one approach allows approximating species delimitations. This is homogeneity in morphological characters, combined—if possible—with homogeneous frequency distributions along environmental gradients and actualistic inferences or paradigmata of ontogenetic connectivity. Clearly, all the restrictions related to the incomplete fossil record must be considered.

## Conclusion

The single organism, regardless whether a prokaryote, a single-celled or a multicellular eukaryote, is characterized by its genotype. Thus, species are groups (clusters) of similar or identical genotypes. Similarities in genotypes can be expressed in similar phenotypes, although often not in a one-to-one relation, complicating species recognition and delimitation based on structure (including gene sequences) and form.

Similarities or identities in genotypes characterizing a species are caused by the transferability of genomes or genome halves to the succeeding generation through reproduction. This either keeps the pool of genotypes constant or enlarges it by demographic mixing in asexual reproduction or changes the genotype composition by a balanced mixing of genomes (sexual reproduction). The latter provides an implicit time-dimensional aspect for the species,

because genotypes and their composition always change in successive generations of sexually reproducing organisms. Due to this time dimension, species can be treated as individuals restricted to distinct regions with regard to birth, ontogeny and death. Species death can involve extinction or a split off into daughter species.

Developmental homogeneities of individuals within a species reflect homogeneous genotypes, which in turn are based on the transferability of genomes. In sexual reproduction, preferred reproduction between genotypes allows adaptation to continuously or rapidly changing environments; in prokaryotes, increased mutation or conjugation causes rapid adaptation to new ecological conditions.

After establishment of new adaptive zones, the above mechanism leads to species differentiation. Depending on number, the onset and the further role of the new adaptive zones (stable or continuously changing), various methods of speciation—grouped into split off and split up speciation—can be established. True speciation is characterized by a complete loss of transferability between the new species without the possibility to fuse (hybridise) when their adaptive zones come in contact or overlap. In case of a broad geographical distribution, the area might be differentiated into several adaptive zones, where transferability between subgroups is restricted or even lost. Temporarily disconnected adaptive zones can again become combined, reinstalling transferability between sub-pools of genotypes. Genotypically and morphologically different subgroups preserving transferability are thus not species; taxonomically, these structurally distinct subgroups can be treated as subspecies. Due to this uncertainty in the transition zone, a delimitation of species during the Recent Period is difficult in genotypically related but geographically separated groups. This is because the future behaviour of these groups as far as inter-group transferability is concerned is unpredictable.

A strong discontinuity in an evolutionary line is caused when a new adaptive zone for the species opens contemporaneously with the closing of the previous adaptive zone. This type of speciation, termed ‘quantum evolution’ or ‘punctuated gradualism’, is a special form of a geologically instantaneous ‘split off speciation’, where the mother species becomes extinct.

Contrary to quantum evolution, an instantaneous change in the transformation rate within an evolutionary line does not lead to new species. The clear differentiation in the species transformation rates can be taxonomically treated as subspecies.

This article presents a possible solution to the biological species problem, showing that the fundamental property ‘transferability of genomes or genome halves’ makes all organisms at the species level comparable. This property explains the differentiation into groups (species) that are homogeneous (typical) in their genetic composition, development, ecological demands and historical fate.

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## References

- Benton, M.J. & Pearson, P.N. (2001) Speciation in the fossil record. *Trends in Ecology & Evolution*, 16, 405–411.
- Bessey, C.E. (1908) The taxonomic aspect of the species. *American Naturalist*, 42, 218–224.
- Cohan, F.M. (2001) Bacterial species and speciation. *Systematic Biology*, 50, 513–524.
- Cohan, F.M. (2002) What are bacterial species? *Annual Review of Microbiology*, 56, 457–487.
- Darwin, C. (1859) *On the Origin of Species by Means of Natural Selection*. Murray, London, 318 pp.
- de Queiroz, K. (1998) The general lineage concept of species, species criteria, and the process of speciation. In: Howard, D.J. (Ed.), *Endless Form: Species and Speciation*. Oxford University Press, Oxford, UK, pp. 57–75.
- de Queiroz, K. (2005) A unified concept of species and its consequences for the future of taxonomy. *Proceedings of the Californian Academy of Science*, 56, supplement 1, 196–215.
- de Queiroz, K. (2007) Species Concepts and Species Delimitation. *Systematic Biology*, 56, 879–886.
- Diamond, J.M. (1966) Zoological classification system of a primitive people. *Science*, 151, 1102–1104.
- Eldredge, N. & Gould, S.J. (1972) Punctuated equilibria: An alternative to phyletic gradualism. In: Schopf, H.T.M. (Ed.), *Models in Paleobiology*. Freeman, Cooper & Company, San Francisco, CA, pp. 82–115.

