Changes were observed from miniature-alpha to beta and gamma, from miniature-beta to gamma, and from miniature-gamma to alpha and beta.

Miniature-alpha appears to be the most unstable and miniature-beta the most stable of the three mutable miniature allelomorphs as far as the changes from one miniature allelomorph to another are concerned.

[1] Demerec, M., these PROCEEDINGS, **12**, 1926 (687–690).

[2] Miniature-1 is constant allelomorph of mutable miniature and it is independent in origin of mutable miniature. It was found by Dr. C. W. Metz in 1922.

[3] Demerec, M., these PROCEEDINGS, **15**, 1929 (834–838).

[4] Demerec, M., *Verh. V. intern. Kongr. f. Vererbungswis.*, **1**, 1928 (183–193).

CHROMOSOME NUMBER AND THE MUTATION RATE IN AVENA AND TRITICUM[1]

BY L. J. STADLER

U. S. DEPARTMENT OF AGRICULTURE AND UNIVERSITY OF MISSOURI, COLUMBIA, MO.

Communicated November 14, 1929

In common barley (Hordeum vulgare) mutation is readily induced by X-rays.[2] Hundreds of recessive mutations have occurred in this species under known conditions of treatment. The rate of mutation is proportional to radiation intensity, and under heavy dosage of dormant seed mutations have occurred at rates exceeding 5 per cent.

The same treatments applied to common oats yield little or no induced mutation. In 1928 oats were included with barley in a number of experiments in which x-ray treatments were applied to dormant or germinating seeds. Oats of both of the commonly cultivated species, *Avena sativa* and *A. byzantina*, were included, the former species being represented by a selection of the agronomic variety Kherson and the latter by a selection of the variety Fulghum. The rate of mutation was determined for seedling characters only, previous trials having shown that about 90 per cent of the induced mutations were recognizable in the seedling stage. Only one mutation was found in the treated oats, a recessive white seedling segregating in the progeny of Kherson oats irradiated as dormant seed. The same treatments if applied to barley would have produced more than 40 mutations.

This extreme difference in mutation rate between oats and barley is not surprising in view of the known chromosomal constitution of the species concerned. Following the discovery by Sakamura[3] of a polyploid series in Triticum species, similar series have been found in Avena (Kihara[4]) and Hordeum (Griffee[5]). In each genus species with 7, 14 and 21 pairs of chromosomes occur. In *Triticum* and *Avena* the most valuable cul-

tivated species have the highest number of chromosomes, 21 pairs. In *Hordeum*, on the other hand, the cultivated species have 7 pairs of chromosomes, and the higher chromosome numbers are found only in wild species of the genus.

The mode of origin of the species of higher chromosome number is unknown, but it may be assumed that the triple complement of 21 pairs of chromosomes ultimately represents a combination of 3 groups of 7 pairs each, and that these three groups are identical in some of their genes. According to Winge's[6] hypothesis, the higher numbers may have been produced by chromosome doubling in a hybrid between species of lower chromosome number and thus the different groups may be as different as the chromosomes of different species. But species nearly enough related to produce a hybrid must still retain much common germ plasm. Whether the higher chromosome numbers resulted originally from the union of identical or different groups of 7, subsequent mutation and hybridization would lead to further differentiation between the chromosome groups. In either case, therefore, some genes would be identical in all groups and other genes would not; and we have at present no sound basis for estimating the proportion of genes in each class.

There is presumptive evidence of gene reduplication in wheat and oats in the segregation ratios of certain hybrids. Nilsson-Ehle[7] in 1911 reported three genes having the same effect on seed color in common wheat. When a red-seeded plant carrying all three dominant genes is crossed with a white-seeded plant in which all three are recessive, the F_2 ratio is 63 red to 1 white. Some red-seeded varieties have only two of the genes dominant, and these, when crossed with white-seeded plants, give a ratio of 15:1 in F_2. Other varieties, in which only one of the genes is dominant, give a simple 3:1 ratio in F_2. The same situation has been found in the inheritance of several other characters in both wheat and oats, as Gaines[8] has pointed out. Although a segregation ratio of 63:1 does not prove that the three genes concerned are identical, the frequent occurrence of such cases in these species of plants suggests that they contain many genes in triplicate. In barley segregation suggesting duplicate or triplicate genes is rare.

