LOCATION OF GENES FOR RESISTANCE TO *PUCCINIA HORDEI* OTTH. IN THE BARLEY VARIETIES SUDAN AND REKA 1 AS DETERMINED BY MEANS OF TRANSLOCATION STOCKS

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INTRODUCTION

Inheritance of resistance to Puccinia hordei Otth. (P. anomala Rostr.) the causal agent of leaf rust of Hordeum vulgare L. has been studied by several workers. No one has attempted, however, to assign genes for resistance to this fungus to the linkage groups of barley. The literature on inheritance of resistance to this fungus and the exploration of genes controlling it in the varieties of cereal crops is inadequate in the light of modern cytogenetics.

The objective of this investigation was to assign the genes for resistance to leaf rust found in Sudan and Reka to the correct chromosome pair or pairs by means of crossing them with appropriate translocation stocks.

Reciprocal translocations are very valuable as genetic markers to locate genes in linkage groups and very effective in the study of inheritance of characters which are difficult to analyze by ordinary means. As to the use of translocations in genetic studies, Anderson (1938) reports the following: "Chromosomal interchanges or reciprocal translocations are excellent tools for the exploration of portions of chromosomes where no known genes are available. For most cases it is only necessary to cross any new or unplaced gene with the translocation stock, backcross with the new gene if recessive or with normal stock if dominant. Classification of the new gene character under consideration and semi-sterility will give a direct linkage test with a known point on the chromosome."

REVIEW OF LITERATURE

Mains (1930) undertook the study of the possible physiologic specialization of Puccinia hordei (P. anomala Rostr.). He (1932) demonstrated the occurrence of two physiologic forms (1 and 2) of P. hordei in the United States.

Levine and Cherewick (1952) used 357 isolates of Puccinia hordei and inoculated a number of varieties of barley which had been used as differential hosts for determining the existence of physiologic specialization of the fungus. Of the 16 varieties used, nine were found to be valuable for the identification of the consolidated races. On the basis of the critical interpretation of previously recorded data, Levine and Cherewick constructed two dichotomous keys for the identification of physiologic races of the dwarf leaf rust of barley and described 54 physiologic races.

Moseman and Roane (1959) isolated 39 cultures of Puccinia hordei from barley grown in the United States over a period of three years, 1956 through 1958, and found that 70 per cent were race 4, a fact that is in agreement with previous findings by Levine and Cherewick (1952). Moseman and Roane used the nine differential varieties of Levine and Cherewick and found none of the rust cultures highly pathogenic on four of them, an evidence of lack of pathogenicity on these varieties. The investigators concluded that the similarity in the reaction of certain varieties to infection by these cultures is an indication that these varieties possess the same gene or genes for resistance to these cultures of the pathogen.

According to Levine and Cherewick (1952), race 4 "constituted 28.9 per cent of the isolates obtained from United States specimens, 31.3 per cent of those from Canadian specimens and 29.7 per cent of the grand total."

Hey (1931) in Europe and Mains and Martini (1932) in North America have found several resistant varieties.

Waterhouse (1927) in Australia was the first to report studies on the inheritance of resistance to leaf rust of barley. In crosses of susceptible Cape, Skinless, and Kinver with resistant Manchuria, Virginia Hooded and _Hordeum distichum rampani typica_, resistance appeared to be completely dominant in F_1. Segregation in F_2 and F_3 indicated a single factor pair difference between resistant and susceptible varieties in all crosses.

Henderson (1945) used nine resistant varieties and one susceptible selection (II-34-35) for a genetic study of inheritance of reaction to _Puccinia hordei_. The results obtained in all resistant x susceptible crosses indicate that each resistant variety possessed a single, incompletely dominant factor for resistance. In crosses involving resistant parents, Henderson found eight to carry the same factor as shown by lack of segregation in their progeny. The eight varieties are Weider (C.I.1021), Bolivia (C.I.1257), Purple Nepal (C.I.1373), Modia (C.I.2483), Morocco (C.I.4975), Barley 305 (C.I.6015), Ricardo (C.I.6306), and Marco (C.I.5647). The investigator found, moreover, that the ninth variety, Estate (C.I.3410), had a different factor for resistance, as crosses between it and the other eight resistant varieties segregated in a ratio of 15 resistant: 1 susceptible. He concluded that at least two loci are involved in determining reaction to leaf rust, but the majority of varieties studied carried a single, incompletely dominant factor conditioning resistance. The two loci found were independent of each other. Henderson (1945) gave these two factors the symbols *Pa* and $\underline{Pa}_1$. There were distinct differences in the degree of resistance exhibited by the varieties studied which had a factor for resistance at a common locus. The investigator suggested that this was due "either to modifying factors present in some varieties or to the existence of multiple allelic series at the first resistant locus." The worker showed that the *Pa* factor for resistance to leaf rust was independent of the factors *Nn* (covered *vs*. naked caryopsis) in Linkage Group III, *Kk* (hooded *vs*. awned) and *Bl bl* (blue *vs*. non-blue aleurone) in Linkage Group IV, *Rr* (rough *vs*. smooth awn) and *Ss* (long *vs*. short haired rachilla) in Linkage Group V while the $\underline{Pa_1}$ factor for resistance to leaf rust was independent of *Bl bl* in Linkage Group IV and *Rr* in Linkage Group V. The characters just mentioned are listed in the various linkage groups in a summary published by Robertson *et*. *al*. (1947).

Honecker (1945) reported partial linkage between the number of rows and certain genetic components of field resistance to mildew and rust in barley.

In later studies of inheritance of resistance, Waterhouse (1947) found that in crosses of resistant x susceptible varieties, the F_1 indicated complete dominance of resistance. The F_2 and F_3 populations verified the simple factor hypothesis. Resistance appeared completely dominant in crosses with moderately resistant varieties and conditioned by a single factor while moderate resistance appeared to be conditioned by a simply inherited dominant factor, as shown by segregations in crosses with susceptible varieties.

