

CHAPTER 1

General introduction

All meiotic individuals (basically, the sexual eukaryotes) undergo an alternation between haploid and diploid nuclear phases and thus spend some proportion of their life cycles as haploids and as diploids. When the haploid phase undergoes somatic growth by mitosis the species is said to have a haploid generation. Likewise, when the diploid phase undergoes somatic growth by mitosis the species is said to have a diploid generation. A life-cycle is haploid when it only has a haploid generation. This generation is self-sufficient and dominates the life cycle whereas the diploid phase is short-lasting, does not develop to be a true generation and is energetically dependent on the haploids. The most common example of a haploid life cycle is that of the bryophytes. On the contrary, a life-cycle is diploid when it only has a diploid generation, which is self-sufficient, and it is the haploid phase that gets to be short-lasting, does not develop to be a true generation and is energetically dependent on the diploids. The most complex, developed eukaryotes have diploid life cycles, namely the animals and the vascular plants with seeds. In between, there is a broad range of *taxa* that have a haploid-diploid life cycle. This is a life cycle where both nuclear phases may develop into generations with individuals that suffer mitotic somatic growth and are self-sufficient. It is the case of many algae, fungi, protists and the vascular plants without seeds.

Many arguments have been put forward about the evolutionary benefits of haploid life cycles and of diploid life cycles. More intriguing has been the evolutionary benefits of haploid-diploid life cycles. Richerd et al (1993) argued that such a life cycle benefits from reducing the cost of sex, whenever this cost is high, to produce the same amount of generations. Questioning this hypothesis, Mable and Otto (1998) argued that in such situations one might reasonably expect that this cost could be reduced through the evolution of asexual reproduction. These authors show an interesting point; and furthermore, such strategy is common in many algae and seaweeds. However, the validity of one alternative to reducing the cost of sex does not invalidate the benefits of the former. At this point it is important to clarify that haploid-diploid life cycles do have several sub-categories within. Hall (2000) differentiates them between biphasic and

polymorphic. In biphasic life cycles the individuals suffer mitosis in the diploid phase, undergo meiosis and then suffer mitosis again in the haploid phase, to finally undergo syngamy thus closing the cycle. In polymorphic life cycles the individuals suffers mitosis either on the haploid or the diploid phase but never at both. It all depends whether, after syngamy, the individuals anticipate or delay meiosis until the next syngamy event. Such a decision depends on their genetic background. Hall (2000) found that polymorphic biphasic life cycles prevail whenever there is a strong selection (in the adult fitness locus), loose linkage (between the adult fitness and the life cycle modifier loci) and meiotic mutations that are not too recessive (in the life cycle modifier locus). The biphasic life cycles can be further split into heteromorphic and isomorphic. In the heteromorphic the ploidy generations are morphologically very distinct. The most notorious case is that of kelp, for which the diploids are the largest autotrophs in the marine environment whereas the haploids are microscopic. In the isomorphic the ploidy generations are so similar to the point of usually being undistinguishable to the naked eye. Such is the case of many Rhodophyte algal species. Wilson (1981) argued that biphasic life cycle species may benefit from exploiting a broader range of environmental conditions if they differentiate their niches. Furthermore, Hughes and Otto (1999) found that for biphasic life cycles to evolve and prevail the distinct ploidy phases must have a certain degree of niche differentiation so that one phase does not competitively exclude the other. In Hughes and Otto (1999) work niche separation was attained by conditional differentiation, that is, the distinct ploidy phases adapt differently to the same environment in which they live in and explore, simultaneously.

Both Richerd et al (1993) and Hall (2000) hypotheses are well placed at the level of questioning the costs/benefits of ploidy alternation in life cycles, where they effectively differentiate between monophasic and haploid-diploid strategies and not between sub-categories within these classes. Unfortunately, it shall be hard for biologists to look for evidence of such hypothesis or to experimentally test them. Hughes and Otto (2001) hypothesis is also properly placed at the level of ploidy strategy. What is unique and remarkable about their hypothesis is the subject of ploidy alternation in biphasic life cycles imposing constrains upon the next hierarchical level of morphological adaptations. Interestingly, evidence in favor of such hypothesis is promptly provided by heteromorphic biphasic life cycles as those of kelp. It turns even more interesting as the work by Hughes and Otto (1999) proves that subtle dissimilarities between ploidy phases may be enough to promote the evolution and

stability of biphasic life cycles. It relates with Engel et al (2001) having determined the subtle dissimilarities between the survival rates of the isomorphic ploidy phases of *Gracilaria gracilis* to be the likeliest drive for their uneven ploidy phase abundances. Such uneven abundances of isomorphic distinct-ploidy phases, when these were expected to occur evenly, has until now been an intriguing matter of phycology.

