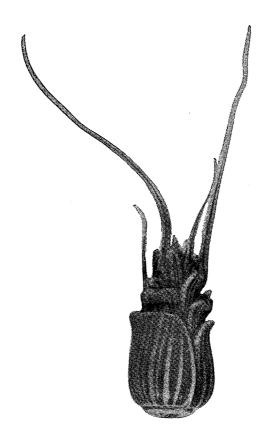
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A spikelet of Aegilops squarrossa strangulata. S. page 6



I. RESEARCH NOTES

New cases of male-sterility and new restorer source in T. aestivum

E. OEHLER and M. INGOLD Federal Agricultural Research Stations, Lausanne, Switzerland

During our studies in view of improving the resistance to disease in cultivated wheat, we have carried out, among others, two groups of crosses. One consists of a line of *Triticum timopheevi* Zhuk. crossed with four cultivars of *T. aestivum* Host (Probus, Mont-Calme 245, Heine VII and Derenburger Silber) and the other, of an hexaploid line, obtained from a cross *Aegilops ventricosa* × *T. timopheevi* 2 (creator: Simonet) crossed with the cultivars Probus, Heine VII and Cappelle. In each case *T. aestivum* was the pollinator.

 F_1 plants are morphologically intermediate between parents, and their fertility is very low, as Wienhues (1951) and Simoner (1957) reported. We can ascertain the same thing for the majority (75 to 95%) of the F_2 plants and for a large proportion (75%) of the F_3 to F_5 plants. The stamens remain indehiscent due to irregularities at the meiosis. Only a small number of ovules is able to develop. After one backcross with T. aestivum, we obtain one to two seeds per ear; with two to three successive backcrosses we got five to ten seeds per ear.

In this material, we could isolate from the F_2 self-fertile plants, the stamens of which being partly or completely dehiscent and producing respectively 5 to 17 and 17 to 22 seeds per ear. We have thus obtained stable and fertile lines of the T. aestivum type.

The progeny of some F'₂ plants showed a different behaviour pattern. After 5 to 7 consecutive backcrosses, the stamens remained indehiscent. There are:

| 3 | plants | T. timopheevi | × | Heine VII |
|---|--------|--------------------|---|--------------------|
| 1 | plant | T. timopheevi | × | Derenburger Silber |
| 1 | plant | Aegtr. vent. Tim 2 | × | Heine VII |
| 1 | plant | Aeotr. vent. Tim 2 | × | Cappelle |

In this material hand-pollinated with T. aestivum, the average fertility reaches 55% (18 seeds per ear) and can even arrive at 100% for certain ears. The same fertility is obtaind, when such plants flower freely side by side with T. aestivum plants, which are acting as pollinators. This case of stamens' indehiscence is thus different from that caused by cytological instability. It is a case of male-sterility comparable to those described by the American authors (Wilson, Ross, Schmidt) obtained from crosses T. timopheevi \times Bison.

These different male-sterile forms were crossed with a series of *T. aestivum* from our own collection.

Table 1. Cultivars and strains of T. aestivum giving no restoration of male-fertility

| | | | <u> </u> | | |
|-------------------------------------|------------|---------|------------|----------|-----------|
| Switzerland | France | Germany | Austria | N. Amer. | S. Amer. |
| Probelle | Chambord | Florian | Lassers SR | Albit* | Fronteira |
| Probus | Champlein | Hauber | Record | Atlas 50 | |
| A 17 | Côte d'Or | | | Taylor | |
| A 205 | C.F.51.24. | | | - | |
| A 273 | Floress | | | | |
| A 394 | Francest | | | | |
| A 912 | Ideal | | | | |
| A 914 | Magali | | | | |
| F ₁ (A 909 × Francest) | | | | | |
| F ₁ (A 909 × NB 1978) | | | | | |
| F_1 (Francest \times A 142) | | | | | |
| F_1 (Francest \times A 205) | | | | | |
| F_1 (Francest \times Fronteira) | | | | | |

^{*} T. aestivo-compactum Sohiem.

All F₁ issued of these crosses are male-sterile. On the contrary, crosses carried out with the French cultivar Frimépi* have all given completely fertile F₁ as well with the male-sterile obtained from *Timopheevi* as with those issued of *Aegtr. vent-Tim* 2.

While examining 20 F_2 progenies, we noticed a segregation between fertile and sterile plants. The proportion seems to be three fertile for one sterile, but these figures have still to be confirmed. Primépi can thus be considered as a restorer cultivar.

Crosses from male-sterile with self-fertile types obtained from the same material ($timopheevi \times aestivum$) also give fertile plants. However, the F_1 is not homogenous. One finds fertile plants, other are sterile, and at last for a certain number only a part of the stamens are dehiscent. The F_1 behaviour can be partially determined by the fact that the self-fertile types used in the crosses are heterozygous for the restorer genes. The presence in F_1 of plants with partially dehiscent stamens indicates a more complex inheritance than the one of the Primépi restorer.

Table 2. Fertility of F₁ (male-sterile × Primépi)

| Number of ears | Seeds per ear | Seeds per spikelet |
|-------------------|------------------|-----------------------|
| 20 | 50 | 2.58 |
| 5 | 56 | 2.68 |
| 35 | 47 | 2.52 |
| | 20 5 | ears ear 20 50 56 |

^{*} creators: Dromigny and Hamel

Effect of EMS on germination of einkorn wheat

Taro Fujii National Institute of Genetics, Misima, Japan

Very high mutagenic effects of ethyl methane sulphonate (EMS) were already reported by many researchers. Normal seeds, and heterozygotic seeds from the cross between chlorina and normal green in *Triticum monococcum flavescens* were used in this experiment. The aim of this experiment was to determine the differences in

mutagenic effect for a specific gene and the difference in the appearance of mass mutation between γ ray and EMS treatments. For two hours steeped seeds were treated with 0.1, 0.3 and 0.5% EMS solutions for 22 hours. For comparison, for 24 hours steeped seeds were subjected to 0.5, 1.0, and 1.5 kr of γ -rays. Moreover, EMS and γ -ray treatments were combined to examine the synergistic effect of both treatments. The treatments were so combined that 2 hours steeped seeds were placed in EMS solution for 22 hours and γ -ray exposure was done after 5 min. washing with tap water. Steeping and EMS treatments were done under room condition at 20°C.