Watson and Butler (1947) studied a hybrid population obtained from a cross of two resistant varieties which differed in the type of resistance. Earlier tests with susceptible varieties indicated that each possessed a single gene which differentiated its type of resistance. The results indicated that the single factor for resistance carried by each is non-allelic and independently inherited. The F_2 ratios obtained, namely 12:3:1, indicated epistasis of one type of resistance over the other. No association between leaf rust reaction and type of lemma appendage, rough _vs._ smooth awns, short _vs._ long haired rachilla and reactions to race 3 of _Erysiphe graminis_ was found. The investigators concluded that there are relatively few loci involved in determining reaction of barley to leaf rust. Zloten (1952) studied the inheritance of reaction to race 4 of leaf rust of barley (_Puccinia hordei_ Otth.) in hybrid populations from crosses made in all combinations between the varieties Montcalm, Ricardo (C.I.6306), Kwan (C.I.1016) and C.I.4219. The results obtained from the crosses studied indicated that reaction to leaf rust was conditioned by factors located at four independent loci. The resistance of each of Kwan (C.I.1016) and Ricardo (C.I.6306) was found to be conditioned by two factors which are completely dominant and independently inherited. Another incompletely dominant gene was found to be common to these two varieties. C.I.4219 exhibited a mesothetic type of reaction but some data showed two factors were involved in crosses of this variety with resistant types. Starling (1955) found that inheritance of resistance to race 4 of leaf rust in crosses of resistant x susceptible varieties involving 13 resistant varieties was conditioned by two independent factor pairs in some varieties while in others resistance was found to be conditioned by a single factor pair. He added that the varieties Juliaca (C.I.1114), Peruvian (C.I.2441), Carre 180 (C.I.3390), Batna (C.I.3391), C.I.3530-2, and Ricardo (C.I.6306) have a factor for resistance at a common locus, supposedly the Pa locus, whereas the varieties Cebada Capa (C.I.6193) and Ariana (C.I.2524) have a factor for resistance at some locus other than the Pa locus. Roane (1959) showed that among the barley rust differentials Reka 1 has a single gene at locus A while Speciale, Sudan, and Oderbrucker each has a single gene at locus C. He also showed the differentials Gold and Lechtaler to have each a single gene at the same locus D, hence disproving an earlier report (1958) that those genes are at different loci.

Origin of Translocations

It is known that translocations occur either spontaneously or as a result of irradiation with x-rays. Kihara and Nishiyoma (1937) showed that crossing-over between duplicated segments which are present in non-homologous chromosomes produced translocations. Sax and Anderson (1933), working on Tradescantia bracteata pollen mother cells, found that interlocking of bivalents at meiosis gave an opportunity for naturally occurring translocations. Kostoff (1938) suggested that spontaneous association between heterochromatic regions could result in translocations. Darlington (1931) stated that accidental entanglement of chromosomes may cause translocations. Burnham (1934) and Anderson (1934) found that crossing-over is greatly reduced in regions near the breakage points. They attributed this to be due, in maize, to the variable and non-homologous pairing in regions near the breaks. Beadle (1937) reported translocations in the progeny of plants that were homozygous for the "sticky chromosome" character in maize while Jones (1938) presented genetic evidence that exchanges may occur between non-homologous chromosomes during the development of the endosperm in the same plant.

Use of Translocations

Gates (1938) was first to observe the attachment of more than two chromosomes to form a ring. He observed this in *Oenothera rubrinervis*. The first investigator to report linkage data between partial sterility and other segregating characters was Belling (1915). Linkage between factors in two linkage groups was first reported by Rhoades (1931, 1933) in maize.

According to Brink and Burnham (1929), the first case of a structural change (interchange) found in the literature was reported by Bridges (1923) in *Drosophila*. Belling (1926) was first to report semisterility as being due to a translocation and the term semisterility seems to have originated with Belling, who applied it to the phenomenon in *Stizolobium*.

Brink (1927) found the same condition in corn. He reported 1115 normal to 1190 partially sterile plants and 1520 to 1154 aborted ovules. He suggested for an explanation of partial sterility in maize that a portion of one chromosome has become attached to a member of a non-homologous pair. In this case it was assumed that all spores receiving the "disjoined" section in duplicate or lacking it altogether are abortive.

Morgan, Bridges, and Sturtevant (1925) reported that cases occur in Drosophila where part of a chromosome is attached to another homologous chromosome. According to Muller (1929), the first case in which both genetic and cytological evidence of a translocation were combined in Drosophila was provided by Stern (1926). In this case a portion of the Y chromosome had become attached to the X chromosome.

Painter and Muller (1929) studied the cytology and genetics of induced translocations. Muller and Altenburg (1930) induced translocations in Drosophila by x-rays. Belling (1914) reported the condition of partial pollen sterility in crosses of varieties of the Florida velvet bean, Stizolobium deeringianum Hort. Later (1926) he interpreted the 1:1 segregation of normal vs. partially sterile individuals in the F_2 on the basis of the chromosome behavior in a reciprocal translocation. Conclusive proof of the chromosome interchange theory to explain the phenomenon of partial sterility was given by McClintock (1930) who demonstrated cytologically that a complex configuration of four chromosomes was found during meiosis in a partially sterile individual. Based on the chromosome interpretation of the phenomenon, an individual heterozygous for an interchange can produce only two types of viable gametes, one containing both interchange chromosomes and the other having the two corresponding normal chromosomes. The two pairs of non-homologous chromosomes involved in the interchange thus behave as a single unit and the intensity of linkage between a gene and the point of interchange can be computed as a function of the frequency that the gene is associated with the interchange or the normal gametes. Taking this into consideration, data have been gathered identifying different translocations and placing the interchange break on genetic maps. In this respect, McClintock (1931) placed the gene order of the genes c, sh, and wx in *Zea mays* L. with reference to a cytologically known point in the chromosome.

In 1929 Brink and Burnham found that reciprocal crosses between normal and partially sterile maize plants gave equal numbers of normal and partially sterile offspring. Self-pollination of partially sterile individuals likewise produced the same two classes in the same proportion. Burnham (1930) reported genetical and cytological studies of partial sterility in maize. Rhoades (1933) investigated cytogenetically a reciprocal translocation in maize. Joachim (1947) adapted the product method for the determination of linkage intensities when there are four F_2 phenotypic classes.

