MUTATION STUDIES, GENETICS AND PLANT BREEDING[1,2]

(Barley, spring wheat, oats)

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The atomic bomb brought to center stage of national attention the radiation studies at Washington State University applied to induced mutations and plant genetics. Barley and diploid wheat plants were exposed to various kinds of radiation from the Test Able atomic bomb at Bikini in 1946. The seeds of those plants were then brought to the Agronomy Department of WSU where Dr. Luther Smith compared the genetic and cytogenetic changes induced by the bomb and x-rays. He found the changes to be similar. Washington State pioneered in genetics and plant breeding even from its earliest history; but this cataclysmic event brought special attention to this work and provided an impetus still felt in the program.

Indeed, early WSU history of the science of genetics included the rediscovery of the Mendelian principles of inheritance by Dr. William Jasper Spillman (Nilan, 1961) who produced the first wheat hybrids and initiated wheat improvement research at WSU 1894-1901. He sought to find the laws governing the transmission of characters from parents to offspring. In 1899, he made

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1/ Part of History of Agronomy and Soils. W.S.U. 1958.

2/ The author gratefully acknowledges the extensive help provided by Drs. R. A. Nilan and C. F. Konzak in providing much of the basic information.

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fourteen crosses between wheat varieties with easily distinguishable, contrasting characters, e.g. the club vs. lax spike traits. Then he analyzed the results in the hybrid and second generations. He tallied the number of plants in the various classes of the second generation, thereby arriving at simple genetic ratios. He noted the hybrid generation was intermediate between parents and that the classes in the second generation were merely combinations of the parental characters and occurred in definite ratios. He independently and unknowingly had rediscovered the Mendelian principles of inheritance and thereby contributed to the enunciation of these genetic laws commonly known today.

Spillman had not known of similar experiments on peas conducted by the Austrian Monk, Gregor Mendel in 1865. Nor was Spillman aware of the work then in progress by three European scientists, Correns, DeVries, and von Tschermak, who were making similar discoveries to those of Mendel on heredity. However, the European scientists searched the literature and referred to Mendel's earlier publication and reaffirmed his conclusions. Spillman's findings were presented November 1901 at the Eleventh Annual Convention of the Association of American Agricultural Colleges and Experiment Stations in Washington, D.C. His paper was entitled "Quantitative studies on the transmission of parental characters of hybrid offspring." The paper was subsequently published as USDA Office and Experiment Station Bulletin No. 115, 1901.

While, by custom, the credit for rediscovery of the Mendelian laws of inheritance went to the three European scientists, in recent years outstanding geneticists of the USA, including Nobel Prize winner Hermann J. Muller, recognized the contribution of Spillman (and attempted to place Spillman in his proper role) as one of the founders of the science of genetics. Here at WSU, the "Agronomy Farm"--one of the finest field laboratories for experimental work in genetics and plant breeding--has been named "The Spillman Farm" (Nilan, 1961).

A WSU Distinguished Alumni Award recipient in 1966 was Dr. Karl Sax--a Spokanite who majored in Agronomy at WSU and received his B.S. degree here in 1916 (Phillips, 1967). He married one of his instructors, Miss Hally Jolivette, and they went to Harvard University where they began their work on genetic problems of agricultural plants. Sax pioneered studies of the effects of radiation on cells and the production of chromosome aberrations. He also did some work on demography which culminated in his widely read book entitled "Standing Room Only" in 1955. He received national and international recognition and numerous honors. His colchicine-doubled tetraploid Forsythia cultivar was named "Karl Sax" in his honor by the nursery trade.

Edward F. Gaines, the first employee of WSU to be titled "agronomist", was a driving force in wheat genetics and breeding 1912-1944. His accomplishments are covered in greater detail in Vogel's history of "Winter Wheat Improvement in Washington to 1982." His name was immortalized with the designation of Vogel's world yield-record-holding variety "Gaines."

About 1950, Dr. Fred Elliott began to utilize irradiation to transfer disease resistance from wheat grass to wheat in interspecific and intergeneric hybrids. This was part of his larger program in spring wheat breeding and plant genetics research. Dr. Elliott resigned in 1957 to join the faculty at Michigan State University.