 γ -rays had almost no effect on germination rate of the normal strain, but the F_1 hybrids were affected. In EMS treatments, germination rate in both strains decreased with increasing concentration of EMS, more markedly in the F_1 lot, as shown in Table 1. (Of course, germination rate of F_1 seeds was a little inferior to that of the normals because they are usually smaller than normal seeds). Moreover, combination of EMS and γ -ray treatments showed severer killing effect than each single treatment; about 50% and only 14% seeds germinated in the normal and the F_1 batch, respectively, in the EMS 0.5% and γ -ray 1 kr treatment lot. Almost all seedlings died out in these lots. Additive or synergistic effects of both treatment are assumed. The appearance of chlorina stripes due to somatic mutation is now under examination.

Table 1. Germination rates of γ -ray and EMS treatments of normal and F_1 seeds

| Treatment | Normal | F ₁ |
|---------------------------------|--------|----------------|
| γ-ray 0.5 kr | 86 | 54 |
| 1.0 | 86 | 50 |
| 1.5 | 88 | 46 |
| EMS 0.1 % | 91 | 50 |
| 0.3 | 80 | 48 |
| 0.5 | 70 | 32 |
| EMS 0.3% + γ-ray 1.0 kr | 78 | 38 |
| EMS 0.5% + γ -ray 1.0 kr | 52 | 14 |
| | 90 | 78 |

Inheritance of the sphaerococcum effect in tetraploid wheat 1)

J. W. Schmidt and V. A. Johnson 2)

The sphaerococcum or shot-wheat kernel has been the distinguishing characteristic of the hexaploid wheat, Triticum sphaerococcum Perc.. Sears (1947) has placed the hemizygous-ineffective recessive gene controlling this in chromosome 3D. Since then, Schmidt, et al. (1963) reported that a similar effect was produced by an incompletely dominant gene not allelic to the gene on chromosome 3D. This appeared as a mutant in common wheat. Because of sterility interactions in monosomic studies, the incompletely dominant gene could not be placed on a particular chromosome or in a genome. However, evidence that the sphaerococcum effect is not restricted to the D genome came from the report by Schmidt and Johnson (1963) of the same character in a tetraploid wheat. In all three of these sources expressions of the sphaerococcum effect is similar: (1) Awn length is reduced and a shortened kernel develops in nearly hemispherical glumes. (2) The rachis becomes strongly zigzag in shape. (3) Reduction in plant height occurs. (4) Flag leaves tend to be short and blunt.

The sphaerococcum variant in the tetraploid wheat was crossed to the tetraploid population in which it was found (C. I. 8594, a durum introduction from China). The F_1 resembled the normal parent although there was some expression of the sphaerococcum effect. More recent crosses show that the heterozygotes can probably be identified. An average of 54 F_2 plants were grown from each of seven F_1 plants. Since there appeared to be no significant deviation from the expected segregation ratio for any of the F_2 populations, all F_2 populations were combined. Of the 377 F_2 plants, 285 were classified as non-sphaerococcum and 92 as sphaerococcum, a nearly perfect one-factor ratio. Classes were clearly defined.

The presence of this *sphaerococcum* variant in a tetraploid wheat is evidence that the *sphaerococcum* effect is not restricted to the D genome in wheat. Furthermore, the heterozygote appears to show considerable heterotic effect in grain yield per head

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²⁾ Professor of Agronomy, University of Nebraska, and Research Agronomist, Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture, Lincoln, Nebraska.

and possibly could have some plant-breeding value. The gene should be useful for linkage studies in tetraploid wheats. Small quantities of seed of the variant are available to those interested.

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An intergeneric hybrid between Eremopyrum orientale (LINN). JAUB. et SPACH. and Aegilops squarrosa LINN.

Sadao SAKAMOTO National Institute of Genetics, Misima, Japan

An intergeneric triploid hybrid between Er. orientale (2n=28; Iran) and Ae. squarrosa (2n=14; material received from Dr. H. KAPPERT) was obtained in 1964.

The shape of the F_1 spikes was intermediate between the parents but the spikelets showed characters of Eremopyrum. Pollen- and seed-sterility were complete. Chromosome pairing at MI of PMC's of the F_1 is given in Table 1. Average chromosome pairing per cell was $0.1_{\rm II} + 20.8_{\rm I}$. All bivalents observed were associated loosely by a terminal chiasma. This indicates that the genomes of Er, orientale are quite different from the D-genome of Ae, squarrosa.

Table 1. Chromosome pairing PMC's of the F_1 hybrid between Er. orientale and Ae. squarrosa

| Chromosom | e pairing | |
|-----------|-----------|-----------------------|
| п | I | No. of cells observed |
| | 21 | 1,067 |
| 1 | 19 | 123 |
| 2 | 17 | 1 |
| Total | | 1,191 |

Altered potency of chromosome 5B in wheat-caudata hybrids

M. D. Upadhya ¹)

Indian Agricultural Research Institute, New Delhi, India

The activity of chromosome 5B to control the intergenomic pairing in hexaploid wheat is suppressed in the presence of the genotype of Ae. speltoides was shown by RILEY et al. (1958; J. Hered. 49:91—98). Where as, Mochizuki and Okamoto (1961; Chromosome Inform. Serv. 2:12—14) had shown that the genome of Ae. speltoides also suppresses the activity of this corresponding chromosome at the tetraploid level, in crosses with T. turgidum. It was further indicated by RILEY (Proc. 2nd Intern. Wheat Genet. Symp.), that in 28 chromosome hybrids between T. aestivum and the diploid species Ae. mutica there is considerable meiotic pairing, similar to that observed in 28 chromosome aestivum-speltoides hybrids. However, in 28 chromosome hybrids between T. aestivum and Ae. longissima, RILEY et al. (1959; Nature 183:1244—46) had shown that the genotype of longissima does not suppress or modify the activity of chromosome 5B.

SARKAR and STEBBINS (1956; Amer. J. Bot. 43: 297-304) and RILEY et al. (1958; J. Hered. 49: 91-98) had proposed that Ae. speltoides is the diploid progenator of the B genome of polyploid wheats. Where as, CHENNAVEERALAH (1660; Acta. Hort. Gotoburgensis 23: 85-178) had suggested Ae. mutica to be the B genome donor of the polyploid wheats.

It is interesting to note that the genomes of both of the diploid species suggested to be the B genome donors share in common the distinctive feature to suppress the activity of chromosome 5B of wheat.

According to Kihara's genomic grouping Ae. speltoides and Ae. longissima belong to the S group and Ae. mutica belongs to the M group. Therefore, the study was made on the effect of the genotypes of the diploid species belonging to the C group, on the activity of chromosome 5B of wheat.