Anderson (1935) pointed out that translocations are of special value in locating a given gene or genes on the observed chromosome, in understanding the linkage relationships of conspicuous markers on the chromosomes, and in providing means of controlling desired portions of chromosomes. In order to identify a chromosome by means of translocations, new interchanges were crossed with a series of tester stocks each homozygous for a known interchange. The F_1 plants were examined for chromosome configurations at diakinesis. When two interchanges are combined in heterozygous form, a ring of six chromosomes is observed at diakinesis if one chromosome is in common while two separate rings of four are obtained if the two interchanges have no common chromosome.

Translocations have demonstrated the relations between chromosomes and genes, and between chromosomes and linkage maps. Chromosomal interchanges have furnished information on chromosome behavior which resulted in marked advances in cytology. For example, in maize Creighton and McClintock (1931) and Brink and Cooper (1935) showed that when genetic crossing-over occurs, a cytological crossing-over occurs also. Stern (1931) has shown the same phenomenon in *Drosophila*.

Translocations are being studied quite extensively in *Zea mays* L. and translocation stocks involving all chromosomes are now available. Translocations are of value as genetic markers to locate genes in linkage groups and are quite effectively used to study the inheritance of characters which are difficult to analyze by ordinary means. Burnham and Cartledge (1939) found a linkage between partial sterility (resulting from a translocation) and resistance *vs.* susceptibility to *Ustilago zeae* (Bekm.) Unger.

In maize when a single translocation is involved in a cross, normally the pollen of the partially sterile progeny will show 50 per cent sterility. In similar crosses of barley, laboratory observations have shown that the per cent of pollen sterility is less than 50 per cent.

Smith (1941) reported two translocations in Hordeum vulgare which occurred naturally. One of them showing 67.2 per cent of the configurations as rings was used in a study of the application of the product method to calculate linkage intensities from F_2 data involving an interchange as developed by Joachim (1947). Jones (1944) presented evidence that outgrowths on the surface of mature kernels of maize resulted when certain regions of chromosome 5 came into contact with others as the result of translocations.

Translocations have been used to identify the linkage group carried by each chromosome, and the locus of the centromeres in the genetic linkage groups. The latter case has been particularly studied in maize by Anderson and Randolph (1945) using homozygous interchanges to determine which genes in the two linkage groups are linked and which are not. If a linkage is found which does not exist in normal stocks, one of the genes is concluded to be in a translocated segment while the other is in the segment which carries the centromere.

In case of direct segregation, the expected reduction in crossing-over in the interstitial segments is indicative of the positions of the centromeres. Hanson (1952) presented data which showed reduction in crossing-over for regions marked by three genetic factors. This was shown for three interchanges with breaks at different positions in one linkage group.

Burnham (1946) reported the appearance of "long" chromosomes at metaphase I of microsporogenesis in one of several lines of Mars barley produced by x-ray treatment. He explains the "long" appearance as the result of failure of one of the two arms of a chromosome to be associated with its homologue. Under these circumstances, a plant with the chromosome abnormality showed 15 to 20 per cent pollen abortion.

Smith (1948) studied three ring-forming plants (heterozygous translocations) and found an average of 66 per cent fertility while normal plants averaged 93 per cent. He concluded that his failure to get 50 per cent fertility was due to disjunctional segregation of the chromosomes. This explanation was questioned by Burnham (1948). The latter stated: "If a species with a single ring of four chromosomes had completely directed or alternate segregation (disjunctional), only in those with genetically short interstitial segments should spore abortion be low or absent. Sterility might be expected to reach a maximum of 50 per cent as the length of this segment increased." Since 50 per cent pollen sterility is no~~w~~t found in barley plants heterozygous for a single translocation, the term "partially sterile" should be used instead of "semisterile."

Burnham (1948) described a naturally occurring semisterile line of maize which usually has a ring of four chromosomes at diakinesis. He showed it to be heterozygous for an interchange between chromosomes 1 and 7 and gave it the designation T1-7a. At mid-prophase in plants heterozygous for the interchange, a cross-shaped synaptic figure was formed. Longley (1950) used cytological study at pachytene to identify a large number of interchanges in maize. This procedure identifies the chromosomes involved and the positions of the translocation breakage point. Miller (1951) used translocations to study the inheritance of oil content in corn. Roman and Ullstrup (1951) used the A-B translocation method in which functional deficient gametes were used to place genes within specific chromosomal segments. They applied the method to the Hm hm alleles concerned with the reaction of maize to infection by Helminthosporium carbonum Ull. race I and showed that the locus is in the distal seven-eights of the long arm of chromosome I, about 22 recombination units from the centromere.

Of equal importance in the use of translocation is the work of Paterson (1952), who used the waxy gene in maize to determine whether, "in translocations involving chromosome 9, the break in 9 was between the wx locus and the centromere or in the other arm. For this test, it is possible to use only interchanges which involved chromosome 9 and had one of the interchanged pieces short enough so that the pollen deficient for it was partially filled." In the interchange heterozygote, Paterson found it necessary to have the wx allele on the normal chromosome and Wx on the interchanged chromosome. He concluded, therefore, that if the break were in the arm not carrying the locus, the partially filled grains were mostly wx; but if the break were between it and the centromere, the grains were mostly Wx.

In barley, it was found by Hagber, Nybom, and Gustafsson (1952) that three of the erectoides mutants are inseparable from different translocations.

Rhoades and Dempsey (1953) studied individual plants of maize carrying two inverted chromosomes, one of the latter carried a deficiency-duplication, and placed the $\underline{Lg_2}$ locus 12 recombination units from the proximal break of the inversion.

Interchange markers have been used by several workers in locating new genes and in studying the inheritance of complex characters. Ibrahim (1954) located three genes for the resistance of one inbred line of maize to the European Corn borer. In barley, Nybom (1954) reported complete linkage between an alboviridis mutant and a translocation.

