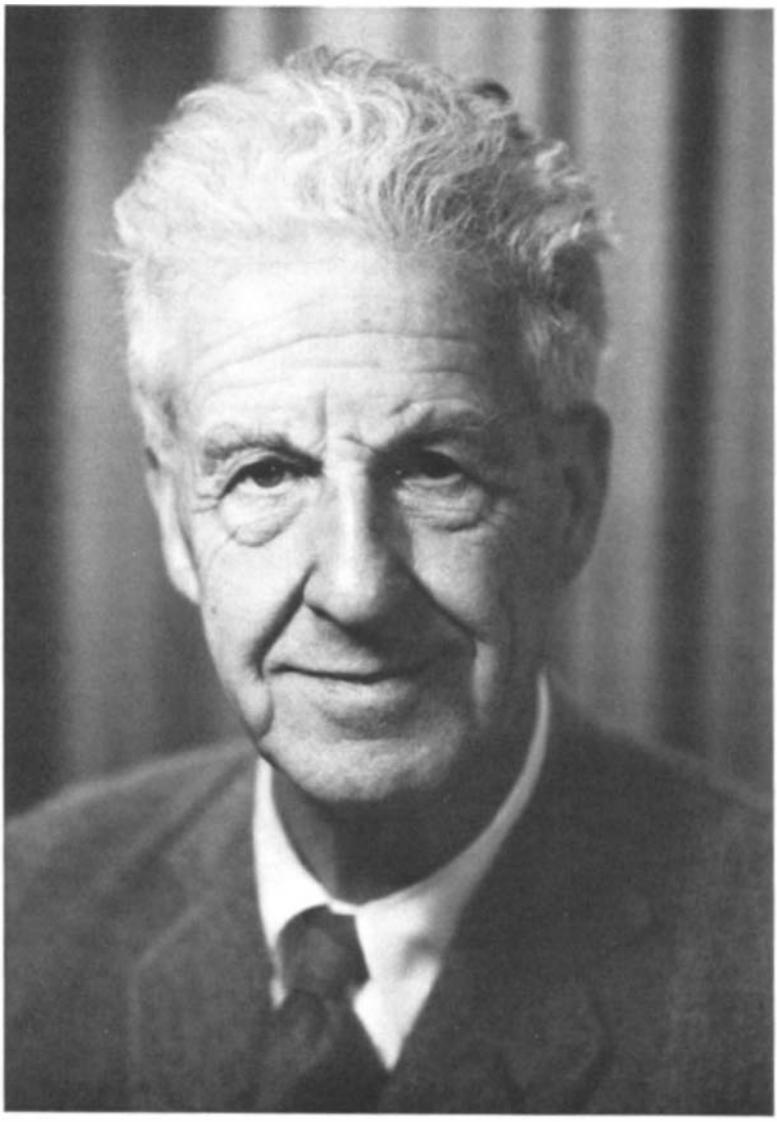

L. C. Dunn
1893–1974

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# L. C. DUNN AND HIS CONTRIBUTION TO T-LOCUS GENETICS

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Leslie Clarence Dunn, who died on March 19, 1974, was a geneticist's geneticist and a biologist's biologist. He had an ingrained awareness of all of the different things that we call genetics and a strong feeling that genetics forged links between all matters of biological interest. This quality made it possible for him to talk on equal terms with almost anyone interested in a biological problem of any kind—this applied both to neighbors bothered by their inability to grow tasty tomatoes in their backyard and to colleagues concerned about genetic bases for racial discrimination. He was fascinated by what we call genetic variation, in whatever form it manifested itself, and I think this made him a true naturalist; he was equally interested in man, mouse, and garden flowers in the sense that they presented interesting biological problems with genetic correlates that were just as real to him although their overall importance might be very different. This catholicism is reflected in the biological material that he worked with during his professional career; it ranged from mice to chickens to man and Drosophila, and even included, after retirement, an excursion into morning glory genetics. To some extent his own broad interests dissatisfied him, because he felt they made him an amateur in everything and a professional in nothing. Clearly, this was not so; what he considered amateurism was partly a reflection of his ability to use genetics as a connecting link among the biological sciences in general.