In the case of a dominant gene homozygous in all three groups, the recessive mutant character would not appear unless recessive mutation occurred coincidentally at the same locus in the three groups. For example, let A represent a dominant gene essential to chlorophyll development, A_1, A_2 and A_3 representing this gene in the three chromosome groups. A normal plant of common barley is $\frac{A}{A}$. Mutation of A to a produces the heterozygote $\frac{A}{a}$ and in the next generation this segregates 25% albino

plants, the homozygous recessive $\frac{a}{a}$. But a normal plant of common oats may be $\frac{A_1}{A_1}\frac{A_2}{A_2}\frac{A_3}{A_3}$. Mutation of A_1 to a_1 produces the heterozygote $\frac{A_1}{a_1}\frac{A_2}{A_2}\frac{A_3}{A_3}$, which by segregation produces the homozygous recessive $\frac{a_1}{a_1}\frac{A_2}{A_2}\frac{A_3}{A_3}$. This plant is green because of A_2 and A_3, and since A_2 and A_3 are homozygous, it will (barring further mutation) produce only green descendants. A segregation of albino plants from the homozygous triple dominant could be produced only by inducing mutation in A_1, A_2 and A_3 in the same cell. If mutations were induced simultaneously in one allelomorph of each pair, the recessive mutant character would appear in $^1/_{64}$ of the progeny. Similarly, in a plant in which the gene is dominant in two of the three groups $\left(\frac{A_1}{A_1}\frac{A_2}{A_2}\frac{a_3}{a_3}\right)$, simultaneous mutation of A_1 and A_2 would be required for segregation of the recessive character, which would then appear in $^1/_{16}$ of the progeny. The probability of occurrence of these coincidences is remote. Recessive mutations of appreciable frequency are to be expected only in those genes which are dominant in only one group $\left(\frac{A_1}{A_1}\frac{a_2}{a_2}\frac{a_3}{a_3}\right)$.

If gene reduplication is the cause of the low apparent mutation rate in oats, common wheat also should yield few mutations under x-ray treatment, and in the species of both *Avena* and *Triticum* it should be possible to show a relation between chromosome number and mutation rate. To test this relation the mutation rate under x-ray treatment was determined in species of varying chromosome number in both genera.

The experimental technique was similar to that previously described for barley,[2,9] except in certain details here stated. Unfiltered radiation from a Universal type Coolidge x-ray tube operated at 108 K. V. P., 4 m. a. tube current, 18 cm. target distance, was used for periods of 5, 10, 20, 30 or 40 minutes. The intensity of the radiation emitted under these conditions (measured in international *r*-units by means of a Victoreen dosimeter) is 290 *r* per minute of exposure. Dormant seeds were treated in all cases. The seeds were planted immediately after treatment, under conditions favorable to tillering. Seedling progenies were grown in the greenhouse from the main head and the first two tiller heads of each plant, later tillers being discarded because they are in some cases derived from earlier tillers. Mutation rates were determined from the progeny of tillering plants only, and in all cases recorded as mutations the mutant character segregated in the progeny of one head and was absent in other head progenies of the same plant. The primordia of the three heads tested are separate in the embryo at the time of treatment, and the frequency of mutation is indicated by the number of seedling mutations found in pro-

portion to the number of head progenies tested. Previous trials[10] with varying doses of x-rays applied to dormant seed of barley have shown that the mutation rate per unit of radiation intensity is a constant. This constant is a convenient basis of comparison of mutation frequency in different species.

The frequency of mutation in *Hordeum vulgare* under these conditions was determined for comparison with the species of *Avena* and *Triticum*. Dormant seed of the agronomic variety Himalaya was irradiated for periods of 10, 20, 30 and 40 minutes. The mutation rate per *r*-unit was $(4.9 \pm 0.9) \times 10^{-6}$.