A second big question about biphasic life cycles that has been running in parallel concerns the drive for the observed uneven abundances of ploidy phases so commonly observed in the field for algal species where ploidy phases are isomorphic, as reported by Santos and Nyman (1998), Engel et al (2001), Mudge and Scrosati (2003), Thornber and Gaines (2003), Scrosati and Mudge (2004a and b) and Dyck and deWreede (2006), among others. In such cases it would be expected to find similar abundances between phases as a consequence of isomorphism. Its answer may come from field observations together with experiments in controlled environments which have shown haploids and diploids may differ both in subtle morphological characteristics and physiological rates. Gonzalez and Meneses (1996) found that the gametophytes (the haploids) of *Chondracanthus chamissoi* are more fertile releasing more carpospores (diploid spores) than the tetrasporophytes (the diploids) release tetraspores (haploid spores). On the other hand, the tetraspores showed higher settlement and germination rates than the carpospores. Also, diploid adults showed higher growth rates than haploid adults. Santos and Duarte (1996) found tetrasporophytic ramets of *Gelidium sesquipedale* to be larger and less branched, while Lewis and Lanker (2004) found differences in branch angle and a tendency for dichotomic vs proliferative primary branches in *Ceramium codicola*. Carrington et al. (2001) found that the gelling kappa-type carrageenans of the gametophytes' distal tissue of *Chondrus crispus* make them stronger, more extensible and stiffer than the tetrasporophytes' distal tissue composed of weaker nongelling lambda-type carrageenans. They further argue that the kappa-type carrageenans may also render gametophytes more resistant to desiccation and herbivory. Thornber et al. (2006) tested for herbivore selectivity among tissue types of the isomorphic red alga *Mazzaella flaccida* and found important differential grazing by snails for gametophyte reproductive tissue over other tissue types.

The examples above may be evidence of the conditional differentiation Hughes and Otto (2001) predicted for the stability and evolution of biphasic life cycles. The occurrence of such features is probably the answer to why it has been observed a change in ploidy dominance from wave protected to wave exposed sites or with the intertidal

height of the sites (Engel et al 2001, Mudge and Scrosati 2003, Scrosati and Mudge 2004a and b), or to why Dyck and deWreede (2006) found that haploids of *Mazzaella splendens* dominate in the summer in wave sheltered sites whereas diploids dominate in wave exposed sites or everywhere in the winter. These later examples are evidence of a spatial or temporal niche differentiation most probably driven by conditional differentiation. Unfortunately, it was never assessed how the morphological and/or physiological differences effectively affected the vital rates of both ploidy phases nor how differences in the vital rates affect the haploid to diploid ratio of abundances (H:D) and its dynamics. It needs to be tested and proven the cause-effect chain that links from differential morphological and physiological adaptations of ploidy phases to dissimilar vital rates between ploidy phases to niche differentiation between ploidy phases and finally to the uneven H:D field observations. It is precisely upon the latter links that rely the objectives of the present thesis. Assuming that an uneven H:D reflects a partition of niches driven by conditional differentiation, the vital rates that affect most the H:D are simultaneously expected to have the greater potential to set a niche separation, to reflect an efficient conditional differentiation and ultimately the conditions for the stability and evolution of biphasic life cycles. Therefore, in the first chapter was estimated the potential for the dissimilarities between ploidy phases in each type of vital rate to impose an H:D. It enabled to determine where it is more likely for conditional differentiation to be found. This was done with a spatially adimensional model solved for steady state. In the second chapter it was estimated the potential of the several types of ploidy dissimilarities to yield an H:D geographical variability. This is the variability of the H:D over the many distinct populations of an algal species, subject to different environmental conditions. This inter-population H:D variability was documented by Engel et al (2001) and Thornber and Gaines (2003). So, the second chapter was also an attempt to validate the findings from the first chapter. In the third chapter it was assessed how the population structure, and in particular the H:D, responds to punctual episodes of instability, even if the environment tends to be stable over time. Being ahead aware of such feature is essential to proceed to the later step of understanding the adaptations of biphasic life-cycles to seasonal environments. In this context, this preliminary analysis on temporal changes is fundamental as it enables to determine whether a certain pattern of dissimilarities between ploidy phases may cause a temporal shift on the H:D that matches the temporal shift required by the environment. Temporal shifts in the H:D were documented by Gonzalez and Meneses (1996), Engel et al