Dunn started in science because he was interested in natural history, and he pursued it for the same reason. The fact that he got paid for doing it seemed like a lucky accident to him, and the fact that his work brought him eminence as a scientist meant little to him. He certainly never sought importance or position, he never played politics, and he seemed completely free from pretense of any kind. As his student and later his colleague, the most important message I took from him was to follow science first, to do my job as I thought right, and never to worry about

whether the result would be "interesting," "topical," "grant-worthy," or a step toward personal advancement. This attitude no doubt reflected the fact that Dunn started in science in a time that was quieter and gentler than now, when it was easier to do science single-handedly without serious worries about either competition or support for elaborate and expensive laboratories; but it also reflected standards of integrity and honesty that were rarely matched then or now. This attitude is fortunately infectious; all of my own first students of course knew Dunn, and he took a personal interest in them as scientific grandchildren. They heard his message and still persist in being almost cantankerously obstinate in doing science for the sake of science alone, avoiding the kind of expediencies that might have tainted them in his eyes. And now they tell their own students, Dunn's great-grandchildren, who never knew him, to follow the same principles. This is one of Dunn's legacies that I think is as important as his contributions to knowledge.

It is the story of these contributions, however, that constitutes the bulk of this memoir. The development of genetics has been so rapid that it comes as a surprise to realize that Dunn's initial entry into biology occurred less than 10 years after the rediscovery of Mendel's principles. Thus, as a student he was exposed to these ideas as something excitingly new that required general confirmation and testing. Probably because of his naturalistic inclination, his bent seemed to lead him always to tackle these new problems at the level of the whole organism and particularly with reference to development and evolution.

When Dunn entered Dartmouth College in 1911, he already considered himself a biologist with good theoretical and practical training. From early childhood he had spent Saturday and Sunday afternoons working with his favorite person, his grandmother, in her garden, and had learned the satisfaction of steady application to simple tasks that gave insight into living things. A high school teacher, Dr. Marie Walcott, had amplified his love for gardens into an interest in taxonomic botany and through that, had given him an awareness of the powerful beauty of science. Dunn's high school biology notebook was apparently such an impressive document that Dartmouth excused him from freshman biology courses, and he began an intensive study of botany. By the time he was a junior, though, Dunn had found Punnett's *Mendelism* and was apparently immediately fascinated with the exactness of the rules governing the behavior of hereditary factors, since until then most of his experience had been with the descriptive aspects of biology. During the winter of the same year, Morgan's *Heredity and Sex* appeared; this captured Dunn's interest entirely, and his path into genetics was fixed. He goaded his major professor, Professor John H. Gerould, and other graduate students into organizing a seminar to discuss Punnett's and Morgan's books and to read some of the papers on genetics that were appearing in the journals. In the Easter recess of that year Dunn went to New York to see Morgan at Columbia, to inquire whether there was any prospect of working in his laboratory either before or after completing his college degree. But Morgan was apparently unimpressed, perhaps even dismayed, by the young botanist's ignorance of systematic zoology and lack of any solid training in zoology. At any rate he held out no good hope, since his space was already overcrowded and his few posts already committed to Columbia graduate students.

Disappointed, Dunn returned to Dartmouth to try to remedy his zoological defects, and to look for a geneticist who would accept him as a graduate student. In time he made an arrangement with W. E. Castle, who, since 1901, had been engaged in testing the ideas promulgated by Mendel in both the new laboratory animal, Drosophila, and mammals.

Dunn's research activities with Castle seem to have been mostly of the search and explore type; this presumably represented attempts to become comfortable with a new way of thinking, as well as of generating and analyzing data. He bred mice and defined some coat-color traits, he classified different types of spotting variants, he studied segregation ratios. But the only real questions they asked proved to be red herrings. Briefly, they tried to test the hypotheses that linkage relations between genes could be altered by selection, and that genes themselves could be changed in a directed way. These were quite reasonable questions for the time since genes were largely being investigated as statistical units in segregation analyses, and little or nothing was known about what they were composed of, how they were associated, or how they controlled phenotypic traits. So it seemed reasonable to Dunn and Castle that selection might well enhance the degree to which certain markers traveled together, and Dunn set out to demonstrate this notion in Drosophila. Unfortunately, Morgan's group was also testing the same ill-fated hypothesis, and they published while Dunn's work was still in progress. He shifted then, but apparently with little real enthusiasm, to a similar study of linked genes in both mice and rats.

This work was disturbed by restlessness aroused by the worsening, in late 1916, of the First World War; at that time Dunn joined the Harvard regiment as a volunteer to receive military instruction from French and British officers. When the United States entered the war, Dunn's university work ceased entirely and he went to officer's training camp and then to France. When he returned in 1919 he completed his dissertation and made arrangements to begin work as a geneticist at the Storrs Agricultural Experiment Station in Storrs, Connecticut.