Four species of *Avena*, *A. brevis*, *A. strigosa*, *A. byzantina*, and *A. sativa*, were treated. Kihara in 1919 reported 7 pairs of chromosomes in *A. strigosa*, and later 7 pairs were reported in both *A. brevis* and *A. strigosa* by Huskins,[11] Goulden,[12] and Aase and Powers.[13] *A. byzantina* and *A. sativa* are the common oats species with 21 pairs of chromosomes in which trials had been made in the previous season.

Seed of all four species was irradiated for periods of 10, 20, 30 and 40 minutes. The plants of *A. brevis* and *A. strigosa* were distinctly injured in early growth by the 30-minute and 40-minute exposures, but recovered sufficiently to produce some tillering plants suitable for the determination of mutation rate from all treatments. The injury to the common oat species was slight in early growth and inappreciable at maturity. An additional and more extensive trial was made in *A. brevis* and *A. strigosa* irradiated for 5 minutes, and in *A. strigosa* irradiated for 10 minutes. The frequency of mutation in the four species is shown in table 1.

TABLE 1

FREQUENCY OF INDUCED MUTATION IN SPECIES OF AVENA

SPECIES	CHROMOSOME NUMBER	MUTATION FREQUENCY DURATION OF TREATMENT (MIN.) 5	10	20	30	40	MUTATION RATE PER *r*-UNIT ($\times 10^{-6}$)
A. brevis	7_{II}	0/268	0/24	2/51	3/39	0/12	4.1 ± 1.2
A. strigosa	7_{II}	2/449	3/484	1/101	1/44	2/38	2.6 ± 0.6
A. byzantina	21_{II}		0/116	0/102	0/64	0/55	0
A. sativa	21_{II}		0/133	0/124	0/101	0/55	0

A. byzantina and *A. sativa*, the species with 21 chromosome pairs, yielded no mutations. The treatments applied to these species in this trial, together with those applied in the previous season, were sufficient to have produced more than 70 mutations in barley, instead of the single mutation found in oats.

The two 7-chromosome species of *Avena* yielded 14 mutations. Although the mutation rates in both of these species were somewhat lower than that of barley, the differences are not statistically significant. The mutant characters in *A. brevis* were "white" (3 cases), "virescent," and "pale yellow;" in *A. strigosa* "white" (2 cases), "pale green" (2 cases), "greenish yellow" (2 cases), "virescent," "banded," and "brownish."

All of these types except "brownish" are similar to mutant types previously found in barley.

The species of *Triticum* treated were *T. monococcum* (7 pairs of chromosomes), *T. durum* and *T. dicoccum* (14 pairs), and *T. vulgare* (21 pairs). The chromosome numbers given have been reported by Sakamura,[3] Kihara,[4] Sax,[14] and others. In the fall of 1928, 30-minute and 40-minute treatments were applied to *T. sativum* of the fall-sown variety Harvest Queen. In the following spring, treatments of 10, 20, 30 and 40 minutes were applied to spring-sown varieties of all four species, of which seed was kindly furnished by Professor H. K. Hayes, of the University of Minnesota. *T. monococcum* was so severely injured by exposures of 20 minutes or more that mutation rates could not be determined from any but the 10-minute treatment. *T. dicoccum* also was severely injured by the 20-minute and 30-minute exposures, but matured a few tillering plants given these treatments. Neither of these species matured seed from the 40-minute treatment. *T. vulgare* was injured by the 30-minute and 40-minute exposure, but recovered sufficiently to permit the determination of mutation rates from a small number of plants. *T. durum* was not seriously injured by any of the treatments used.

The frequency of mutation in the species of Triticum is shown in Table 2.

TABLE 2

FREQUENCY OF INDUCED MUTATION IN SPECIES OF TRITICUM

SPECIES	CHROMOSOME NUMBER	MUTATION FREQUENCY DURATION OF TREATMENT (MIN.) 10	20	30	40	MUTATION RATE PER *r*-UNIT ($\times 10^{-6}$)
T. monococcum	7_{II}	4/133				10.4±3.4
T. dicoccum	14_{II}	0/61	1/26	0/20		2.0±1.3
T. durum	14_{II}	0/55	3/237	1/73	2/79	1.9±0.5
T. vulgare	21_{II}	0/52	0/17	0/377	0/299	0

T. monococcum, though tested only on a small scale, yielded four mutations. Its mutation rate per unit of radiation intensity was higher than that of barley, though the difference is not statistically significant. The mutant types were "white" (2 cases), "virescent," and "yellowish-green."