Monosomic plants for chromosome 5B of variety Chinese Spring of T. aestivum were crossed with Ae. caudata as the \nearrow parent. The two hybrids raised to maturity showed to be having 28 chromosomes. Analysis for the chromosome associations was

¹⁾ Present address: Department of Horticulture, College of Tropical Agriculture, University of Hawaii, Honolulu, Hawaii, U.S.A.

made of the first meiotic metaphase from 50 microspores from each plant. Out of 100 cells analysed, 93 cells showed to be having chromosome associations varying from $1_{\rm II} + 26_{\rm I}$ to $1_{\rm III} + 5_{\rm II} + 15_{\rm I}$ (Table 1). For only those cells showing the chromo-

Table 1. Chromosome behaviour of 28 chromosome hybrids at first metaphase of meiosis

| ype of chromosome associations | Number of cells |
|----------------------------------|-----------------|
| 28 _I | 7 |
| $1_{\text{II}} + 26_{\text{I}}$ | 9 |
| $2_{\Gamma\Gamma} + 26_{\Gamma}$ | 30 |
| $3_{II} + 22_{I}$ | 28 |
| $4_{\Gamma\Gamma} + 20_{\Gamma}$ | 5 |
| $5_{II} + 18_{I}$ | 3 |
| $6_{II} + 16_{I}$ | 4 |
| 1_{III} + 25_{I} | 1 |
| $1_{III} + 1_{II} + 23_{I}$ | 1 |
| 1_{III} + 2_{II} + 21_{I} | 6 |
| 1_{III} + 3_{II} + 19_{I} | 3 |
| $1_{III} + 4_{II} + 17_{I}$ | 2 |
| $1_{III} + 5_{II} + 15_{I}$ | 1 |
| Total | 100 |

some associations, the mean frequencies of trivalents and bivalents per cell, was 0.15 (range $0\sim1$) and 2.62 (range $1\sim6$) respectively. The range of chromosomes entering associations was found to be from 2 to 13 with a mean of 5.74 chromosomes per cell. Similarly, the mean chiasmata frequency per cell was found to be 2.98.

Kihara and Lilienfeld (1935; Cytologia 6:195—210) had observed the occurence at first metaphase of meiosis in the vulgare-caudata F_1 hybrid a range of 3~5 bivalents with a mean of 4 bivalents and trivalents varying from 0~2. Kihara (1958; Proc. 10th Intern. Cong. Genetics, Montreal 2 142—171) also reported the occurence of 0~5 bivalents (mode 3) and trivalents in the F_1 of the vulgare-caudata cross, where as the reciprocal cross had the range of 0~4 (mode 2) bivalents, along with trivalents and quadrivalents.

The present observations and those reported by earlier workers clearly indicate a higher degree of chromosome pairing in 28 chromosome aestivum-caudata hybrids than that observed in euhaploids of aestivum or the 28 chromosome aestivum-longissima or aestivum-rye hybrids. Thus suggesting that the genotype of Ae. caudata seems to modify the potency of chromosome 5B to result in the lowering of the threshold value inhibiting intergenomic chromosome associations.

This lowering of the threshold value of chromosome 5B by the *caudata* genotype results in increased intergenomic pairing in the 28 chromosome *aestivum-caudata* hybrids. But that this altered threshold value is not of the same magnitude as is brought about by *speltoides* or *mutic* genotypes.

Kihara's data on the occurrence of higher frequencies of trivalents and quadrivalents in the *caudata*-wheat hybrids over that in the reciprocal *wheat-caudata* hybrids, further indicate that the effect of *caudata* genotype on chromosome 5B is enhanced by the *caudata* cytoplasm in the F₁ hybrids.

The present conclusions are further supported by the observations of Kihara and Lilienfeld (1935; Cytologia 6: 195–210) on the high degree of chromosome pairing in the triploid F_1 hybrid between $Ae.\ caudata$ and $T.\ durum$. The mean chromosome associations per cell, at the first metaphase of meiosis, calculated from their data are 4.8 biv. (range 1~8) and 1.08 triv. (range 0~4). These frequencies are much higher than those observed in the triploid F_1 hybrid between $Ae.\ longissima$ and $T.\ dicoccum$ by Kihara (1949; Cytologia 14: 135–144).

Transfer of v₁ gene from common wheat to emmer wheat

K. TSUNEWAKI Laboratory of Genetics, Kyoto University, Kyoto, Japan

The polyploid series of wheat offers an excellent opportunity to investigate the transfer of major genes between species which differ in chromosome numbers. The effects of allopolyploidy on the phenotypic expression of those genes can be studied by such transfer, which provides a new means to elucidate the genetic role of polyploidy in wheat evolution.

A virescent seedling found by Neathy (1933) in F_6 of a cross, Garnet \times Double Cross, was revealed to be under control of a simple, recessive mendelian factor, that was later designated by v_1 and located on chromosome 3B (Sears 1954).

In 1959 the author obtained from Dr. E. R. Shars a virescent strain of a common wheat variety, Chinese Spring, that possesses v_1 gene. An emmer wheat, T carthlicum var. stramineum was crossed to this v_1 -carrying Chinese Spring, and the F_1 hybrids were successively backcrossed to the emmer parent. In the first backcross generation plants with 14 pairs of chromosomes were cytologically selected for further backcrosses.

Germination and segregation of virescent plants in the selfed progenies of seven B_1 and a single B_2 plant are shown in Table 1. Two facts are evident from this table. In the selfed progenies of V_1v_1 heterozygotes virescent plants were much fewer than expected from the 3:1 ratio of greens vs. virescents. At the same time, their germination rate was significantly lower than that of their normal sib lines. This can not be attributed to a maternal effect, since the germination rate of backcrossed seeds, which were set on V_1v_1 and V_1V_1 plants, were almost the same, i.e., 91 and 93 per cent, respectively. Nor, can it be due to a genetic disharmony caused by combining genes of emmer and common wheats, because seeds of normal sib lines germinated well, in spite of having almost the same gene combinations.

If the germination rate of a normal seed population were 91 per cent (=germination rate of V_1V_1) and its decrease in the selfed progenies of V_1v_1 to 75 per cent were attributable only to seed lethality of v_1v_1 the seed population raised by self-pollination of the heterozygote should have consisted of 78 per cent greens and 22 per cent virescents; these figures fit well the 3:1 ratio of greens vs. virescents. In fact, occasionally obtained virescent segregants of emmer wheat were very weak.