Burnham _et al_ (1954) reported a group of translocations produced by x-raying, followed by intercrossing and cytological examination of the F_1's. Burnham (1954) developed a set of five tester translocations which he subsequently used to determine the chromosomes involved in terms of the temporary letter designations a, b, c, d, e, f, and g. Based on the cytological examination of the F_1's at meiosis, Burnham (1954) described a method to identify a chromosome involved in a translocation. This is done by selecting a tester set by which the chromosomes in any new translocations may be identified in terms of those in the tester set. A ring of six indicates that one chromosome involved in the two interchanges is the same; two rings of four indicate that the two chromosomes involved in one are different from those involved in the other. If the same chromosomes are involved, only pairs of a ring of four will be observed depending on whether the breaks are at a similar or at very different positions. Burnham (1956) concludes that "in the higher plant species with completely directed segregation, the interchange heterozygotes may be identified cytologically by the association of two or more chromosome pairs at meiosis, or genetically by linkage between characters normally independent."

Finally, the work of Ramage and Suneson (1958) showed how translocation stocks can be used to locate a certain gene. The investigators tested the "early" gene in barley against the break-points of 11 chromosomal interchanges and concluded that it is independent of the break-points of 9 of these interchanges but completely linked with the break-point of (f-g) and closely linked with the break-point of (b-g). Ramage and Suneson, in conclusion, found the gene conditioning "earliness" in barley to be associated with the g chromosome.

MATERIALS AND METHODS

In this study, translocation stocks of barley were furnished by Dr. C. R. Burnham, Division of Agronomy and Plant Genetics, University of Minnesota. Several ways by which translocation stocks may arise were mentioned by Burnham (1956). In this study, the translocation stocks were all developed from the variety Mars by x-raying. They are 5051-2(a-b), 5059-5(a-e), 5030-2(b-d), 5034-2(b-f), 5062-3(b-g), 5038-2(c-d), 5056-2(c-e), 5053-6(e-f), and 5057-1(f-g). The letters in parentheses indicate the chromosome pairs, designated by the letters a through f, which are involved in the translocation stocks. Seed of the varieties Reka 1 (C.I.5051) and Sudan (C.I.6489) were furnished by Dr. C. W. Roane of the Department of Plant Pathology and Physiology,— Virginia Polytechnic Institute.

Seed of each of the two varieties Sudan and Reka and of each of the nine translocation stocks described above were planted in six-inch clay pots in the greenhouse for tests of reaction in the seedling stage to race 4 of leaf rust caused by *Puccinia hordei*.

As soon as the seedlings had developed to the first leaf stage, one week to 10 days after planting, they were inoculated with uredospores of race 4 of the leaf rust fungus. Race 4 is very useful for studying the genetics of leaf rust resistance in the barley leaf rust differentials since all except Egypt are resistant to it. The method used in inoculating consisted essentially of wetting the leaves by spraying them with a fine mist of water and wetting agent; namely, Triton X-100, manufactured by Rohm and Haas. The spores of the fungus were mixed with a talcum powder and blown onto the plants. A wet surface was maintained on the leaves during the infection period by putting the plants in a moisture chamber constructed of aluminum frames and covered with a muslin cover for 24 hours. The muslin cover was kept wet by repeated sprinkling with water during the infection period. Humidity was maintained in the moisture chamber by means of a fog machine and the temperature in the greenhouse was maintained between 68°F and 70°F as far as possible. As reported by Waterhouse (1927), a period of 24 hours under conditions of high humidity was satisfactory for infection.

The varieties were classified for reaction to leaf rust about 2 weeks after inoculation. Three separate readings were taken 12, 14, and 16 days following inoculation to make sure that adequate time for expression of reaction in the various lines and changes in the reaction types were taken into consideration.

Classification was made according to pustule type and was similar to the infection types described by Waterhouse (1927), Mains (1930), Straib (1937), d'Oliveira (1939) and others. Five classes of pustules were recognized as suggested by Levine and Cherewick (1952). They can be described as follows:

No pustules developed ————————————type 0

Only a few small pustules visible. Infection chiefly visible as small necrotic flecks without pustules ——————type 1

Pustules rather small - each delimited by a necrotic ring ——type 2

Pustules of moderate size, more numerous than in previous classes, without necrosis, pustules usually surrounded by chlorotic area ——————————————————type 3

Pustules relatively large, numerous, generally not surrounded by chlorosis. No evidence of hyper-sensitive reaction by host plant ——————————————————type 4

Superscript C stands for chlorosis, N stands for necrosis, and + indicates a stronger reaction.

Qualitatively, these classes may be described as follows:

Type 0 = extremely resistant
Type 1 = very resistant
Type 2 = moderately resistant
Type 3 = moderately susceptible
Type 4 = very susceptible

From the aforementioned plantings, reciprocal crosses of the two resistant varieties with the nine translocation stocks were made.

After all crosses were completed, the hybrid seeds were planted in the greenhouse. Individual F_1 plants of each cross were inoculated and classified for reaction to leaf rust as described earlier. The reactions of these plants were expressed in relation to their parents. All F_1 plants were found to be resistant, ranging in type reaction from 0 to 2.

In the early boot stage, all F_1 plants were checked to make sure they were derived by crossing by examining the microspore mother cells for ring-of-four configurations. This was done by staining the microsporocytes with acetocarmine prior to their examination. Young spikes were fixed in Carnoy's solution (6 ml 95 per cent alcohol: 3 ml chloroform: 1 ml propionic acid solution) for further examination if desired.

The mature anthers of the same plants were then checked for pollen sterility. This was done by placing a drop of a 2 per cent aqueous solution of potassium iodide on pollen from mature anthers and examining under a microscope. Sterile pollen grains are shrunken and devoid of starch. Under greenhouse conditions of this experiment, even pollen from normal plants showed a reduced percentage of fertility and only those plants which showed over 10 per cent sterility were classified as partially sterile.

About 80 F_2 seeds from each of the F_1 plants shown to be hybrid were planted in four-inch peat pots in the greenhouse. In the one-leaf stage, inoculations with leaf rust spores were conducted in the manner described earlier. A temperature of 68°F-70°F was maintained in the greenhouse, as far as possible. The classification for reaction to leaf rust was accomplished most easily 13-15 days after inoculation. All type reactions obtained are presented in tables 1 and 6.

After seedling reactions were determined, all F_2 plants were transplanted to the field. Just prior to anthesis, the pollen was examined as pointed out earlier and all F_2 plants were classified as to partial sterility <u>vs</u>. fertility. All plants were numbered so that the partially sterile or fertile plants could be checked as to whether they had been resistant or susceptible in the seedling stage.