There, he was the only member of the small station staff who was not also a member of the staff of the Connecticut Agricultural College which then had about 350 students. The director of the Storrs Agricultural Experiment Station was Dr. Edward H. Jenkins, a chemist trained at Yale University who was also the director of the Connecticut Agricultural Experiment Station at New Haven. That experiment station was the first in the United States (founded in 1877) to be devoted only to testing and research. Dr. Jenkins hoped to develop a similar research program at Storrs, and wanted Dunn to devote himself entirely to research in genetics, especially as applied to poultry breeding. The freedom to develop his own program in the new area of genetics, just exactly as he wished, was the motivating factor that took Dunn to Storrs.

At Storrs, Dunn embarked on an extensive program of inbreeding by brother-sister matings, and on a study of the factors influencing "hatchability" of eggs, that is, a study of embryonic mortality. The inbreeding experiments, stimulated by the study of inbreeding in corn at the New Haven Experimental Station, were intended to measure changes in various elements of vigor, in morphological characters, and in egg production, as well as to produce inbred lines of chickens that at that time

did not exist. The results, published mostly in technical journals and bulletins, were not of great consequence, since most "inbred" lines died out after two generations or so. A few families survived and were turned over to Henry Wallace where they formed foundation stock for the Hybred Corn Company's introduction of crossbred chickens.

Again, this was a period when Dunn did a good deal of seemingly superficial work that nevertheless gained him a wide knowledge of the practice and the theory of genetics in a familiar species. He studied plumage colors, egg production, egg characteristics, and growth rates; almost every measurable characteristic of a chicken was at one time or another measured and related to specific genes or genetic background. By 1924, Dunn began to home in on the subject matter that was to form the basis for much of his future work, namely lethal factors in embryos. He became especially interested in so-called parrot-beak embryos, which provided the first analyzed cases of chondrodystrophy in birds. In collaboration with Walter Landauer, who provided a thorough study of the anatomy and development of chondrodystrophic embryos, it was demonstrated that virtually identical chondrodystrophic phenotypes could be due to heredity or to purely nongenetic causes. They found the same two sets of factors to operate in causing the condition known as rumplessness in fowls to be a gene mutation in some cases and nongenetic alterations in development in others. This collaboration not only resulted in a lifelong friendship between the two men but, interestingly, established a kind of reciprocal approach to problems of development that each was to follow through the rest of his career. Dunn chose what he thought was the simplest path, namely the analysis of single-gene mutations that produced developmental aberrations that should be easy to analyze. Landauer elected to deal with all the complexities of nongenetic variation, which led eventually to the development of the field of chemical teratology.

Dunn's course diverged briefly in 1927 from his growing interest in developmental genetics. On his first sabbatical leave he worked for a time at the Naples Zoological Station on invertebrate development without finding an interesting problem. Then he spent some months at Edinburgh with Frank Crew where he began observations on selective fertilization in fowl and also returned to the mouse to study an enzyme step (dopa-oxydase) in the determination of spotting patterns. The latter problem proved interesting to him, and he went to Berlin to pursue it further with Richard Goldschmidt at the Kaiser Wilhelm Institute for Biology. No significant advances in research were made during that stay, but Dunn was strongly influenced by the atmosphere of scientific attachment to fundamental problems that he found at the Institute, where Carl Correns (then director), Goldschmidt, Max Hartman, Otto Warburg, and others met in seminars filled with vigorous debate. This atmosphere apparently revived Dunn's memories of graduate school days and evoked both a desire to return to a more academic environment as well as the impetus to make a fresh start on problems of what he was by then calling *developmental genetics*. Goldschmidt was a leader in this new field, and had just published his *Physiologische Theorie der Vererbung* which attempted to interpret the effects of genes on development in terms of relative rates of different physiological processes. The reasoning

behind this concept came first from studies on intersexuality in the gypsy moth, where crosses between geographical races often produced intersexes. The physiological mechanism that Goldschmidt proposed was that sexuality was a genetic trait and that each individual begins life with the capacity to develop into either a male or a female. This led him to the conception of a balance mechanism for sex determination, and to proposals for such mechanisms in development generally. Dunn wanted to follow this lead because he thought such an approach might unite genetics and embryology in a mechanistic way.

