Role of whole genome duplication in tinkering process

Charles Darwin fought against the argument of perfect creation in his works. Jacob suggested that natural selection (NS) operates as an improvizing tinkerer rather than a planned engineer. It is now realized that NS in fact operates as a tinkerer to express evolutionary novelties. Evolution, like a tinkerer, is endlessly improvizing structural complexity and is far from perfection. However, the result is always unsatisfactory.

The core precept of Darwinism hinges on the availability of variations in the genome on which NS would tinker for the best possible outcome, resulting in evolutionary novelty. Apart from point mutations, recombination, gene duplications, etc., duplication of the entire genome has also now been identified as a potent source of variations, having great potentialities in yielding evolutionary novelty. The fundamental role of gene duplication in the origin of phenotypic diversity attracted Ohno[1], who postulated that it creates a superfluous locus that is free to accumulate otherwise 'forbidden' mutations as long as the original copy still performs the essential ancestral tasks. Whole genome duplication (WGD) has already been implicated to play a vital role in speciation for diversified orders of organisms. Dehal and Boore[2] provided evidence that two rounds of WGD occurred in early vertebrate evolution, which has resulted in increased complexity and genome size of vertebrates. Supportive data to this hypothesis include observation of 4 : 1 ratio of few gene family clusters in vertebrates compared to invertebrates. Aury *et al.*[3] have identified the occurrence of WGD in *Paramoecium* and have found over-retention of signalling molecules and transcription factors apart from the genes performing several very basic cellular processes, resulting in relative enhancement of these gene activities. According to Aury *et al.*[3], speciation here has been a neutral consequence to WGD and may not imply evolutionary advantage. Recent evidences on the effects of WGD on functional novelty in *Arabidopsis* come from the work by Spillane *et al.*[4]. Together with *SWN* and *Curly Leaf* (*CLF*), *MEA* forms a family of enhancer of zeste-related proteins [*E*(*z*)] like genes in *Arabidopsis*. Study indicates a duplication separating *CLF* and the common ancestor of *SWN* and *MEA* before divergence of monocotyledons and dicotyledons ~200 myr ago. A recent study has pointed out that imprinted gene *MEA*, is responsible for seed development and has originated during whole genome duplication ~35–85 myr ago from its paralogue *SWN*, which interestingly is not imprinted as it has retained the ancestral function of the precursor gene. Post-duplication, *MEA* has undergone positive Darwinian selection consistent with neo-functionalization and the parental conflict theory. It has been shown that WGD occurred even before the radiation of teleost in ray-finned fish[5]. Documented evidences of WGD are still very few because it is quite possible that WGD might have occurred at several times during evolution in several lineages, all of which have not yet been detected. In most cases, it is found that speciation explosion followed WGD.

At this point it would be pertinent to take a cautious approach in emphasizing the effect of WGD. Sudden duplication of the entire genome, in fact, leads to an inherently unstable state[6], resulting in several sorts of genic imbalances that might not always be congenial to NS. But in several other cases, WGD has been tolerated and maintained by NS. This establishes that WGD provides the requisite raw materials for NS to tinker with where, apart from evolutionary novelties, hypothetically, extinction of lineages also took place. It is probable that many of the speciation explosions during animal evolution (like Cambrian Explosion) might be a result of WGD. Hence WGD always favours tinkering, whereas NS determines the outcome. Any event of WGD will invariably throw up two possible initial outcomes to be selected by NS, in the form of either retention or rejection of duplicate copies of genes (Figure 1). Following retention the two copies, as evidences point out, are subjected to differential rates of molecular evolution to attain functional novelty through neo/sub-functionalization of, arguably, the duplicate copy. Here the debatable point is, which molecular mechanism will discriminate the duplicate copy against the original one to be subjected to novelty, while retaining the older function through the original copy[7]? Presently, the answer is unknown. Loss/retention of gene duplicates has been shown to be independent in differ-