When final classification was completed for the F_2 population of each of the 18 crosses under study, all possible phenotypic classes were tabulated and summarized. Chi-square (X^2) was calculated for each segregating character pair (resistant _vs._ susceptible and partially sterile _vs._ fertile) and for every possible dihybrid combination. To calculate recombinations, the product method (Joachim, 1947) was used.

EXPERIMENTAL RESULTS

The varieties Sudan and Reka were resistant to rust. Under greenhouse conditions, the rust reaction of Sudan was 1^N while the reactions of the translocation stocks were $4, 4^C$ and 4^+. These results are presented in table 1. The range of the rust reaction of the F_1 of each cross is presented in table 1. The range of the rust reaction of the F_1 plants of Sudan x a-b and Sudan x c-d is 0-2, that of the F_1's of Sudan x a-e, Sudan x b-g, and Sudan x e-f is 1-2 while that of the F_1's of Sudan x b-d and Sudan x b-f is $1\text{-}2^N$. The rust reaction of the F_1's of Sudan x c-e is 0-1 and that of the F_1's of Sudan x f-g is $1^N\text{-}2$.

The F_1 plants should be heterozygous for the translocations and should be partially sterile. Cytological examination of the microspore mother cells in the F_1 plants revealed the presence of a ring of four chromosomes plus five pairs and all were partially sterile. An F_2 was produced from F_1 plants which were proven to be heterozygous for the translocation. In growing the F_2 generation, the F_1 plants were allowed to self. While barley is normally self-pollinated, the individual spikes of the F_1 plants were covered with glassine bags just before anthesis to prevent the possibility of cross pollination. After a few weeks, the bags were removed and the spikes were allowed to reach maturity. Rust reactions and pollen condition of F_2 of all crosses of Sudan with the translocation stocks are presented in table 1.

Table 1: – Data for reaction to *Puccinia hordei*, race 4, of the parents and F_1 and for partial sterility *vs.* fertility and *P. hordei* reaction of F_2 of crosses of Sudan with nine translocation stocks.[a]

Cross and parental reactions to leaf rust	Range of reaction of F_1	Pollen classification	No. of F_2 plants with indicated reaction to leaf rust[d]						
			Resistant			Susceptible		Total	Grand Total
			0	1	2	3	4		
Sudan (1^N) x a-b(4)[b]	0-2	P.S.	5	11	18	8	2	44	89
		Fert.[c]	2	11	19	7	6	45	
Sudan (1^N) x a-e(4)	1-2	P.S.	12	12	7			31	72
		Fert.	6	11	5	13	6	41	
Sudan (1^N) x b-d(4)	$1-2^N$	P.S.	5	15	8	5	2	35	88
		Fert.	1	20	20	8	4	53	
Sudan (1^N) x b-f(4^C)	$1-2^N$	P.S.	6	13	6	2	5	32	76
		Fert.	3	27	2	6	6	44	
Sudan (1^N) x b-g(4^C)	1-2	P.S.		17	7	1	4	29	66
		Fert.	6	18	5	7	1	37	
Sudan (1^N) x c-d(4^+)	0-2	P.S.	4	13	13		7	37	92
		Fert.	2	4	35	6	8	55	
Sudan (1^N) x c-e(4)	0-1	P.S.	4	12	8			24	64
		Fert.	3	6	12	10	9	40	
Sudan (1^N) x e-f(4)	1-2	P.S.	2	10	15	6	1	34	91
		Fert.	5	14	22	11	5	57	
Sudan (1^N) x f-g(4^C)	1^N-2	P.S.		14	6		4	24	68
		Fert.	12	13	6	5	8	44	

[a] Rust reaction symbols are explained in text.

[b] Reciprocal translocation between indicated pairs of chromosomes.

[c] Partially sterile (P.S.) *vs.* fertile (Fert.) plants.

[d] See text for description of reaction type.

Table 2 shows a sample calculation for F_2 data indicating independent assortment in Sudan x b-g stock while table 3 shows a sample calculation for F_2 data showing incomplete linkage in Sudan x e-f translocation stock. Table 4 is a sample calculation for F_2 data showing a case of complete linkage in cross Sudan x a-e.

A summary of the classification of the reaction of the F_2 of Sudan and the translocation stocks to leaf rust race 4 as well as partial sterility vs. fertility is presented in table 5. The X^2 and P values for the F_2 of each cross of Sudan and the translocation stocks are also given. The data given in table 5 show that the classification of F_2 plants for reaction to leaf rust and partial sterility vs. fertility fit a 3:3:1:1 ratio, with P values greater than 0.05, in crosses of Sudan x a-b, Sudan x b-d, Sudan x b-f, Sudan x b-g, Sudan x c-d, Sudan x f-g, and Sudan x e-f. In these crosses the gene for resistance behaves as if it is independent of the chromosomes involved in the translocation, i.e. chromosomes a, b, c, d, f, and g.

In crosses of Sudan x a-e and Sudan x c-e a ratio of 2 resistant partially sterile: 1 resistant fertile: 1 susceptible fertile was found. This indicates that in both cases the gene for resistance was completely linked to interchanges involving chromosome e. As indicated above, the gene appears to be independent of chromosomes a and c. Therefore, it must be upon the e chromosome.

Table 6 shows the rust reaction of Reka to be 0 while that of the translocation stocks to be $4, 4^c$ or 4^+. Individual reaction ranges of F_1's of all crosses are also included.

Table 2: - Sample calculation for F_2 data showing independent assortment in Sudan x 5062-3(b-g)

Class	Obs.	Exp. (e) 3:3:1:1	Deviation (d)	d^2	d^2/e	$\chi^2=\sum(d^2/e)$	P from χ^2 for goodness of fit
Res. and P.S.[a]	24	24.75	0.75	0.56	0.022	2.038	0.7-0.5
Res. and Fert.[b]	29	24.75	4.25	18.06	0.729		
Sus. and P.S.[c]	5	8.25	3.25	10.56	1.280		
Sus. and Fert.[d]	8	8.25	0.25	0.06	0.007		

[a] Resistant and partially sterile plants

[b] Resistant and fertile plants

[c] Susceptible and partially sterile plants

[d] Susceptible and fertile plants

Table 3: - Sample calculation for F_2 data showing incomplete linkage in Sudan x 5053-6(e-f).