From the results of this investigation, it is concluded that v_1v_1 homozygotes of emmer wheat are mostly seed lethal. Apparently, the gene v_1 upsets the physio-

logy of embryos more drastically on the tetraploid than on the hexaploid level. Sears (1954) reported that plants of common wheat monosomic for chromosome 3D and heterozygous for v_1 bore 15 to 20 per cent shriveled seeds, and all their virescent offspring were disomic. From this he assumed seed lethality for mono-3D v_1v_1 . Since emmer wheat homozygous for v_1 seems to be equifunctional to nulli-3D v_1v_1 , though the latter has the remaining six chromosome pairs of D genome, Sears' result confirms to a certain extent the present finding that v_1v_1 emmer is seed lethal.

A large number of genes become duplicated as a consequence of polyploidy and one of the duplicates may mutate without impairing any preexisting characters. Common wheat, in fact, has many duplicated genes which are homoeologous in nature, including the present V triplicates. The results of the present investigation demonstrate that a hexaploid is more tolerant of a deleterious gene than a tetraploid, and consequently has a greater potentiality for maintaining an apparently deleterious gene.

Table 1. Germination and segregation of virescent plants in the selfed progenies of B_1 and B_2 plants of T. aestivum var. Chinese Spring $(V_1v_1) \times T$. carthlicum var. stramineum (V_1V_1) , the latter being the backcross parent

| Pa | arent | Line | Seeds | Seeds | Germi- | | | Virescent | χ^2 |
|----------|----------------|--------|-------|-----------------|---------------|--------|-----------|-----------|----------|
| Genotype | Generation | No. | sown | germi- nated | nation (%) | Normal | Virescent | (%) | (3 : 1) |
| V_1v_1 | В1 | 1145–3 | 20 | 14 | 70.0 | 13* | 1 | 7.1 | 1.52 |
| " | ,, | 1149-1 | 72 | 54 | 75.0 | 51* | 3 | 5.6 | 10.89** |
| ,, | , | 1149–3 | 71 | 57 , | 80.3 | 53* | 4 | 7.0 | 9.83** |
| " | $\mathbf{B_2}$ | 10521 | 49 | 34 | 69.4 | 34* | 0 | 0.0 | 11.33** |
| | Total | l | 212 | 159 | 75.0 | 151* | 8 | 5,3 | 33.81** |
| V_1V_1 | B ₁ | 1147-1 | 20 | 19 | 95.0 | 19 | 0 | _ | _ |
| ,, | ,, | 1147–2 | 70 | 62 | 88.6 | 62 | 0 | - | _ |
| ,, | ** | 1149-2 | 90 | 83 | 92.2 | 83 | 0 | _ | - |
| ,, | ** | 1151–3 | 90 | 82 | 91.1 | 82 | 0 | - | _ |
| | Total | | 270 | 246 | 91.1 | 246 | 0 | _ | |

^{* :} Some plants showed variegation of leaf color in mid-winter, indicating to be heterozygous for v_1 .

^{** :} Significant at the 1 per cent level.

Wheat collecting expedition to Afghanistan

G. M. HALLORAN

Agricultural Research Institute, Wagga Wagga, New South Wales, Australia

From mid-August to mid-November, 1965 the author accompanied a British Expedition from the University of Reading's Exploration Society for wheat collecting in Afghanistan. The principal aim of the expedition was to obtain species and varieties of *Triticum* and of closely related genera, but at the same time samples of *Hordeum* species, both cultivated and wild, grasses, pasture legumes, field peas and beans were collected.

The expedition obtained about 1,900 samples, approximately 1,500 of which were of species and varieties of Triticum. The wheats were predominantly Triticum aestivum ssp. vulgare types but many species and subspecies of the tetraploid and hexaploid levels of Triticum were obtained, although all were of reasonably rare The wheat collection has, however, to be closely examined and taxonooccurrence. mically classified before a detailed account of the species and subspecies constitution Two almost identical sets of specimens were collected, one of which can be given. will be grown at the Plant Breeding Institute, Cambridge, England and the other at the Agricultural Research Institute, Wagga Wagga, New South Wales, Australia. Specific interest will be shown in this collection in Australia from the points of view of the nature and genetics of phasic development, disease resistance, and in particular possible resistance to take-all, Ophiobolus graminis, and possible new sources of good baking-quality characteristics. It is anticipated that other interests of wheat breeders in the material of this collection will be made known, to whom seed supplies will become available late in 1966 subsequent to initial seed multiplication.

It was the aim of the expedition to attempt to collect as wide a range of new genetic diversity as was possible from the country. The approach to a problem of this nature was very difficult to place upon a very rational basis, that is the choice of the particular areas of the country wherein collection would be thought to have

The editorial comment: The results of the Kyoto University Scientific Expedition to the Karakoram and Hindukush, 1955, Vol. 1 has just been published by the Committee of the Kyoto University Scientific Expedition to the Karakoram and Hindukush, Kyoto University, Kyoto, Japan, 1965. The volume contains the research reports on "Cultivated Plants and Their Relatives" including wheat and related genera. (K.Y.)

most adequately satisfied this aim. One criterion for the choice of collecting are as was that of a wide difference of habitat type. For this purpose three habitat types were specified wherein intensive collecting activities would be carried out. These habitat types were as follows:

- 1. Very high altitude habitat,
- 2. Moderately high altitude habitat,
- 3. Lowland semi-desert habitat.

The second criterion was that of remoteness and inaccessibility of the wheat-growing area, an area which had not been extensively covered by previous wheat-collecting expeditions and one wherein a large number of very isolated wheat-growing communities were likely to be found.

The area chosen, embracing habitat types 1 and 2, was that along the valley of the Hari Rud River eastward from Herat through the centre of the country along the Ghorband River to Kabul in the east of the country. Collections in the lowland semi-desert habitat which, however, were not inaccessible or remote areas, were made around Kandahar and Boost in the south and Kunduz, Tashkurghan and Mazar-i-Sharif in the north of the country. Because the expedition was somewhat late for the harvest in the lowland semi-desert areas of the country a comprehensive collection from this habitat was not able to be made. However, samples kindly given the expedition by the Afghan Ministry of Agriculture from a comprehensive collection made recently by that organization in these areas, were a very useful addition to the expediton's specimens from this habitat type.

It is anticipated that the wheat collection made by the expedition, its documentation and subsequent study will succeed in two broad aims. Firstly, it is hoped that some substantial contribution may be made to a widening of the gene pool for characters of importance of the wheat crop in supplying wheat breeders with new sources of genetic variation for breeding. Secondly, it is envisaged that a study of the collected specimens from the point of view of their distribution, taxonomy, developmental physiology and cytology may provide useful information upon the evolutionary history of *Triticum* and of its related genera in this part of the world.