The opportunity for a new start came quickly. The sabbatical year was curtailed by the sudden death of Dunn's father-in-law, and the family returned to Storrs. For his new endeavors Dunn brought with him chickens from Scotland and Germany, and stocks of mice with suspected mutant genes that he had collected from dealers and fanciers all over England. Hardly had he settled these animals in Storrs when he was approached by Gary Calkins, professor of zoology at Columbia University, to fill a vacancy in that department that arose as a result of the retirement of E. B. Wilson and the departure of T. H. Morgan, C. B. Bridges, and A. H. Sturtevant to found a new laboratory at the California Institute of Technology. This was not the first offer that Dunn had had, of course, but he had declined all others without investigation and without regrets, since he had been eager to continue research in the free and untrammeled atmosphere of his small laboratory in the country. But the Columbia offer came at just the right time; it promised a return to an academic environment, and perhaps a broader scope for the new direction that Dunn wanted to take in research. In any case he accepted what he considered a flattering offer for a full professorship in 1928.

On moving to Columbia, Dunn focused his attention on a genetical and developmental analysis of pattern formation in the mouse. The establishment of pattern is of course the essential feature of development, and Dunn thought it could be analyzed most effectively by studying its genetic control. For this purpose he returned to studies of spotting patterns in mice, with an eye to testing Goldschmidt's central themes that mutations alter the rates of processes in development and that the whole of development implies a merging of many subpatterns, all of which must be physiologically integrated with one another. But since he was returning to Columbia, which had such strong associations with Drosophila even though Morgan was gone, Dunn could not resist expanding his work to Drosophila as well. So he began to collect Drosophila mutations such as *Minute* and *Bar*, which could be interpreted as affecting developmental rates, with the thought of analyzing each of them separately, and then in combination. The Drosophila work was never as close to his heart as was the mouse work, probably because he never found the fruit fly as interesting as a mammal—and what could be examined in Drosophila could generally be done as well in the mouse; if work with the mouse was more difficult and more time consuming it was nevertheless more interesting.

In his sabbatical of 1932–1933, Dunn made one last attempt to combine work on Drosophila and on mice, going to the University of Oslo to do Drosophila experiments with Otto Mohr and to work on mice with Christina Bonnevie. The experiments with Drosophila proved the general point that there was quite a strict

proportionality between the retardation induced by various *Minute* mutations and the size of the *Lobe* eye, and therefore were supportive of Goldschmidt's ideas. This set of experiments incidentally led them to detect an apparent back mutation, in which a white-eye mutation reverted to honey, or part way to wild type. This represented a wholly new concept, because until then mutation had been thought to be a one-way process. Dunn worked hard to nail this point down, and derived from it a lasting interest in gene structure and organization as related to mutation, matters that had previously never been of great concern to him because he had felt that gene structure was far less interesting than gene function.

The mouse aspect of Dunn's work in Oslo may have been particularly significant, and foreshadowed events to come. It was his first real excursion into abnormal embryology in the mouse, and it involved a mutation that produced a short tail, the so-called Shaker Short. Bonnevie and Dunn produced only descriptive studies of this mutant, and Dunn was never satisfied that they had got a real explanation or interpretation of the defects. Nevertheless, this work probably set him up for an interest in the T-locus mutations that had just come his way before he went to Oslo; the stocks were left in the care of a young student, Paul Chesley.

The first *T* mutation was reported in 1927 by a Russian surgeon, Nelly Dobrovolskaia-Zavadskaia. A cancer researcher studying the effects of radiation on mice, she found in the offspring of an irradiated male two categories: short-tailed and normal-tailed. She proved that the short-tailed offspring carried a new dominant mutation, which she named in Greek *Brachyury*. When she bred two short-tailed animals together she was puzzled because she could never get a true-breeding short-tailed line. She made the correct assumption, but had no proof for it, that the *Brachyury* (*T*) mutation was lethal when homozygous.

Shortly after reporting on *T*, Dobrovolskaia-Zavadskaia was joined in Paris by another young Russian émigré, N. Kobozieff. The two pursued the study of *T* by crossing short-tailed animals to two other apparently normal strains of mice, one a French laboratory line, and one descended from a wild mouse that Zavadskaia had trapped while on holiday near the Spanish border. To their surprise a striking abnormality appeared in the offspring of both crosses: taillessness. They inbred the offspring from each cross and were further confounded when, this time, the abnormal phenotype bred true! At this point, Dobrovolskaia-Zavadskaia came to the United States to lecture for some Russian refugee organizations, and appeared at Dunn's laboratory at Columbia looking for a solution to her puzzle. The best solution she could see was "to give up the confusing tails and return to my proper field which is cancer research." Dunn was fascinated with the problem and answered, "If you are going to give them up, just give them to me."