- Futuyma, D.J. (1998) *Evolutionary Biology* (3<sup>rd</sup> edn). Sinauer Associates, Sunderland, MA, 763 pp.
- Gayon, J. (1996) The individuality of the species: a Darwinian theory? – from Buffon to Ghiselin, and back to Darwin. *Biology and Philosophy*, 11, 215–244.
- George, T.N. (1956) Biospecies, chronospecies and morphospecies. In: Sylvester-Bradley, P.C. (Ed.), *The Species Concept in Paleontology*. Systematics Association, London, pp. 123–137.
- Ghiselin, M.T. (1974) A radical solution to the species problem. *Systematic Zoology*, 23, 536–544.
- Goodfellow, M., Manfio, G.P. & Chun, J. (1997) Towards a practical species concept for cultivable bacteria. In: Claridge, M.F., Dawah, H.A. & Wilson, M.R. (Eds.), *Species: The Units of Biodiversity*, Chapman & Hall, London, pp. 25–29.
- Hey, J. (2001) *Genes, Categories and Species*. Oxford University Press, Oxford, 240 pp.
- Hey, J. (2006) On the failure of modern species concepts. *Trends in Ecology and Evolution*, 21, 447–450.
- Hohenegger, J. (1989) Klassifikation von Organismen und das “Natürliche” System. *Sitzungsberichte der Österreichischen Akademie der Wissenschaften, Mathematisch-naturwissenschaftliche Klasse, Abteilung 1*, 197, 135–181.
- Hohenegger, J. (1992) Species as the basic units in taxonomy and nomenclature. In: International Information Centre for Terminology (Ed.), *Proceedings of the 3<sup>rd</sup> Infoterm Symposium 1991*. Termnet, Vienna, pp. 15–30.
- Hohenegger, J. (2007) Population genetics and micropaleontology – Possibilities for testing different speciation models. *Denisia*, 20, 59–74.
- Hull, D.L. (1978) A matter of individuality. *Philosophy of Science*, 45, 335–360.
- Hull, D.L. (1997) The ideal species concept – and why we can’t get it. In: Claridge, M.F., Dawah, H.A. & Wilson, M.R. (Eds.), *Species: The Units of Biodiversity*. Chapman & Hall, London, pp. 357–380.
- Hunt, G. (2010) Evolution in fossil lineages: Paleontology and The Origin of Species. *The American Naturalist*, 176, Supplement, S6–S76.
- Linnaeus, K. (1758) *Systema Naturae*, 10<sup>th</sup> edition. Impensis direct. Laurentii Salvii Salvius, Holmiae (Stockholm), 1620 pp.
- Mallet, J. (1995) A species definition for the modern synthesis. *Trends in Ecology and Evolution*, 10, 294–299.
- Malmgren, B.A.S., Berggren, W.A. & Lohmann, G.P. (1983) Evidence of punctuated gradualism in the late Neogene *Globorotalia tumida* lineage of planktonic foraminifera. *Paleobiology*, 9, 377–389.
- Mayden, R.L. (1997) A Hierarchy of species concepts: the denouement in the saga of the species problem. In: Claridge, M.F., Dawah, H.A. & Wilson, M.R. (Eds.), *Species: The Units of Biodiversity*, Chapman & Hall, London, pp. 381–424.
- Mayr, E. (1942) *Systematics and the Origin of Species*. Columbia University Press, New York, NY, 334 pp.
- Mayr, E. (1963) *Animal Species and Evolution*. Belknap Harvard University Press, Cambridge, MA, 797pp.
- Mayr, E. & Ashlock, P.D. (1991) *Principles of Systematic Zoology* (2<sup>nd</sup> edn). McGraw-Hill, New York, NY, 475pp.