Class	Obs.	Exp. (e) 3:3:1:1	Deviation (d)	d^2	d^2/e	$X^2=\sum(d^2/e)$	P from X^2 for goodness of fit	$\frac{ab}{cd}$ [a]
Res. and P.S.	27	34.125	7.125	50.76	1.48	6.42	0.1-0.05	0.6643[b]
Res. and Fert.	41	34.125	6.875	47.27	1.38			
Sus. and P.S.	7	11.375	4.375	19.14	1.68			
Sus. and Fert.	16	11.375	4.625	21.39	1.88			

[a] $\frac{ab}{cd}$: a = number of resistant, partially sterile plants
b = number of resistant, fertile plants
c = number of susceptible, partially sterile plants
d = number of susceptible, fertile plants

[b] Refers to the recombination value obtained from tables developed by Joachim (1947) and corresponding to ab/cd value.

Table 4: - Sample calculation for F_2 data showing complete linkage in Sudan x 5059-5(a-e).

Class	Obs. Value	Exp. Ratio 2:1:1	$X^2=\sum(d^2/e)$	P from X^2 for goodness of fit	Exp. Ratio 3:3:1:1	$X^2=\sum(d^2/e)$	P from X^2 for goodness of fit
Res. and P.S.	31	36	1.637	0.5-0.30	27	21.629	<0.01
Res. and Fert.	22	18			27		
Sus. and P.S.	0	0			9		
Sus. and Fert.	19	18			9		

Table 5: - A summary of the classification of the reaction of the F_2 plants from crosses of Sudan with the translocation stocks to *Puccinia hordei*, race 4, and for partial sterility *vs.* fertility, with X^2 and P values for goodness of fit to indicated ratio.

Cross	Number of F_2 plants with indicated classification				Total	X^2 for 3:3:1:1 ratio	P from X^2 for goodness of fit
	Res. and P.S.	Res. and Fert.	Sus. and P.S.	Sus. and Fert.			
Sudan x a-b	34	32	10	13	89	1.008	0.8 - 0.7
Sudan x a-e	31	22	0	19	72	21.629	$<$0.01
Sudan x b-d	28	41	7	12	88	4.241	0.3 - 0.2
Sudan x b-f	25	32	7	12	76	2.174	0.7 - 0.5
Sudan x b-g	24	29	5	8	66	2.038	0.7 - 0.5
Sudan x c-d	30	41	7	14	92	4.116	0.3 - 0.2
Sudan x c-e	24	21	0	19	64	23.495	$<$0.01
Sudan x e-f	27	41	7	16	91	6.420	0.1 - 0.05
Sudan x f-g	20	31	4	13	68	7.140	0.1 - 0.05

Table 6: – Data for reaction to *Puccinia hordei*, race 4, of the parents and F_1 and for partial sterility *vs.* fertility and *P. hordei* reaction of F_2 of crosses of Reka and nine translocation stocks.[a]

Cross and parental reactions to leaf rust	Range of reaction of F_1	Pollen classification	No. of F_2 plants with indicated reaction to leaf rust						
			Resistant[d]			Susceptible		Total	Grand Total
			0	1	2	3	4		
Reka(0) x a-b(4)[b]	0-1	P.S.		16	12		8	36	86
		Fert.[c]		15	22	12	1	50	
Reka(0) x a-e(4)	0-2	P.S.	2	17	21	2	7	49	94
		Fert.	9	11	13	10	2	45	
Reka(0) x b-d(4)	1-1	P.S.		13	13	1	4	31	80
		Fert.	4	4	26	14	1	49	
Reka(0) x b-f(4^{c})	0-2	P.S.	1	11	28			40	83
		Fert.	4	9	11	15	4	43	
Reka(0) x b-g(4^{c})	1-2	P.S.	6	9	10	2	7	34	83
		Fert.	3	8	25	10	3	49	
Reka(0) x c-d(4^{+})	$0\text{-}1^{N}$	P.S.		7	23	2	5	37	95
		Fert.		10	31	11	6	58	
Reka(0) x c-e(4)	0-2	P.S.	14	10	8		4	36	67
		Fert.	17	2	1	8	3	31	
Reka(0) x e-f(4)	$0\text{-}1^{N}$	P.S.	1	5	29			35	82
		Fert.	5	7	12	17	6	47	
Reka(0) x f-g(4^{c})	1-1	P.S.	2	6	13	4	3	28	80
		Fert.	4	17	19	10	2	52	

[a] Rust reaction symbols are explained in text.

[b] Reciprocal translocation between chromosomes a and b.

[c] Partially sterile (P.S.) *vs.* fertile (Fert.) plants.

[d] See text for description of reaction types.

The F_1 plants which were heterozygous for the translocations were checked for partial sterility. All F_1's were partially sterile. Cytological examination of the microspore mother cells in the F_1 plants revealed the presence of a ring of four chromosomes plus five pairs. Thus it was demonstrated that all F_1 plants were crosses between translocation stocks and Reka. The classification of F_2 plants from crosses of Reka with the translocation stocks for rust reaction and partial sterility vs. fertility are presented in table 6.

A summary of the classification of the reaction of the F_2 plants from crosses of Reka with the translocation stocks for reaction to leaf rust race 4 and partial sterility vs. fertility and X^2 and P values for goodness of fit to the expected ratios are presented in table 7.

The F_2 data for reaction to leaf rust and partial sterility vs. fertility fit a 3:3:1:1 ratio with a P value greater than 0.05 in crosses of Reka x a-b, Reka x a-e, Reka x b-d, Reka x b-g, Reka x c-d, Reka x c-e, and Reka x f-g (table 7). In these crosses the gene for resistance behaves as if it is independent of the chromosomes involved in the translocation; i.e. chromosomes a, b, c, d, e, and g.

In crosses of Reka x b-f and Reka x e-f a ratio of 2 resistant partially sterile: 1 resistant fertile: 1 susceptible fertile was found (table 7). This indicates that in both cases the gene for resistance was completely linked to interchanges involving chromosome f. As shown above the gene appears to be independent of chromosomes b and e. Therefore, it must be upon chromosome f.