The short-tailed and tailless mice arrived at Columbia in 1931. The first job was to give a thorough test to the idea that *T* was lethal in homozygotes. Zavadskaia had found some crippled short-tailed animals, partly paralyzed with hemiplegia, and she thought they might be homozygotes. But as soon as Dunn and his student Chesley began to breed the stock, they became convinced that the homozygotes were dying as embryos and that the crippled animals were extreme variants caused by modifier genes or nongenetic developmental fluctuations. The first task then was to

find a class of dying embryos that represented the expected Mendelian proportion of 25%. They found them almost immediately by dissecting pregnant females at midgestation. At 10 days of pregnancy some of the embryos had strikingly abnormal traits; instead of having truncated tails they had truncated bodies that ended abruptly just behind the forelimbs. This abnormality was then traced further back in time at half-day intervals, and Dunn and Chesley learned that they could first recognize the abnormal class at 8 days. At that time the embryology of the mouse had not been well described for that stage, so the two had to work out a schedule for normal development, and had to study the normal embryology of the mouse for comparision with their mutant. Eventually they determined that the fundamental defect in *T/T* embryos depended on a failure of notochord differentiation. Chesley's dissertation describing this work is now recognized as a classic of its kind; it was not only the first thorough study of the effects of a lethal gene in mammals, but also the first clear demonstration of genetic interference with an inductive system. It laid the basis for a whole series of studies of the effects of genes on early development in mammals.

Having confirmed Zavadskaia's suspicion that the failure of *T/+* mice to breed true was due to lethality of *T*-homozygotes, Dunn then went on to analyze the genetics of taillessness. First he tested and confirmed Zavadskaia's observation that matings between two tailless mice of the same origin did in fact produce only tailless offspring; he concentrated on the line derived from the Spanish mouse because its wild origin intrigued him. An analysis of litter size in those matings provided the clue that a balanced lethal system was operating; in other words, in crosses of short by short, where he had already shown that one category of embryos died, he had found litter size at birth reduced by one quarter. In litters from two tailless animals, the progeny size was reduced by one half. He surmised that the tailless mice were double heterozygotes for two lethals—the *Brachyury* gene that they had already defined, and a new one, apparently a recessive, which at first was merely inferred.

The main puzzle now took quite a different form, since the two lethals involved appeared to represent alleles at the same locus. There was one precedent for a balanced lethal system, described by H. J. Muller, but in that case the two lethals involved showed occasional crossing-over in heterozygotes and were clearly not allelic. Dunn's problem now was to explain why, although all the data collected indicated that the two genes in question were strict genetic alternatives and did not show crossing-over, they showed complementation and produced a viable heterozygote that carried two lethal genes at the same locus. An unprecedented situation, this made him suspect that genes—at least these genes—might be bigger things than were supposed, and that different alleles controlled different sets of processes in early development. He began to call them *pseudoalleles*, which is a term still used for this and other genetic systems whose genetic structure is not well understood.

In any case the situation quickly became more complicated. Dunn then analyzed both tailless lines that Zavadskaia had sent: Line A derived from the laboratory stock and Line 29 from the Spanish mouse. Each produced only one kind of offspring (tailless) that survived to birth, and in each the homozygous *Brachy*

mutation killed one quarter of the embryos, and in each another lethal apparently killed another quarter. But those other lethals proved to be different in the two tailless lines. This was shown when Dunn crossed Line A by Line 29 and found with gleeful surprise that a class of normal-tailed offspring was present at birth. It was quickly shown by breeding tests that these normal-tailed animals carried, as predicted, *no* wild-type alleles at the *T* locus, but rather two different lethals. Again, this raised the problem of complementation between two lethal alleles, and again Dunn suggested an explanation based on their controlling different sets of embryonic processes. This was possible to test, since if correct, one would expect the two recessives to affect the embryo in different ways, perhaps by killing it at different times or in different ways.