Luther Smith certainly brought the mutagenesis focus to WSU through his early studies on induced mutation and by his cooperative work on the irradiation effects of Test Able atomic bomb at Bikini on exposed seeds. Unfortunately, he succumbed to cancer in 1952--very early in his career. One of his last postdoctorate students was Dr. R. A. Nilan, who came to the Department in 1951 and who carried on this work with such a promising beginning. He was joined by Dr. C. F. Konzak, a successor to Dr. Fred Elliott in 1958, and by Dr. Andris Kleinhofs in 1967. This three-some made tremendous accomplishments at WSU in the area of induced mutations and their applications to genetics and plant breeding. They point out that most induced mutations are akin to those that occur naturally, and that artificial mutagens help nature by speeding the process many-fold, perhaps as much as a thousand times, and harnessing it to bring about new directions in genetics and plant breeding.

Dr. Luther Smith (1951) recognized several of his students for their work in radiation studies; it seems appropriate to mention them again here: R. S. Caldecott studied the influence of heat treatments on effects of X rays and vice versa; C. C. Moh conducted in-depth genetic analyses of atomic bomb and X ray induced mutations in barley and wheat; Louise Roberta Sieburth studied the relation of atmosphere to biological effects of X rays, initiating research in an aspect that was not only continued at WSU, but which became important over the entire field of radiation biology; T. S. Osborne, an AEC fellow from Oklahoma A & M, studied the relationships of temperature and atmosphere to the effects of X radiation; Walter L. Nelson, a former AEC research fellow, studied hereditary variation in susceptibility to X ray effects; Mary Hafercamp studied the age factor in the response of seeds to X radiation, using wheat seeds stored at the Dryland Experiment Station, Lind, Washington. The seeds had been stored there for more than 30 years.

During Luther Smith's time, the radiation studies were supported by the American Cancer Society, the U.S. Public Health Service, the Atomic Energy Commission, the State of Washington Initiative 171 funds from the liquor tax, and by regular funds of the Experiment Station.

Beginning with the team efforts developed by R. A. Nilan and C. F. Konzak, outside support for radiation and mutagenesis research grew amazingly in amounts and the range of sources, taking on international as well as U.S. support. Support even further escalated as Dr. Kleinhofs added his biochemistry expertise to the team. It is estimated that these programs within the Department of Agronomy and Soils over the last 35 years received more than 5 million dollars in outside grants and support. Drs. Nilan, Konzak and Kleinhofs, and their associates and graduate students, are known internationally as well as nationally for their mutation work. They have been active in genetics and plant breeding programs world-wide. Even now, Dr. Konzak is an Associate Editor of the Egyptian Journal of Genetics and of Experimental and Environmental Botany, while for several years Dr. Nilan was an Associate Editor of Radiation Botany (now Experimental and Environmental Botany) and has served on the Editoral Board of Mutation Research from 1964 to the present.

Dr. Smith had published a monograph on barley genetics just prior to his death. A second monograph was published by Dr. Nilan in 1964. These monographs have been important references to barley genetic colleagues over the world.

In 1965, the highly successful interdepartmental, intercollegiate Program in Genetics (now Program in Genetics and Cell Biology) had its beginning in the Department. For the past 20 years, the Office of the Program has been housed in the Agronomy area and the agronomy faculty have played a major role in the development of the Program. Indeed, R. A. Nilan was Chair of the Program during its first thirteen years.

The Agronomy Genetics Mutation group has studied the effects of a wide variety of mutagens and have used several to induce mutants for use in their plant breeding and genetics research. They have pioneered in what is now popularly called "bio-engineering." Among the mutagens they used in their studies were X rays, neutrons, gamma rays, and a considerable array of chemicals, including ethyleneimine, ethyl methanesulfonate (and many analogues) diethyl sulfate, nitrosourea, and sodium azide. The discovery of the unusual ability of diethyl sulfate to induce high yields of visible mutations but low levels (almost none) of chromosome aberrations and low levels of seedling injury was made by R. E. Heiner, studying under the direction of Dr. Konzak. The mutagenic activity of azide and the means to increase its potency was discovered by this group (Nilan, 1975). Graduate students R. Spence, E. Sideris, W. Owais, and several trainees and post doctoral students, C. Sander, M. Niknejad, M. Awan, and G. Mustafa were among those studying various azide treatment conditions and interactions with other mutagens in efforts to understand the action mechanisms involved. Research by Owais, Kleinhofs, and Nilan led to the identification of the true mutagenic compound synthesized from azide by an enzyme present in cells of many plants, but not animals. As a result of this research, now extended by other laboratories over the world, azide has become one of the more important mutagens of choice both in plant breeding and in biotechnological or genetic engineering applications.