(2001), Servi re_Zaragosa and Scrosati (2002), Thornber and Gaines (2003) and Dyck and DeWreede (2006). Finally, in the forth chapter it was accessed how a spatially high resolution variability may affect the space and time intra-population H:D. The spatial scale addressed in this chapter was small and intended to reflect possible intra-population environmental ranges as differences in the intertidal height, degree of wave exposure, distance to fresh water inputs or distance from shore. Such short scale intra-population H:D variability was documented by Lindgren et al (1998), Mudge and Scrosati (2003), Scrosati and Mudge (2004b) and Dyck and DeWreede (2006).

During its execution this thesis was criticized on the account of the models being ramet based when supposedly should be genet based. It is felt it is due a clarification on the subject ahead of the reading of the thesis. Macroscopic marine primary producers often possess the ability to give rise to new fronds that grow from a previously existing holdfast. All fronds from the same holdfast are then genetically identical. Considering the concept of genet and ramet introduced by Harper (1980), if an individual is considered as a genetic unit (a genet), the clonal production of new fronds increases individual sizes but not population numbers. On the contrary, if an individual is considered a frond (a ramet), the clonal production of new fronds increases population numbers and not individual sizes. However, clonality may also occur at the spore or zygote stages (by parthenogenesis), giving rise to physically separated holdfasts and ramets, nevertheless with the same genetic code. For this reason Scrosati (2002) updated the concept of genet to admit for clonal replication of new genets in the spores and zygotes. Genet vs ramet based models have been, and still are, something of a controversial matter in plant demography, ecology and evolution. Hughes and Otto (2001) work on the conditions for biphasic life cycles to be an evolutionary stable strategy (ESS) did not consider the ability of growing new fronds from the same holdfast. So, considering Scrosati's (2002) definition of genet, Hughes and Otto (2001) genetic model was simultaneously ramet and genet based. Nevertheless, when studying the evolution of biphasic life cycles, or any other life cycles or strategies, the first instinct of a biologist would probably be to focus on genet based models as it is more easily associated to the fundamental genetic component of the models. However, it may be a big mistake to follow that reasoning. Evolution occurs because the more fit prevail while the less fit succumb when competing for resources. Taking competition out of the equation leads to no evolution. A genet based model mistakenly considers that a holdfast with many ramets consumes exactly the same amount of resources and has the

exact same competitive strength of a holdfast with only one ramet. It neglects that fronds with different sizes consume different amounts of resources. It considers any herbivore gets equally satisfied by eating one or ten ramets, as long as they are all from the same genet. And so it implies that the needs of heterotrophic organisms are to consume genetic information and not biomass. It further neglects genets may alter their competitive ability by clonally producing more ramets. In fact, upon careful thought, it is hard to think of anything more biased and misleading than a genet based model to study the evolution of life cycles that incorporate the clonal replication of other stages besides spores and zygotes. Therefore, although it is unquestionable that a study about stability and evolution of life cycles must rely in models that output genet proportions, the fundamental model units that support the model building and mechanics must be ramets. Such models accomplish this by using ramets that never lose track of their genetic identity. The reason above justifies why during the present thesis it was chosen to always follow the ramet based approach. Still, there is another strong reason that concerns comparison with published reports on field observations. Often, in those reports, it is not clear whether samplers were concerned with not sampling ramets from the same genet, which would be essential for a genet based H:D discussion.