The two species with the doubled number of chromosomes mutated at a lower rate. The test of *T. dicoccum* was small and only one mutation was found. The mutant type was "white." In the more extensive trial with *T. durum* 6 mutations were found. The mutant types were "yellow" (3 cases), "light yellow," "white," and "shriveled." In addition a white seedling segregation was found in the progeny of an untillered plant. All of the segregation ratios were apparently monogenic, like those from the 7-chromosome species, and probably all were due to the mutation of a gene dominant in only one group.

T. vulgare, though much more extensively tested, yielded no mutations. The treatments applied were sufficient to have given about 40 mutations in barley.

The results as a whole support the hypothesis that the frequency of induced mutation observed in polyploid species is low because of gene reduplication. This does not imply that other factors may not affect mutation rate as well. It is possible that the basic mutation rate may differ materially in different groups. The data available in *Avena* and *Triticum* are not yet sufficiently extensive to permit strict comparison between individual species. But it is clear in both genera that the mutation frequency (as measured by visible effects) decreases sharply with increasing chromosome number. Apparently the proportion of mutable genes not reduplicated is very small in the species with the triple chromosome number. In the species with the double chromosome number a considerable proportion of the mutable genes appear to be dominant in only one of the two chromosome groups.

In untreated plants the rate of change due to recessive mutation is doubtless decreased similarly by polyploidy. Probably this is the reason for the fact that seedling chlorophyll characters, though of fairly common occurrence as recessives in barley, have been found only rarely in wheat and oats. These characters are relatively inviable or lethal, and in regularly self-fertilized species their causal genes would quickly be eliminated from the germ plasm. The deleterious characters found are probably due to mutations of relatively recent occurrence, and these apparently have been much less frequent in wheat and oats than in barley.

The expression of dominant mutation, on the contrary, would not be affected by polyploidy. Three-fourths of the progeny of the mutating plant would have the new dominant gene in one of the chromosome groups, and these would show the dominant character. In fact, the frequency of dominant mutation would probably be somewhat higher in the polyploid species, since the number of genes which might mutate would be increased. But if the mutations induced by x-rays are representative of mutation under natural conditions, the proportion of dominant mutations is extremely small.

[1] These experiments were supported in part by a grant from the NATIONAL RESEARCH COUNCIL, Committee on the Effects of Radiation upon Living Organisms.

[2] Stadler, L. J., *Science*, n. s., **68**, 186–187 (1928).
[3] Sakamura, T., *Bot. Mag.*, **32**, 151–154 (1918).
[4] Kihara, H., *Bot. Mag.*, **35**, 19–44 (1919).
[5] Griffee, Fred, *Univ. Minn. Studies Biol. Sci.*, **6**, 319–331 (1927).
[6] Winge, Ö., *Compt. Rend. Trav. Lab.*, Carlsberg, **13**, 131–275 (1917).
[7] Nilsson-Ehle, H., *Lunds Univ. Arsskr. N. F.*, Afd 2, Bd. 7, Nr. 6, 3–84 (1911).
[8] Gaines, E. F., *J. Amer. Soc. Agron.*, **19**, 202–205 (1927).
[9] Stadler, L. J., *J. Hered.*, (1929) (in press).
[10] Stadler, L. J., *Anat. Rec.*, **41**, 97 (1928).
[11] Huskins, C. L., *Sci. Agr.*, **6**, 303–313 (1926).
[12] Goulden, C. H., *Minn. Agr. Expt. Sta. Tech. Bul.*, **33** (1926).
[13] Aase, Hannah C., and Le Roy Powers, *Am. J. Bot.*, **13**, 367–372 (1926).
[14] Sax, K., *Science*, n. s., **54**, 413–416 (1921).