Table 7: - A summary of the classification of the reaction of the F_2 plants from crosses of Reka with the translocation stocks to *Puccinia hordei*, race 4, and for partial sterility *vs.* fertility, with X^2 and P values for goodness of fit to indicated ratio.

Cross	Number of F_2 plants with indicated classification				Total	X^2 for 3:3:1:1 ratio	P from X^2 for goodness of fit
	Res. and P.S.	Res. and Fert.	Sus. and P.S.	Sus. and Fert.			
Reka x a-b	28	37	8	13	86	2.432	0.5 - 0.3
Reka x a-e	40	33	9	12	94	1.425	0.7 - 0.5
Reka x b-d	26	34	5	15	80	6.066	0.2 - 0.1
Reka x b-f	40	24	0	19	83	21.706	<0.01
Reka x b-g	25	36	9	13	83	2.814	0.5 - 0.3
Reka x c-d	30	41	7	17	95	5.911	0.2 - 0.1
Reka x c-e	32	20	4	11	67	6.034	0.2 - 0.1
Reka x e-f	35	24	0	23	82	28.177	<0.01
Reka x f-g	21	40	7	12	80	7.333	0.1 - 0.05

Analysis of the ratio of resistant to susceptible plants irrespective of partial sterility vs. fertility for the F_2 data from crosses of Sudan with all translocation stocks are presented in table 8. X^2 for 3:1 ratio and P values from X^2 for goodness of fit are also included. Data presented in table 9 offers a similar treatment for the F_2 population from crosses of Reka with all translocation stocks. The totals for the crosses with all translocations, as well as each of the F_2 populations, had a satisfactory fit for the expected ratio of 3 leaf rust resistant: 1 leaf rust susceptible. The X^2's for goodness of fit to this ratio were 0.092 and 0.087 for the combined crosses involving Sudan and Reka, respectively.

Table 10 provides a sample calculation for X^2 and P values for deviation from the expected recombination ratios based on actual frequencies of each class from F_2 data for the cross of Sudan x a-b. Data in table 11 summarize X^2 and P values for deviation from the expected recombination ratios based on actual frequencies for F_2 data from all crosses of Sudan with the translocation stocks. Table 12 offers a similar treatment for F_2 data from all crosses of Reka with the translocation stocks. If we consider, however, partial sterility vs. fertility, totaled for the crosses with all translocations, a significant deviation from the ratio 1 partially sterile: 1 fertile plant is observed. A X^2 value of 22.48, with P value less than 0.01, is obtained for the F_2 from all crosses involving Sudan with all translocation stocks (416 Fert./290 P.S.). The F_2 from crosses involving Reka with all translocation stocks (424 Fert./326 P.S.)

Table 8: - Analysis of the classification of the reaction of the F_2 plants to *Puccinia hordei*, race 4, with X^2 for 3:1 ratio and P values for each cross of Sudan and the translocation stocks.

Cross	Resistant Plants	Susceptible Plants	Total	X^2 for 3:1 ratio	P from X^2 for goodness of fit
Sudan x a-b	66	23	89	0.032	0.9 - 0.8
Sudan x a-e	53	19	72	0.072	0.8 - 0.7
Sudan x b-d	69	19	88	0.544	0.5 - 0.3
Sudan x b-f	57	19	76	0.000	$>$0.99
Sudan x b-g	53	13	66	0.988	0.5 - 0.3
Sudan x c-d	71	21	92	0.228	0.7 - 0.5
Sudan x c-e	45	19	64	0.748	0.5 - 0.3
Sudan x e-f	68	23	91	0.004	0.98- 0.95
Sudan x f-g	51	17	68	0.000	$>$0.99
Total	533	173	706	0.092	0.8 - 0.7

Table 9: - Analysis of the classification of the reaction of the F_2 plants to *Puccinia hordei*, race 4, with X^2 for 3:1 ratio and P values for each cross of Reka and the translocation stocks.

Cross	Resistant Plants	Susceptible Plants	Total	X^2 for 3:1 ratio	P from X^2 for goodness of fit
Reka x a-b	65	21	86	0.016	0.9 - 0.8
Reka x a-e	73	21	94	0.352	0.7 - 0.5
Reka x b-d	60	20	80	0.000	$>$0.99
Reka x b-f	64	19	83	0.196	0.7 - 0.5
Reka x b-g	61	22	83	0.100	0.8 - 0.7
Reka x c-d	71	24	95	0.003	0.99- 0.98
Reka x c-e	52	15	67	0.240	0.7 - 0.5
Reka x e-f	59	23	82	0.404	0.7 - 0.5
Reka x f-g	61	19	80	0.064	0.9 - 0.8
Total	566	184	750	0.087	0.8 - 0.7

Table 10: - Sample calculation for determining X^2 value and P value for deviation from the expected recombination ratios based on actual frequencies of each class from F_2 data for cross Sudan x a-b.

F_2 plants with indicated classification	Obs.[a]	Exp.[b] (e)	Dev. (d)	d^2	d^2/e	$X^2=\sum(d^2/e)$	P from X^2 for goodness of fit
Res. and P.S.	34	32.7	+1.3	1.69	0.052	~~0.052~~	0.95 - 0.90
Res. and Fert.	32	33.3	-1.3	1.69	0.051	.397	
Sus. and P.S.	10	11.3	-1.3	1.69	0.150		
Sus. and Fert.	13	11.7	+1.3	1.69	0.144		

[a] Observed number of plants taken from data in table 5 for cross Sudan x a-b.

[b] Expected recombination ratios calculated:

Res. and P.S. 66/89 x 44/89 x 89 = 32.7
Res. and Fert. 66/89 x 45/89 x 89 = 33.3
Sus. and P.S. 23/89 x 44/89 x 89 = 11.3
Sus. and Fert. 23/89 x 45/89 x 89 = 11.7

Table 11: - Chi-square values and P values for deviation from the expected recombination ratios based on actual frequencies for F_2 data from each cross of Sudan with the translocation stocks.