Another feasible approach to study the phylogeny of cultivated hexaploid wheat 1)

Guillermo S. RYAN
Instituto de Fitotecnia, Castelar, Argentina

The phylogeny of cultivated hexaploid wheat is always an exciting subject. It is not necessary to refer here to well known pioneering research work made by Kihara, Sarkar, Stebbins, McFadden, Sears and others. The main approaches to the problem can be divided as follows:

- a) cytogenetic studies, including genome and karyotype analysis, chromosome affinity, interspecific crosses and so on,
- b) deductions based on analogies between different phenotypes.

It is believed that plant reaction to narrowly specific pathogens might be used as a third feasible approach that could shed light on controverted results obtained with the above techniques, as suggested by VAVILOV'S use (1914) of fungi as "physiologic reactors" in systematic botany. If it is assumed that characters of disease reactions in hexaploids or tetraploids derivate from their diploid ancestors, this gives way to screen the possible genome contributors, in order to trace the source of these characters.

To test this working hypothesis, seedlings of 129 strains from the *Aegilops* collection of the Instituto de Fitotecnia, were infected in the greenhouse with conidia of wheat powdery mildew (*Erysiphe graminis tritici*) from the most common physiologic race found in Argentina. Table 1 summarizes results obtained; strains are grouped in four sections according to Kieara (1949 and 1957).

Before drawing conclusions, the following remarks must be emphasized:

- a) samples employed came from plots open pollinated during several years,
- b) Aegilops strains originated from ten sources, listed in Table 2,
- c) no attempts were made to purify this material or to eliminate identical strains from different sources.
- d) botanical names are those indicated by the sources mentioned in Table 2,
- e) some species contribute with only one strain while others are represented by more than ten,
- f) no representatives from Comopyrum and Amblyopyrum sections were included.

 In spite of the aforementioned factors, it is possible, nevertheless, to make the following statements:

¹⁾ Paper No. 339 of the Institute de Fitotecnia, CIA, INTA, Castelar, Argentina.

Table 1. Reaction of Aegilops strains infected with wheat powdery mildew (Erysiphe graminis tritici)

| | jt | I | cylindrica | 14 | C D | pyrum |
|-------------------|-----------|-----------------------------------|---------------------|-----------------------|--------------------------------|-----------|
| | ı | I | caudata | 7 | С | Cylindro- |
| - | 11 | vulgaris | triaristata | 14 or 21 | CuMt or CuMtMt2 | |
| E | 1 2020 | glauca typica | triuncialis " | 14 | СнС | |
| 12 | | _ peregrina | variabilis " | 14 | C¤S₹ | |
| i | | I | kotschyii | 14 | CuSV | |
| 1111 | | armageti typica macrochaeta | biuncialis " | 14 | СчМъ | |
| 1111 | | typica procera erigerens | ovata " | 14 | CuMo | |
| I | | 1 | umbellulata | 7 | Ç | Polyeides |
| Susceptible | Su | Variety | Aegilops species | some number (n) | Genome- type ¹) | Section |
| Strains tested to | | | | Chromo- | | |

| Sitopsis | Ø | 2 | speltoides " | ligustica | 11 | 7 (5) 1 | ∞ |
|------------|-------|-----|---------------------|---|---------|------------|----------------|
| | ល | 2 | longissima | 1 | 1 | က | m |
| | % | 7 | sharonensis | 1 | I | 1 (1) | H |
| | Sp | 7 | bicornis | l | 87 | I | 7 |
| Vertebrata | a | 2 | squarrosa " " | typica anathera meyeri strangulata | 2 2 (1) | 91116 | 15 |
| | DΜΨ | 14 | ventricosa | I | 15 | 1 | 16 |
| | DCuM1 | 21 | juvenalis | l | I | H | , , |
| Chic | Cha | C++ | unidentified | ſ | 1 | 12 (2) | 12 |
| | | | | Total | 32 | 26 | 129 |
| | | | | | | | |

1) According to Kihara (1949 and 1957).

²⁾ Numbers within brackets stand for samples with seedlings of the two types of reaction; they were included in the column corresponding to the prevalent type.

³⁾ Type of intermediate reaction, with limited growth of mycelium.

- a) most of Aegilops species appear to be resistant to wheat mildew,
- b) there are, however, some diploid species with one or more susceptible strains.

 Genomes A and D of hexaploid wheat are clearly identified and recognized but the identity of genome B is not yet firmly established, in spite of amount of accumulated evidence. Correlatively, the following question arises: which is the source of mildew susceptibility as found in cultivated wheat?

It is well known that mildew susceptibility is almost a general condition in all cultivated wheat varieties. Inversely, most diploid wheats are resistant to this pathogen. Ae. squarrosa (genome D contributor) possesses some susceptible strains, though differences between varieties strangulata and the others resemble those found by Hiratsuka (1959) with Puccinia graminis and P. triticina; the question of which Ae. squarrosa variety is the most probable ancestor, remains unsolved.

Even considering that this would account for hexaploid susceptibility, the one corresponding to tetraploid condition would still remain unexplained. It would be difficult to assume that a previous non-existing factor in a tetraploid, could arise in it, transmitted through the hexaploid which has been, in turn, partly built by the said tetraploid. Furthermore, some authors consider (e. g. Sarkar and Stebbins 1956) that "the genes for different characters in the emmers (tetraploids) are all or mostly derived from the same genes existing in the ancestral diploids" and not from mutation.

Section Sitopsis has been considered contributor of genome B because of cytological as well as morphological reasons. But it has not been yet conclusively shown which is the most probable donor species. Within the four (or three if Ae. sharonensis is included in Ae. longissima) species represented in Table 1, two were discarded for different reasons; Ae. speltoides (Sarkar and Stebbins 1956) and Ae. bicornis (Sears 1956) remain as probable constituents of cultivated wheat. The latter would have additional support from its susceptible reaction to mildew as compared with the resistance found in all Ae. speltoides strains tested in this work.

Table 2. Sources of Aegilops strains employed in this work

- 1. Dr. R. Riley: Plant Breeding Institute, Trumpington, Cambridge (England).
- 2. Jardin Botanique de l'Ecole Nationale d'Agriculture de Grignon, (S. et O.) (France).
- 3. Dr. R. Ecochard: Versailles (S. et O.) (France).
- 4. Dr. B. C. JENKINS: Winnipeg (Canada).
- 5. Dr. E. R. SEARS: University of Missouri (U.S.A.)
- 6. Dr. Oehler: Station Fédérale d'Essais Agricoles, Lausanne (Switzerland).
- 7. J. Parisiaos (Cyprus).
- 8. Station Centrale de Génétique et d'Amélioration des Plantes, Etoile de Choisy, Versailles (S. et O.) (France).
- 9. Department of Agricultural Botany, University of Reading (England).
- 10. Ecole National d'Agriculture de Grignon (S. et O.) (France).