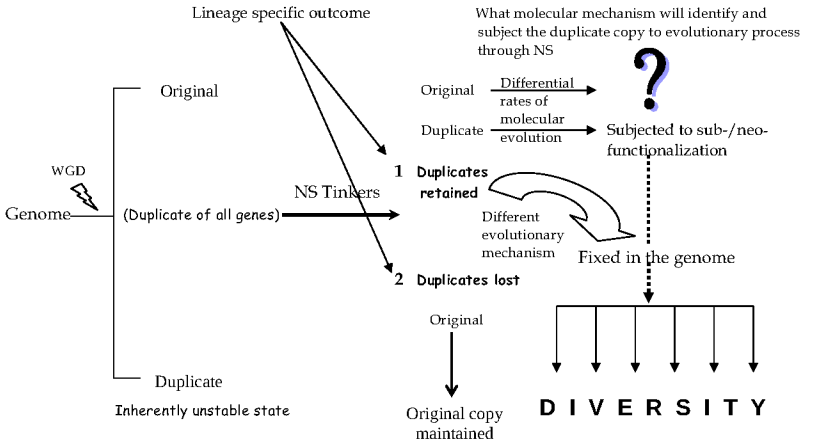

Figure 1. Possible mechanism by which WGD is expected to assist the evolutionary process. For a single event of WGD, initial unstable state of sudden genome duplication is either tolerated for or against by NS, marking the initial scope for tinkering. Where selected for, the next tinkering takes place in either retention or rejection of duplicates in a lineage-specific manner. The retained copies are subjected to asymmetric rates of molecular evolution leading to neo/sub functionalization of arguably the duplicate copy. The question mark in the figure points to the argument about which molecular/evolutionary process would discriminate the duplicate copy from that of the original to be subjected to attain novel function. Presently it is unknown. The retained duplicate should be subsequently fixed in the genome, probably by a different mechanism from that of the initial duplicate retention, leading to diversity and subsequent speciation.

ent populations of the same lineage. Thus NS has ample scope in improvizing the evolutionary output. Such an inclined selective force is directed towards species diversity through attainment of functional novelty. Retained gene duplicates have undergone diverse evolutionary fates in subsequent post-WGD lineages. This is suggestive of selection pressure acting on gene duplicates (Figure 1). WGD exhibits a tendency to reoccur in lineages where NS initially tolerated it. This subjects the concerned genome through cycles of gene loss and retention with each event of the WGD[3]. Hence in addition to the initial retention of duplicates, another important aspect is the subsequent stabilization/fixation of the duplicates in the genome. Most likely a different evolutionary mechanism operates for initial retention of duplicate copies and the fixation of duplicate copies in the genome, leading to functional novelty (Figure 1).

Occurrence of WGD in some species of plants leading to polyploidization has also been recognized to have evolutionary advantages. The phenomenon of WGD demands high-energy investment for duplication as well as maintenance of the superfluous genome. From the perspective of evolution, it can be argued that there should always be a 'cost-benefit ratio' guiding the persistence or abandonment of any biological event. The living system will not persist with a structure and/or function that is not beneficial. Why should WGD then be consistently persisted with within the living system, if it does not give it any 'benefit'? WGD in fact is sort of a handicap to an organism. The benefit should be in having a selective advantage of WGD that provides abundant spare repertoire of genomes for NS to tinker with, resulting in asymmetric rates of molecular evolution for any two duplicate gene copies. It might be argued as to why we call WGD as 'advantageous'? Early in evolution WGD occurred in several lineages, followed by either its selection leading to speciation in some cases, or causes of extinction in others by NS. However, where it was selected for, WGD continued to occur leading to speciation events over an elaborate timescale, thus implicating its evolutionary advantage. This also shows that irrespective of the outcome, WGD provides a source of tinkering. The evolutionary advantage of WGD over single/repeated segmental gene duplication is that more tinkering opportunities are created in the former case. The emergence of structural and functional novelty is usually gradual and slow, progressively increasing the complexity and diversity of new function and of new species. Hence it might be assumed that tinkering is useful for both microevolution as well as macroevolution.

Introducing the concept of 'tool-kit' and 'house-keeping' genes would provide further options regarding the various intricacies of evolutionary processes[8]. It has been shown that with the increase in complexity, the percentage of protein coding gene actually decreases; an observation that emphasizes the increased role of non-coding regions of the genome in establishing complexity. The tool-kit gene set must include non-coding genes also apart from signalling molecules and transcription factors. As a result, WGD would lead to extra copies of both housekeeping genes as well as tool-kit genes for NS to tinker with, optimized to afford speciation. We hypothesize that retention followed by fixation of duplicates of tool-kit genes due to WGD, has the capability to exponentially increase phenotypic diversity, thereby providing enhanced opportunities for tinkering and thus leading to evolutionary novelty. Support to our hypothesis comes from the work of Hittinger and Carroll[9], who have dissected the evolution and divergence of the paralogues GAL_1 and GAL_3, involved in the genetic switch controlling the yeast galactose usage pathway, from a bifunctional ancestral gene. They have found such divergence of function to be a result of complete sub-functionalization of the promoter region rather than that of the structural coding region. According to Carroll[10], 'Evolution of form is very much a matter of teaching very old genes new tricks'. He also states that 'the expansion of the tool-kit correlates with increased animal complexity, but not with diversity'. Apparently the statement is contradictory. It is true that diversity is measured through phenotypic expression. It may be argued that increasing complexity, as a result of WGD, may not always have phenotypic manifestations.

To summarize the contribution of WGD in the tinkering process, it can be assumed that WGD is a handicap and starts its journey in a streetcar. During the journey some duplicates leave (loss) the streetcar, and some change their attitudes (functions) with their original copies (sub- or neo-functionalization), resulting in unplanned combination of duplicates, allowing more opportunities for NS to tinker with in promoting evolutionary novelties.

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