Electrophoretic and molecular hybridization studies indicated that the genomes of the rainbow and brown trout code for eight electrophoretically distinct types of lactate dehydrogenase (LDH) subunits. The interactions of these subunits in vivo produce 18 isozymes which are naturally arranged into four groups. A tissue-specific distribution of the isozyme groups was observed. (The SCI indicates that this paper has been cited over 85 times since 1968.)

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"By the time I joined Markert in 1963, he had an active interest in the genetic basis and evolutionary significance of piscine isoenzyme systems. I believe that our initial interest in salmonid isozyme systems stemmed from the report of Tsuyuki and Gadd of the existence of 12-16 electrophoretic species of hemoglobin molecules in the sockeye salmon and rainbow trout. Our interest was further stimulated by the report of Hochacha of the existence of as many as 14 isozymes of LDH in the tissues of the brook trout, lake trout, and their hybrid, the splake, and that of Lebherz and Rutter of multiple isozymic forms of glyceraldehyde-3-phosphate dehydrogenase in rainbow trout tissues. With this background and the hypothesis of Ohno et al. that salmonids are tetraploid species, we focused on defining the molecular basis of the LDH isozyme patterns of fish. With this background and the hypothesis of Ohno et al. that salmonids are tetraploid species, we focused on defining the molecular basis of the LDH isozymes of the Salmonidae employing the rainbow trout as a model system.

"Electrophoresis resolved the LDH isozymes of the rainbow trout into four distinct tissue-specific groupings which we designated a, b, d, and e. The group designated a was comprised of the least negatively charged isozymes while group e was comprised of the most negatively charged molecular species. Groups a, b, and d each contained five equally spaced isozymes while group e appeared to contain only three isozymes. The group e isozymes were found only in the neural and pigmented retinas and vitreous humor and there was some indication that they might occur exclusively in the neural retina. Likewise, the group a isozymes were restricted almost exclusively to skeletal muscle.

"Of primary importance in determining the interrelationships among the multiple molecular forms of trout LDH was the establishment of the molecular composition of each group of isozymes. Employing the technique of molecular hybridization, the fastest and slowest migrating molecular species of each isozyme group were observed to undergo random subunit reassociation to form the three intermediate heteropolymers characteristic of their series. Thus, each group was a family of isozymes composed of two types of parental heterotetramers and three heterotetramers of intermediate electrophoretic mobility. From intraspecific, intergroup and interspecific hybridizations, it was concluded that each group of isozymes possessed distinctive physicochemical properties which differentiated it from all other groups. And, as many as eight different LDH subunits appeared to be encoded within the trout genome.

"There was a reasonable body of evidence supporting the Ohno hypothesis of the tetraploid nature of salmonids. However, it was not clear whether the progenitor might have been an autotetraploid purebred ancestor or an allotetraploid hybrid ancestor. We favored the allotetraploid hypothesis because of the rarity of mutant alleles at LDH loci and because it was conceivable that hybrid cells might require the products of both parental genomes in order to function effectively.

"Perhaps this paper has been cited frequently because the observations we reported have withstood the test of time."

1. Markert C L & Faulhaber J. Lactate dehydrogenase isozyme patterns of fish.
2. Tsuyuki H & Gadd R E A. The multiple hemoglobins of some members of the Salmonidae family.