Nuclear DNA and the evolution of wheat 1)

H. RESS and M. R. WALTERS
Department of Agricultural Botany, University of Wales, Aberystwyth

The analysis of nuclear DNA variation provides a useful approach to investigating ancestry. In the wheats it provides new evidence on the diploid source of genomes comprising the cultivated polyploid forms. It shows also that, in this group at least, the hybridisation and subsequent polyploidy are accomplished without appreciable alteration in the DNA content of individual chromosomes. A previous report by Pai, Upadhya, Bhaskaran and Swaminathan (1961) of a chromosome diminution of the order of about 30 per cent. In the wheat polyploids in shown to be incorrect as indeed is also made clear in a subsequent report by two of the above authors (Upadhya and Swaminathan 1963). There is thus no evidence in wheat for a DNA diminution or for the associated change in the structural organisation of chromosomes inferred in polyploid Hemiptera (Schrader and Hughes-Schrader 1956, 1958).

There are, of course, impediments to beware of in applying this kind of DNA analysis to problems of ancestry. The first relates to the accuracy of the DNA determinations and hence to the precision of the analysis. In this respect a most obvious precaution is to make all preparations in as near standard conditions as possible. This means that wherever possible the material from all species or types selected for comparison should be fixed, stained and scored together in the same batch. This is particularly important in eliminating variation in Feulgen staining, one of the chief sources of error variation.

Another objection that could be made to the method and to the validity of the conclusions derived from it is that the samples or varieties of the species used may not be typical (see UPADHYA and SWAMINATHAN, loc. cit.). Short of making widespread surveys within cultivated and other species there is no certain way of telling to what extent the objection is valid. At the same time it was shown that, where tested, no significant differences in DNA amount occurred between different species known to have the same genome constitution. It is therefore a reasonable assumption that DNA differences between varieties within species are not likely to be greater, and hence, likely to be negligible.

¹⁾ Cited from Heredity 20, Part 1, pp 73-82. 1965, with understanding of the authors.

Conclusions and summary:

- 1. Comparisons are described of ₂C nuclear DNA amounts, measured by Feulgen photometry, in cultivated and related species of wheat.
- 2. DNA amounts were the same in species of similar genome constitution investigated, viz. AA or AABB.
- 3. Triticum timopheevi, usually classified AAGG, has a lower nuclear DNA amount than T. durum (AABB).
- 4. Aegilops speltoides, on the basis of DNA comparisons, is a more likely contributor of the B genome found in the cultivated AABB tetraploids and AABBDD hexaploid than Ae. becornis or Agropyron triticeum.
- 5. There is no evidence of appreciable change in nuclear DNA subsequent to the hybridisation and polyploidy by which the cultivated wheats arose.

References

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Schrader, F. and Hughes-Schrader, S. 1956. Polyploidy and fragmentation in the chromosomal evolution of various species of *Thyanta* (*Hemiptera*). Chromosoma 7: 469-480.

_____, ____ 1958. Chromatid autonomy in Banasa (Hemiptera: Pentatomidal). Chromosoma 9: 193 - 200.

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Radiosensitivity in pollen grains of Triticum and Aegilops

T. MABUCHI and S. MATSUMURA National Institute of Genetics, Misima, Japan

Relation between radio-sensitivity and ploidy or genome constitution was investigated in several species of *Triticum* and *Aegilops*. Anthers before dehiscence were irradiated by X-rays (173 kVp, 25 mA, 1 mm Al filter) at 1, 2 and 3 kr with dose rate of 200 r/min. Pollen grains were dusted on the emasculated florets of the same species. Seed set, mean grain weight and germination were recorded.

Seed setting was noticeably impaired even at 1 kr and reduced to $24\sim75\%$ and $3.1\sim25\%$ of control at 2 and 3 kr, respectively. Germination rate also decreased to $35\sim88\%$ of control at 1 kr and was markedly affected $(3.3\sim22.5\%)$ at 2 kr. Percentage of viable seedlings (per cent success of crossing) was calculated from seed setting \times germination rate. It was for diploid Triticum and Aegilops species about 25% of control at 1 kr, and for several species of tetra- and hexaploids it was ranging from 30 to 55%. It is, therefore, concluded that diploid species are most sensitive to radiation, T. monococcum as well as Ae. squarrosa, but there is no clear difference between tetra- and hexaploid Triticum species.

Radiosensitivity of aged wheat seeds

Seiji Matsumura and Taro Fujii National Institute of Genetics, Misima, Japan

Difference in radiation susceptibility between aged and fresh seeds was examined. 7.5 and 15 kr were applied to seeds of diploid wheat. No germination was observed in the control lot of two year old seeds, and we had no seedlings for irradiation. Non-irradiated one year old seeds showed very low germination rate; a few seeds germinated in the 7.5 kr lot and no germination was observed in the 15 kr lot. Unfortunately germination rate of fresh seeds was as low as 60%; 7.5 kr of γ -rays did not much reduce it (54 per cent), and a few seeds germinated in the 15 kr lot. Thus, the decrease in germinability by irradiation of one year old seeds was in the whole series markedly lower than that of fresh seeds. A similar tendency was also observed in seedling length which was measured 14 days after sowing, and the decrease of survival rate in April was also marked in the one year old series as compared with that of fresh seeds.

10 and 20 kr of γ-rays were given to tetra- and hexaploid seeds taking into consideration their higher resistance to radiation than that of diploid seeds. nability of two year old seeds was very low in both, tetra- and hexaploid lots, and no germination was observed at 20 kr irradiation. But germination rate of control lots was better for younger than for older seeds. This trend was marked in irradiated lots. For instance, germination rate in 20 kr lots of one year old tetraploid seeds was 11.4% amounting to index 55 when the index of 100 is given to the germination rate in the control lot. On the other hand, 68% germinated in the 20 kr lot of fresh seeds, Seedling length and survival rates the index being 73 against that of the control. also showed a similar tendency. In the hexaploid species, a similar decrease of germinability was observed both in irradiated fresh and in one year old seeds. Namely, sensitivity was almost the same in fresh and in one year old seeds of this species From the results, it can be said whereas two year old seeds had higher sensitivity. in general that older seeds have higher radiation susceptibility than younger ones.