Just at this juncture in 1935 when it was clear that the unraveling of the T locus would require much embryological work as well as genetic analysis, Dunn was fortunate to be joined by an associate, Salome Gluecksohn-Schoenheimer, who had trained with Spemann in the classical German school of amphibian embryology. With her background she was able very efficiently to study the embryonic effects of the two different recessive lethals, and soon showed that, as suspected, the lethals had different effects when homozygous. Both of them interrupted development very early on; in one case the homozygous embryos died almost immediately after implantation, in the other the embryos failed even to implant. The main point for Dunn was that the embryological evidence confirmed the genetic hypothesis, that these were two lethals related by allelism but different in function.

The clear evidence was that the T locus contained sets of multiple alleles, whose complementary interactions suggested to Dunn a complexity on the part of genes or loci or both, and led him to want to know more about what multiple allelic systems represented. Although such systems had been recognized and studied since the early part of the century and had served to dispel the primitive "presence-absence" notion of the relationship of a Mendelian dominant to its recessive counterpart, little was known about them except that, as could be read from Curt Stern's (1930) *Multiple Allelie*, there were no general rules. Dunn strongly suspected that T-locus mutations were not alleles in the normal sense, but that they served as variants within a region that had some structural and functional unity. He set out to dissect that region by collecting more mutations for embryological and recombinational analysis in attempts to get an estimate of the total variation it contained. This point will be returned to later.

While the pure genetics of his system remained a major concern to Dunn from the 1930s on, he was necessarily caught up in another strange functional aspect of the T locus. His very first breeding analyses of tailless mice had turned up a fact that was no less than astonishing. Male tailless mice did not obey Mendel's rules. In matings to wild-type females, males of the genotype Dunn symbolized as $T/t$ usually produced 80% or more of normal tailed ($+/t$) offspring, instead of the 1 normal:1 short ratio expected. Females, on the other hand, behaved normally. The first question that needed asking was whether this was due to a defect in transmission of $T$ or a superiority on the part of $t$. $T$ was already known to segregate normally from $T/+$ males, and Dunn soon showed that in $t/+$ males the recessive lethal gene was transmitted in the same high ratios as from $T/t$ males. This was

an especially interesting point, since it meant, apparently, that a *lethal* mutation had some clear advantage over its normal allele. In his characteristic way Dunn systematically eliminated any trivial explanations he could think of; he showed that the effect was truly genotypic and not phenotypic and ruled out possibilities of modifier genes, differential viability, and errors in classification. He was also very perspicacious in calling the phenomenon distortion of *transmission* rather than *segregation* because he did not want to limit himself to thinking of meiotic disturbances; since transmission was actually all that he was measuring, he wanted to leave entirely open the question of mechanism. In any case it was clear that he had on his hands a major departure from conventional genetics, for which no explanation could be offered, but nevertheless a departure that would have serious implications for evolutionary theory. It was not realized at the time, nor in fact is it even widely appreciated today, how important an evolutionary force nonrandom transmission of gametes can be. As has been shown by R. C. Lewontin, a student of Th. Dobzhansky and a colleague of Dunn who became interested in this question during his stay at Columbia, even minor transmission distortions represent a potential selective advantage well able to swamp the obvious and well-known selective pressures exerted on deleterious genes.

This implication of this new evolutionary force by no means evaded Dunn, especially since he remembered that one of his original lethal *t* alleles had come from a wild Spanish mouse. It occurred to him right away that the unlikely existence of a lethal gene in a wild population might be due to the protective influence of a high transmission of that allele through males. The next obvious step was to find out whether any other wild mouse populations carried such peculiar alleles. In the laboratory of Howard Schneider at the Rockefeller Institute, Dunn found a population of mice that had been derived some five years earlier from wild ancestors trapped in New York and in Philadelphia. He tested them by matings to *Brachy* heterozygotes and the first few litters proved his point; heterozygosity for *t* was present. Testing of a large number of mice showed that the proportion of heterozygotes was surprisingly high, in the neighborhood of 50%. Dunn derived several tailless lines from inbreeding and made another astonishing observation; the population contained not just one but two different *t* mutations, one lethal and one semiviable. The semiviable produced complete sterility in homozygous males, so in an evolutionary sense it was also lethal. Both alleles had, as was expected, the very high transmission ratios necessary to maintain them in the population.