In wheat and barley mutation breeding programs, seeds usually are treated with a mutagen. The modified or new characteristics appear in the second generation (M_2), and by the third generation are generally quite stable in these self-pollinating plants. It is then possible to test these mutant selections by methods usually employed for evaluating advanced generation progeny from crosses. Most of the induced changes are deleterious to survival or growth; but the plant breeder can locate the "winners" by using large populations, or the desired trait(s) can be transferred to an uninjured genetic background by backcrossing or by crossing with an unrelated cultivar.

An early success using chemically-induced mutation resulted in a winter barley variety released by Washington State University in 1966. It was appropriately named "Luther" in memory of Dr. Luther Smith. Alpine barley seeds were treated with diethyl sulfate in 1960 by graduate student R. E. Heiner, under the direction of Dr. Konzak. The M_1 generation was grown in isolation on the Carl Beckley farm near Benge, Washington. About 5000 M_2 progeny were grown at Pullman in 1961 and eight mutants with short straw were selected by Carl Muir and Dr. Nilan. One of these became the variety, "Luther" released just four years after the mutant was isolated--a record for time of variety development. Luther was shorter strawed, more lodging resistant and more winter hardy. Luther yielded about 1/4 ton more than its parent, Alpine. During the late 60's and early 70's, Luther was the highest yielding barley in the Pacific Northwest and was grown on about 120,000 acres annually. Luther also proved an excellent parent for barley improvement through hybridization (Nilan, 1975). It's short height gene already has been exploited in several winter barley varieties, including Boyer and Showin (WSU) and in Mal and Hesk (OSU), as well as in lines nearing various stages of evaluation. A second short-strawed mutant from the same treatment also proved useful as a parent. The spring barley malting varieties Blazer and Advance also have an induced mutant in their origins.

Dr. Konzak has used the induced mutation technique in spring bread and durum wheat genetics and improvement research. Much of Konzak's research has dealt with the induction, genetics and breeding evaluation of mutant semidwarfing gene sources. While only a few of these mutant semidwarf gene sources have proved useful in breeding as alternatives to the widely exploited Norin 10 (Daruma) and Italian (Akakomugi, also Japanese) sources of semidwarfing, several mutant sources induced elsewhere have been used in variety development and studied at WSU. Several WSU-developed, mutant, semidwarfing gene sources also are proving of interest for breeding.

One of Dr. Konzak's mutant semidwarf durum selections, named Durox, was registered for production in France in 1981 and released in Idaho in 1984. Because of funding constraints, durum and oat breeding research at WSU was terminated in 1982.

Konzak (1984) reported that nearly 300 commercial varieties of cereals, legumes, fruits, and ornamentals exhibit improved traits due to induced mutations. Among the list are six from Washington State University: Boyer, Luther, Blazer and Advance barley; Duron durum what; and Dr. X Chrysanthemum. The WSU Agronomy and Soils mutation group's pioneering record of crop improvement through induced mutations speaks for itself.

While it was their pioneering mutation research that first brought them international recognition, Drs. Nilan and Konzak were first and foremost practical plant breeders, responsible for the development and release to Pacific Northwest growers an impressive sequence of superior barley (Nilan) and spring wheat cultivars. In fact, their mutation research was effective and of wider interest because they integrated the work with their genetics and germplasm improvement programs toward practical breeding goals. The list of barley varieties developed at WSU under Dr. Nilan's leadership include: Luther, 6-rowed winter feed (1966), Kamiak, 6-rowed winter feed (1970), Vanguard, 2-rowed spring malting (1970), Steptoe, 6-rowed spring feed (1972), Blazer, 6-rowed spring malting (1974), Boyer, 6-rowed winter feed (1975), and Advance, 6-rowed spring malting (1979).

For the past 8 years Nilan and, more recently, Steve Ullrich have been devloping mutant barley lines that will permit breweries to produce beer free of chill-haze without using chemical additives. Chemical mutagens, especially sodium azide, have been used to alter specific genes so that the subsequent mutant lines are low in certain phenols that contribute to beer haze. This research is being conducted with funds from the Carlsberg Brewery of Copenhagen

Denmark and the Anheuser Busch Brewery of the USA and in cooperation with the Department of Physiology, Carlsberg Research Center, Copenhagen.