The decrease in germinability due to age was largest in diploid wheat and smallest in hexaploid wheat. We may assume from the results that at a higher ploidy level, we may expect a higher stability not only in respect to radiations but also to the physiological changes caused by aging.

II. NEWS

Interim Report of a Project under Japan - U. S. Scientific Cooperative Program

With the aid of a grant from the Japan Society for the Promotion of Science as part of the Japan-U. S. Cooperative Science Program, a project entitled "Genetic study on phylogenetic differentiation of cultivated wheat in the Pacific Region" has been started on September 1, 1964. This project aims to elucidate phylogenetic relationship between Japanese and U. S. wheat populations through the comparative genetic study of wheat varieties in those two countries. The project is expected to be carried out for three years until the end of August, 1967.

I. Japanese Member

| Matsumura, Seiji | National Institute of Genetics, Misima |
|---------------------|--|
| YAMASHITA, Kosuke | Kyoto University, Kyoto |
| Mochizuki, Akira | Hyogo Agricultural College, Sasayama |
| Tanaka, Masatake | Kyoto University, Kyoto |
| Sasaki, Mutsuo | Tottori University, Tottori |
| Окамото, Masasuke | Shiga Agricultural College, Kusatsu |
| Nishikawa, Kozo | Gifu University, Gifu |
| Muramatsu, Mikio | Kihara Institute for Biological Research, Yokohama |
| TSUNEWAKI, Koichiro | Kyoto University, Kyoto |

II. Research Plan

- 1. Study from biochemical standpoint
 - (i) Nucleic acid level Quantitative analysis of DNA content (NISHIKAWA)
 - (ii) Protein level Analysis of species-specific proteins (MATSUMURA)
- 2. Study from the cytogenetic standpoint
 - (i) Nucleic differentiation
 - (a) Gene level Distribution of genes controlling dwarfing, necrosis, chlorosis etc. (Tanaka, Nishikawa, Muramatsu and Tsunewaki)

- (b) Chromosome level Differentiation of chromosome structure (SASAKI and OKAMOTO)
- (c) Genome level Production of aneuploid series of Japanese varieties (Mochizuki)
- (ii) Cytoplasmic differentiation Reciprocal nucleus-substitution between Japanese and American varieties (Tsunewaki)
- 3. Study from the radiological standpoint Variation of radioresistance and analysis of pertinent genetic and physiological factors (Matsumura)
- 4. Study from the morphological standpoint Survey of morphological characters and analysis of genetic factors controlling them (Yamashita and Tanaka)

III. Research Activities

1. Biochemical study

- (i) Nucleic acid DNA contents per nucleus of a common wheat (Chinese Spring), a synthesized hexaploid wheat (ABD No. 1), and its parental species were microspectrophotometrically measured using resting nuclei in embryos. According to the result obtained, there exists the following relationship between their DNA contents; (Chinese Spring) = (ABD No. 1) = (T. dicoccoides + Ae. squarrosa var. typica).
- (ii) Protein Endosperm protein and amylase in embryos have been studied by the cyanogum method of electrophoresis. Seven bands for the former and eight bands for the latter were separated and found to be different quantitatively or qualitatively among various species of wheat and its relatives. Common wheat appeared in this regard to be comparable to the combination of emmer wheat and Ae. squarrosa. Einkorn and Ae. squarrosa were markedly different from each other.

2. Cytogenetic study

(i) Gene distribution — Distribution of genes controlling necrosis and chlorosis has been investigated in Japanese and U. S. wheat populations. No difference was found on the chlorosis genes, while distribution pattern of necrosis genes was quite different between the two populations; Ne₁ gene predominates in Japan, while Ne₂ is prevalent in U. S. A.

In order to clarify the genic system controlling the second type of necrosis (Net-system), F_1 hybrids between Chinese Spring (net_1net_2) monosomics and Prelude (Net_1net_2) were test-crossed to ABD No. 1 (net_1Net_2) and the resulting hybrids are under observation.

Distribution of complementary genes, Dw_1 and Dw_2 , for dwarfness is also under investigation by making crosses between Japanese and U.S. wheats to two testers, T. spelta (Dw_1dw_2) and ABD No. 1 (dw_1Dw_2) .

Four varieties of both Japan and U.S.A. were crossed to Chinese Spring monosomics in order to investigate genes for waxiness of foliage.

- (ii) Chromosome differentiation Mono-5A, 5B and 5D of Chinese Spring were crossed to seven Japanese varieties. Chromosomes 5A, 5B and 5D of these Japanese varieties will be studied in those F₁'s in regard to their morphology and will be compared with those of American varieties.
- (iii) Genomic differentiation As a means of investigating genomic differentiation between Japanese and American wheats, nullisomic series of Japanese wheat will be compared to that of American variety. Since no monosomic and nullisomic series are available in any Japanese variety, Norin 10 and Shinchunaga have been backcrossed several times to Chinese Spring monosomics. Completion of their monosomic series is expected in a near future. Several nullisomics are already obtained and compared with those of an American variety, Wichita. At this moment no remarkable difference was detected between them.
- (iv) Cytoplasmic differentiation Six varieties of both Japan and U. S. A. were crossed in diallel combinations. These F₁ hybrids will be backcrossed in order to obtain reciprocal nucleus-substitution lines.
- 3. Radiological study Up to date, the investigation is limited to the radiological characterization of some representative wheat species. Einkorn, emmer, timopheevi and common wheat, including wild and cultivated forms, were grown in γ-greenhouse. Einkorn wheat was the most sensitive under chronic irradiation, while the other three groups of wheat were not remarkably different from each other.
- 4. Morphological study Using a number of strains or varieties of Asian or American origin, morphological investigation has been undertaken in order to identify their "taxonomic" variety. In Japan erythrospermum and ferrugineum races predominate over all the other races, while in America graecum and erythrospermum are common.