This intriguing situation led Dunn into mouse population genetics both as practiced in the field and as simulated by mathematical formulae and computers. He managed to collect mice from a good geographic spread of North America, from Japan, and from Europe. In virtually all populations that were adequately sampled there were lethal or semilethal *t* alleles (never viables) with high transmission ratios, and heterozygotes in high frequencies that often approached 50%. There was no question that *Mus* as a genus was polymorphic for *t* mutations and therefore in some not understood way these genes had to be considered part of the normal genome of the mouse. Dunn and his students, T. Prout and D. Bruck, began to try to reconcile the two selective factors that had to be involved, namely embryonic mortality and transmission advantage. Applying Hardy-Weinberg statistics that

took both factors into account, they came to a conclusion Dunn found rather absurd. Nor only *should* most wild populations have *t* mutations but they should also have them in even *higher* proportions than were actually found; in fact, the alleles should come close to fixation, that is, 90–100% heterozygotes. Since Dunn felt that data properly collected from natural populations could not lie, and his populations had only 50% or so heterozygotes at most, he looked for what was lacking in Hardy-Weinberg statistics. The obvious parameter to suspect was population size, which of course in Hardy-Weinberg calculations is infinite. Not much was known about population structure in wild mice at the time, but Dunn's "amateur" naturalism led him to suspect that they probably lived in very small mating units, e.g. one's kitchen or someone else's barn. It proved too difficult to analyze the real situation quickly or accurately, so with the computer-oriented Lewontin, filched temporarily from Dunn's colleague Theodosius Dobzhansky, computer models were built to simulate stochastic mating processes in small populations. They showed that in small populations of no more than 10 individuals there were indeed restrictions on the degree of heterozygosity the population could attain without running too serious a risk of extinguishing itself by fixing the mutant allele. In fact, when a starter population containing *t* mutations in any frequency was allowed to produce more than a couple of hundred generations it virtually always died out. But, since real wild populations did nevertheless nearly all have *t* alleles, this suggested that the model was again falling short of reality. The implication was clear nevertheless that mice do live in small populations, and this has since been measured and confirmed. The element Dunn thought most likely missing from the stochastic model was migration from one small deme to the next, most probably in the form of a wandering male carrying with him a *t* allele with an infectiously high transmission ratio.

To return now to the purely genetic aspects of the T locus, it must be pointed out that in the course of maintaining the two original tailless lines, which were thought to be true-breeding, it became clear that not even this rule was to be followed in its entirety. The balanced lethal system broke down every now and then, and about one in every 500-1000 offspring was not tailless, but completely normal. These "exceptions" as Dunn called them were put through breeding analysis and proved, like complementing Line A/Line 29 double heterozygotes, to carry two different recessive *t* mutations. But in the first few cases analyzed, one of the recessive genes was different from any so far studied; it was viable when homozygous but nevertheless interacted with *T* to give a tailless phenotype, and thereby, by definition, was a mutation at the same locus. Since Dunn by then had no evidence from extensive data on the breeding of $T/+ \times T/+$ that *T* or + mutated to *t* alleles, his interpretation was that the recessive lethal *t* mutations in the tailless stock had undergone further mutation to a viable state. Later, it was shown that a comparable process of mutation could give rise also to new lethals. The constant generation of new *t* mutations from preexisting ones produced a plethora of material to be sorted out and categorized as lethal or viable, and measured as to transmission ratio. As a general rule it appeared that viables had also changed with respect to their effects on transmission ratio; if they affected it at all, and some did not, they gave abnormally low

ratios, thus producing yet another puzzle. Further breeding tests were done to make sure that the new class of *t* mutations comprised also genetic alleles of *t* lethals as well as *T*, and again no recombination occurred in any combination tested.

At any rate, Dunn began to suspect that he might be dealing not with a conventional *gene*, but with a *region* of genetic material. He thereupon began a search for other genes—particularly tail mutations—that might map in that same region. He systematically tested for linkage with *T* any mutation that turned up in his lab, any that he could collect from other labs, and any that he could buy from fanciers. The first linked marker (*Kink*) that he found was obtained from "The Sunshine Mousery of Manetee, Florida, The Home of America's Fanciest Mice." The marker, which came as a pleiotropic effect associated with the waltzing behavior that interested the fanciers, provided new insights in two rather different ways.