Dr. Nilan assumed the position of Dean of the Division of Sciences, College of Sciences and Arts, at Washington State University in 1979, and although he still maintains a small research program, the leadership of the barley research in the Department has been transferred to Dr. Steve Ullrich. He and Nilan have released Andre 2-rowed spring malting (1983), Cougbar, 6-rowed spring feed with malting potential (1984) and Showin a 6-rowed winter feed (1984).

Dr. Konzak developed the oat varieties Cayuse (1966) and Appaloosa (1978); the durums Wandell (1971, the first semidwarf durum released in the U.S.), Waid (1980), and Durox (France, 1980, Idaho, 1983); the facultative (winterhardy) spring wheat, Walladay (1978); soft white spring wheats Urquie (1975), Waverly (1981), Edwall (1984), Penawawa (1985), and Wakanz (1986); the hard red spring wheats Wared (1974), Wampum (1978), and Spillman (1986). In addition, he developed a new dual-quality wheat (bread and pastry-making properties) through intensive development research collaborative with Mr. Gordon Rubenthaler, project leader of the Western Wheat Quality Laboratory. This new wheat is expected to be released in 1986, and the sister selections will be made available as germplasm stocks. Over his career, Dr. Konzak markedly improved the base yield potential of the breeding pool of soft white and hard red spring wheat germplasm at Washington State University, incorporated resistances to stripe, leaf, and stem rusts, to mildew, and to the local populations of the Hessian fly, and developed separate breeding populations of soft white and hard red spring wheats, and the dual-quality soft white spring wheat materials mentioned earlier.

Recent cooperative research indicates considerable potential for practical hybrid wheats using a new chemical hybridizing agent applicable with the seed as a soil application or to young seedlings. New dominant semidwarfing gene stocks developed in the course of Dr. Konzak's research could be important to the success of this new development toward higher yielding wheats that can offer more adaptive control over pests and diseases to result in more stable wheat crop production in the region.

Dr. Konzak also has been an innovator in plant breeding technology. Like Dr. Vogel who is famous for his development of threshers, nursery planters, weeders, etc., Dr. Konzak has collaborated with research and development sections of machinery companies and Drs. G. H. Hyde, J. Simpson and Mr. John George, faculty of the Department of Agricultural Engineering, to enhance such improvements. Through his ingenuity and vigorous support of equipment for plant breeding machinery, Dr. Konzak collaborated earlier with several companies to bring their growth chambers here for further testing and refinement. Dr. Konzak and his colleagues have put these to good use in the plant breeding programs.

Konzak and Experimental Aide Mike Davis, along with Agricultural Engineers mentioned above, developed a special experimental seed drill that would plant seeds more uniformly in narrower rows, more like that used by farmers, and as a consequence was able to increase experimental spring wheat yields about 10% on average. In addition, he and his colleagues have helped to improve a recently developed planter able to accurately space seeds as early generation progenies to aid plant breeders in selection work. With such a machine, mechanical work by trained personnel can be reduced appreciably and the work done more accurately, thereby freeing minds and hands for more technical matters.

In this age of computers, Dr. Konzak led the development of early applications of this advanced technology to the WSU plant breeding program. In part through his initiative and concepts presented at international meetings, programs to document germplasm of the multitudes of plant accessions all over the world have been developed. Inventories are now being indexed by computers to facilitate interchanges of information and germplasm. The computer expedites handling and ensures complete and accessible information on each stock. All WSU nursery data are now computerized.

As a further modernization of the technology, Dr. Konzak and his technicians developed adaptations to a commercially-produced nursery plot combine to achieve higher efficiency by harvesting and acquiring plot data at the same time and at the same rate so long as a 15-20 second break is made between plots for combine shake out. The 15-20 second period is used for weighing the samples on a sturdy electronic scale and for collecting the yield data in a portable data collector or computer, bagging and transferring the sample, and since only the combine operator handles the samples, interplot mixtures can be controlled. The manufacturer currently offers the innovation as an accessory and other manufacturers have hastened to follow the lead. Components of one type of electronic scale used on the plot combines also were developed at WSU with construction contracted to a Spokane Valley firm.

Excellent rapport and collaboration have prevailed between this group and the USDA winter wheat improvement group housed here. Many of their interests and problems overlap. They recognized these and cooperated to find common solutions to the problems. Together they provided a tremendous resource group and trained many, many graduates who now occupy prestigious positions around the world.

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