IV. Activities for the Information Exchange

1. Kansas meeting - On July 5, 1964, Wheat Genetics Planning Session was held

- in Kansas City, Missouri; attendants were Dr. H. Kihara from Japan, Dr. E. R. Sears, Dr. E. G. Heyne, Dr. J. W. Schmidt, Dr. W. Q. Loegering, Dr. C. Konzak and Dr. N. P. Neuretter from U. S. In this meeting cooperative research relationships between U. S. and Japanese scientists in the general area of wheat genetics were discussed.
- 2. Dr. C. Konzak visiting Japan Coming to Japan in March, 1955, he discussed the cooperative research plans between his group and the Japanese members. Two formal sessions were held, i.e., at the National Institute of Genetics on March 5, and at the Kyoto University on the sixth.
- 3. Dr. K. Nishikawa visiting U. S. After completing his duty as a research associate in Dr. Sears' Laboratory, he visited Dr. Heyne, Dr. Schmidt and Dr. Konzak, and discussed the future plans of the present project.
- 4. Internal meetings At the start of this project, all Japanese members gathered on August 31, 1964 at the National Institute of Genetics, setting up a definite working program to be carried out for the 3 year period of the project. On March 13, 1965, the second meeting was held in the same institute to report the results of the first one-year term and to discuss the plans for the second year. The third meeting was held on February 18, 1966, where the results obtained in 1965 was reported. The research activities described above are based on the reports of the last two meetings. (S. M.)

Outline of the Kyoto University Botanical Expedition to the Caucasus, May - August, 1966

(under planning)

K. YAMASHITA Biological Laboratory, Kyoto University, Kyoto, Japan

1. Members

- 1) Hitoshi Kihara, D. Sc., Director, National Institute of Genetics
- 2) Kosuke Yamashita, D. Ag., Professor, Biological Laboratory, Kyoto University
- 3) Masatake Tanaka, D. Ag., Assistant Prof. Faculty of Agr., Kyoto University
- 4) Sadao Sakamoto, D. Ag., Researcher, National Institute of Genetics

2. Objects

The aim of the expedition is to continue our work on the origin of wheat carried out since many years in Japan, and to collect new materials urgently needed for the solution of many problems.

In the Caucasus fifteen cultivated and wild wheats are known, including six It is the most important region from the view point of the origin of cultivated wheats. Also, many wild wheat relatives are expected to be abundant there. Two successful earlier Japanese botanical expeditions have been undertaken from the same view point. In 1955 the "Scientific Expedition to the Karakoram and Hindukush (K.U.S.E. 1955)" was organized by Dr. H. Kihara as its leader. During this expedition various cultivated forms of wheat and Aegilops species were collected in Pakistan, Afghanistan and Iran. Particularly, the distribution and variations of Aegilops squarrosa, the donor of D genome to bread wheat (Triticum aestivum), were exhaustively investigated. The secound expedition, "Botanical Mission of the University of Kyoto to the Eastern Mediterranean Countries", led by Dr. K. Yamashita was sent in 1959 (B.M.U.K. 1959). Its chief aim was the study of the distribution and differentiation in Eastern Mediterranean countries of the Sitopsis section of Aegilops, especially Ae. speltoides, the putative donor of B genome to emmer and bread wheats.

Based on the results of those two expeditions covering Central Asia and the Mediterranean countries, it became imperative to send a botanical mission to the Caucasus in order to resolve newly arisen problems. As the new trends in the breeding of wheat, the collection of living materials became also important for the preservation of various genotypic and plasmatic characters.

3. Financial Supports

The present expedition is financially supported by the Research Grant from the Ministry of Education and by personal contributions.

4. Tentative Itinerary of the Expedition

The present expedition will start at the end of May and will take the whole of June, July and will return in the middle of August, 1966. The area of the expedition will cover the Republics of Georgia, Armenia, Azerbaijan and Dagestan.

| May | 21 (1966) | Lv. | Tokyo | |
|------|----------------|-----|------------|---|
| | 22 | Ar. | Copenhagen | Studies of the herbaria in the museum |
| | 24 | Lv. | Copenhagen | |
| | | Ar. | Leningrad | Visit to the Botanical Institute for the studies of the herbaria, and for the arragement of the detailed sche- dule of the trip to the Caucasus Regions |
| May | 27 | Lv. | Leningrad | |
| | | Ar. | Moscow | Official visits to various organizations for the arrangements of the details |
| | 29 | Lv. | Moscow | |
| | | Ar. | Erevan | |
| July | 30 ~ August 10 | 0 | | Observation and collection of wheats and <i>Aegilops</i> in Tbilisi, Kutaisi, Kirovabad and other areas in the Caucasus Regions |
| Augu | st 10 | Lv. | Erevan | |
| | | Ar. | Moscow | Official visits to various organizations for final arrangement of the return trip |
| | 13 | Lv. | Moscow | |
| | 15 | Ar. | Tokyo | |

5. Pending Questions

- a. Relationships between two wild emmer wheats, Palestinian T. dicoccoides, Caucasian T. araraticum (= T. armenianum) and their relation to T. timopheevi.
 - b. Distribution, if any, of T. dicoccoides in the Caucasus.
- c. Origin of emmer wheat based on the ecological characteristics of *T. aegilo-poides* or *T. monococcum*, the donors of A genome, and *Ae. speltoides*, the putative donor of B genome to emmer wheat.
- d. Geographical distribution of six endemic species: T. araraticum, T. carthlicum, T. paleocolchicum, T. timopheevi, T. macha and T. zhukovskyi.

6. Collections expected

- a. Collection of the following 15 wheat species: T. aegilopoides, T. thaoudar, T. monococcum, T. araraticum, T. dicoccum, T. durum, T. turgidum, T. polonicum, T. carthlicum, (= T. persicum), T. paleocolchicum (= T. georgicum), T. timopheevi, T. macha, T. vavilovii, T. aestivum, T. compactum and T. zhukovskyi.
- b. Addition to our sortiment of the following Aegilops species: Ae. speltoides, Ae. squarrosa, Ae. triuncialis, Ae. biuncialis, Ae. triaristata, Ae cylindrica, etc.
- c. Collection of wild plants of the following genera closely or distantly related to Triticum and Aegilops: Agropyron, Elymus, Eremopyrum, Haynaldia, Henrardia, Heteranthelium, Hordeum, Secale and Taeniatherum.

Announcement for further issues

WIS Nos. 23 and 24 will be published during the fiscal year from April, 1966 to March, 1967. Manuscripts for those issues are accepted any time and they will go to press in sequence as soon as they cover planned pages of each number. WIS is open to all contributions regarding methods, materials and stocks, ideas and research results related to genetics and cytology of *Triticum*, *Aegilops*, *Agropyron*, *Secale*, *Haynaldia* and related genera. The manuscripts should not exceed 3 printed pages. List of stocks is exempted from this page limit. Illustrations have hitherto not been accepted, but from the next issues, one text-figure (smaller than $7 \, \mathrm{cm}^2$) will be accepted, if indispensable.

Communication regarding editorial matters should be addressed to:

K. Yamashita Wheat Information Service Biological Laboratory, Yoshida College Kyoto University, Kyoto, Japan

Acknowledgement

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