First, linkage estimates with *Kink* revealed still another genetic peculiarity associated with *t* mutations. Whereas the initial studies done with *T* alone showed the *T-Kink* distance to be about six units, experiments done with *T* and *Kink* on one chromosome and a lethal *t* mutation on the other immediately revealed that *t* mutations suppressed recombination over the whole distance between the markers. This of course raised further doubts about the allelism of *T* and *t*; since the genetic definition of alleles is that they show no recombination, it was patently impossible to test for, or argue, allelism in the case of a gene that prohibits recombination. For example, although *Kink* and *t* showed no functional interaction at all, they also behaved as genetic alleles in segregation analysis. The recombination suppression associated with lethal *t* alleles of course raised further questions as to whether they were induced chromosome aberrations rather than mutations. Dunn investigated this point cytologically as well as he could at the time, but found no evidence for either inversion or duplication-deficiency changes, nor have any been found with the increasingly sensitive karyological methods available today. Linkage studies with *Kink* revealed another difference between viable and lethal *t* mutations; viables did not act as crossover suppressors. This provided one reassuring fact; at least the allelism claimed for *T* and viable *t*'s could be assumed to stand, and perhaps by analogy the case for allelism of lethal *t*'s was strengthened.

The second important implication that Dunn saw in the linkage of *T* to *Kink* and then to a second tail mutation, *Fused*, was that the spatial juxtaposition of these loci on the chromosome and their similarity of effects might not be fortuitous. This was quite the opposite view from that generally held, since Drosophila geneticists had by and large not discovered any clusters of different genes with related functions. In fact, the (1945) paper by Dunn and Ernst Caspari, "Neighboring Loci with Similar Effect on Development," may have been the first expression of that idea—that genes are not distributed randomly on chromosomes. Dunn was impressed later by the suggestion by E. B. Lewis that such situations might well arise from tandem duplications followed by mutational divergence; he thought that this was an obvious mechanism for increasing gene number as well as enhancing the sophistication of genetic regulatory systems. It is interesting that the same ideas, and even some data to support them, are now being applied to the same chromosome in the mouse, but in terms of genes determining cell surface antigens, not tails.

*Kink* was a marker suitable for these first analyses of recombination, but it was difficult to work with over the long haul, since two mutations affecting the adult phenotype of the tail were necessarily not always easy to discriminate. The situation was much improved when Mary Lyon discovered, and sent to Dunn, another marker in the region of *T*; this mutation, called *tufted*, affected a quite different trait, hair growth, but was very close to the locus of *Kink*. Dunn (as well as of course Mary Lyon who was also interested in the T locus) immediately began to use the marker to analyze T-locus genetics. They found, as would have been expected, that the *t* alleles that suppressed recombination with *Kink* also suppressed recombination with *tufted*. More important, both found that new viable exceptions generated by lethal *t* mutations all involved a recombinational event between the loci of *T* and *tufted*. This suggested very strongly that in fact *t* mutations comprised regional chromosomal differences, not single-gene mutations, since apparent crossovers could separate not only lethal factors from viable ones but also the element responsible for high transmission.

In the course of his work on the T locus, Dunn and his associates isolated and defined well over 100 recessive *t* mutations. More than 20 lethals of independent origin were found to fall into six complementation groups, each of which affected early embryonic development at a different and very specific stage. Dunn thought that the T locus represented an important center for controlling the first steps in establishing the patterns of embryogenesis, and that once its genetic complexity was unravelled, it might serve as a model for understanding gene organization and gene regulation in mammals. This promise may be fulfilled; work that was just under way at his death has shown that T locus genes appear to participate in development in a critical way by specifying cell surface components that guide embryonic cells through their first steps in differentiation. More than that is something that would have pleased the evolutionist in him very much; it seems that the role of T-locus genes is not limited to the mouse, but that similar genes, conserved through evolution, operate in the development of all mammals.

I have tried in this brief memoir to give a picture of Dunn as a man and scientist, and to concentrate on what I thought were the important factors that led him first to genetics and then to such an intensive and elegant concentration on one small region of mouse chromosome. In doing this I have relied on my own impressions gained during our long friendship, and on autobiographical material he left behind, particularly his Columbia University "Oral History" which was made available to me by his wife Louise. The memoir is sadly lacking in many respects because it follows only one thread, and gives no impression of his many other interests and accomplishments. I am tempted to do justice to him by discussing these: his books, which served to introduce generations of students and laymen to principles of genetics, his interest in human variation and his clear view of race, and his crusading for political freedom were all important aspects of his life. But, in fact, without a whole volume or maybe two to do it in, the job is just too big to attempt. Anyway, the story outlined here is probably the one that he would have thought most interesting.