- Meier, R. & Willmann, R. (2000) The Hennigian species concept. In: Wheeler, Q.D. & Meier, R. (Eds.), *Species Concepts and Phylogenetic Theory*. Columbia University Press, New York, NY, pp. 30–43.
- Paterson, H.E.H. (1985) The recognition concept of species. In: Vrba, E.S. (Ed.), *Species and Speciation*. Transvaal Museum Pretoria, Pretoria, pp. 21–29.
- Paterson, H.E.H. (1993) *Evolution and the Recognition Concept of Species*. In: McEvey, S.F. (Ed.), *Collected Writings of H.E.H. Paterson*. John Hopkins University Press, Baltimore, MD.
- Raven, P.H. (1976) Systematics and plant population biology. *Systematic Botany*, 1, 284–316.
- Rissler, L.J. & Apocada, J.J. (2007) Adding more ecology into species delimitation: Ecological niche models and phylogeography help define cryptic species in black salamander (*Aneides flavipunctatus*). *Systematic Biology*, 56, 924–942.
- Rosselló-Mora, R. & Amann, R. (2001) The species concept for prokaryotes. *FEMS Microbiology Reviews*, 25, 39–67.
- Schemske, D.W. (2010) Adaptation and The Origin of Species. *The American Naturalist*, 176, Supplement, S1–S25.
- Sylvester-Bradley, P.C. (1977) Biostratigraphical tests of evolutionary theory. In: Kaufmann, E.G. & Hazel, J.E. (Eds.), *Concepts and Methods in Biostratigraphy*, Dowden, Hutchinson and Ross, Stroudsburg, PA, pp. 41–63.
- Simpson, G.G. (1944) *Tempo and Mode in Evolution*. Columbia University Press, New York, NY, 237 pp.
- Simpson, G.G. (1961) *Principles of Animal Taxonomy*. Columbia University Press, New York, NY, 247 pp.
- Sites, J.W. Jr. & Marshall, J.C. (2003) Delimiting species: a Renaissance issue in systematic biology. *Trends in Ecology and Evolution*, 18, 482–470.
- Smith, A.B. (1994) *Systematics and the Fossil Record*. Blackwell Scientific Publications, Oxford, UK, 223 pp.
- Sneath, P.H.A. (1985) Future of numerical taxonomy. In: Goodfellow, M., Jones, D. & Priest, F.G. (Eds.), *Computer-Assisted Bacterial Systematics*. Academic Press, Orlando, FL, pp. 415–431.
- Stackebrandt, E. and Goebel, B.M. (1994) Taxonomic note: a place for DNA-DNA reassociation and 16S rRNA sequence analysis in the present species definition in bacteriology. *International Journal of Systematic Bacteriology*, 44, 846–849.
- Templeton, A.R. (1989) The meaning of species and speciation: A genetic perspective. In: Otte, D. & Endler, J.A. (Eds.), *Speciation and its Consequences*. Sinauer Associates, Sunderland, MA, pp. 3–27.
- Veron, J.E.N. (1995) *Coral in Space and Time. The Biogeography and Evolution of the Scleractinia*. Comstock/Cornell, Ithaca, NY, 321 pp.
- Wiley, E.O. (1978) The evolutionary species concept reconsidered. *Systematic Zoology*, 27, 17–26.
- Wiley, E.O. (1981) *Phylogenetics. The Theory and Practice of Phylogenetic Systematics*. Wiley and Sons, New York, NY, 439 pp.
- Wilkins, J.S. (2010) What is a species? Essences and generation. *Theory in Biosciences*, 129, 141–148.
- Willmann, R. (1985) *Die Art in Raum und Zeit. Das Artkonzept in der Biologie und Paläontologie*. Paul Parey, Berlin, Germany, 207 pp.