Cross	X^2 deviation based on actual frequencies	P from X^2 for goodness of fit
Sudan x a-b	0.397	0.95 - 0.90
Sudan x a-e	19.610	<0.01
Sudan x b-d	0.090	>0.99
Sudan x b-f	0.275	0.98 - 0.95
Sudan x b-g	0.190	0.98 - 0.95
Sudan x c-d	0.559	0.95 - 0.90
Sudan x c-e	16.113	<0.01
Sudan x e-f	0.635	0.90 - 0.80
Sudan x f-g	1.358	0.80 - 0.70

Table 12: - Chi-square values and P values for deviation from the expected recombination ratios based on actual frequencies for F_2 data from each cross of Reka and the translocation stocks.

Cross	X^2 deviation based on actual frequencies	P from X^2 for goodness of fit
Reka x a-b	0.127	0.99 - 0.98
Reka x a-e	0.886	0.90 - 0.80
Reka x b-d	2.199	0.70 - 0.50
Reka x b-f	22.732	<0.01
Reka x b-g	0.002	>0.99
Reka x c-d	1.350	0.80 - 0.70
Reka x c-e	5.523	0.20 - 0.10
Reka x e-f	23.728	<0.01
Reka x f-g	0.048	>0.99

gave a X^2 value of 12.81, with P less than 0.01. A consideration of table 11 with X^2 values for deviations from expected recombination frequencies based on actual frequencies of each character considered alone for F_2 population from individual crosses shows that a significant deviation is found only in Sudan x a-e and Sudan x c-e (where linkage occurred). Data of all other crosses have a satisfactory fit. Similarly, considering table 12, a significant deviation is obtained only in F_2 from Reka x b-f and Reka x e-f (where linkage occurred). Data of all other crosses have a satisfactory fit. It seems that data from all crosses should be treated and considered separately for any possible deviation from a certain ratio.

Averaged over all crosses, an excess of fertile plants over partially sterile plants was obtained. It is not known if this could be attributed to a possible cytological manifestation of the translocations or the arbitrary choice of 10% sterility used as the dividing point between classifying a plant as partially sterile or fertile. All plants were treated uniformly during classification, which was conducted in the laboratory.

DISCUSSION OF RESULTS

Barley plants which are homozygous for reciprocal translocations, when crossed with those which have a normal complement of chromosomes, are expected to produce F_1 plants which are heterozygous for the translocation and therefore show partial pollen sterility. In the F_2 generation, fertile and partially sterile plants are expected to be produced in a one to one ratio.

When simple Mendelian characters segregate giving a 3:1 ratio which is independent of a ratio of 1:1 for fertile vs. partially sterile, a ratio of 3:3:1:1 is expected for the two pairs of characters in the F_2. This would be expected for a Mendelian character located on a chromosome not involved in the translocation under study. If, however, the gene under consideration is completely linked to one of the chromosomes involved in the translocations, the 3:1 and 1:1 ratios will not be independent, but instead will produce a 2:1:1 ratio of heterozygous dominant, partially sterile; homozygous dominant, fertile; and homozygous recessive, fertile. If linkage is complete no homozygous recessive, partially sterile progeny will be produced. Crossing-over between the locus of the gene and the point of translocation will lead to the production of homozygous recessive offspring which are heterozygous for the translocation and, therefore, partially sterile. From the frequency of this group the recombination percentage can be determined using the product method described by Joachim (1947).

In the cross Sudan x e-f the recombination class, susceptible and partially sterile, was present (table 5). The frequency of their occurrence shows a satisfactory fit in comparison to expectations on the assumption of independent assortment as indicated by a X^2 value of 6.42 which is not significant at a P value of 0.1 but is near the significance level of 0.05. Thus crossing-over must have occurred to the extent of approximately 30 per cent as calculated using the product method (Joachim, 1947). The limited size of the F_2 prevents an accurate estimation of this cross-over frequency and further study should be made. Other tests obtained for chromosome e and f in crosses of Sudan x e-f and Sudan x f-g respectively (table 5) indicated that the gene and the translocation locus may be either independent or loosely linked.

A X^2 value of 0.635, suggesting complete independence, was obtained from the expected recombination ratios based on actual frequencies from F_2 data from Sudan x e-f (table 11). This is in sharp contrast with the X^2 value of 6.42 for the 3:3:1:1 ratio for the same cross (table 5). The distance of the gene from the translocation locus may offer an explanation for the failure of getting a linkage test. No cytological differences were observed in the type of ring configurations that were exhibited by the F_1 plants involving the e-f translocation as compared to configurations exhibited by F_1 plants involving other translocations.

In the cross Sudan x f-g, a higher X^2 value is observed as compared to the previous cross, namely, Sudan x e-f (table 5). The unexpected result may be accounted for by the great distance of the gene for rust reaction on the f chromosome from the breakage point of the translocation. It is very hard to test if a gene is linked once it is far enough from the breakage point of the interchange. Furthermore, the X^2 value for the cross Sudan x b-g provides an evidence that the gene in question is not linked with the g chromosome. Of equal importance in attempting to explain the results given by Sudan x f-g is the limited size of the F_2 population.

In Sudan, the factor for resistance is associated with chromosome e. Linked with chromosome e, Burnham (1957) found the zebra characteristic, which was 14.6 crossover units from the breakage point of c-e translocation. His study was based on 111 F_3 lines. Turcotte (1957) reported that the brittle character in barley is linked with the c-e translocation with a recombination value of 6.2±3.4 per cent.

Chromosome e makes up linkage group and chromosome 4 to which belongs the factor pair Kk (Hooded vs. awned spikes) according to Larter and Leisle (1957). This factor pair is linked to Ml_g ml_g (resistance vs. susceptibility to powdery mildew), according to Hiura and Herta (1957), with a recombination value of 17.8±2.5 per cent.

In the cross Reka x f-g the recombination class, susceptible and partially sterile, was present. The cases of complete linkage exhibited by the crosses Reka x b-f and Reka x e-f give enough evidence that the gene is linked with chromosome f. The limited size of the F_2 population prevented an accurate estimation of the amount of crossing-over and further study should be made.

Based on actual frequencies for F_2 data, a X^2 value of 0.048 was calculated for Reka x f-g (table 12). A X^2 value for 3:3:1:1 ratio was calculated for the same cross and found to be 7.333 (table 7). Again, the size of the F_2 population seems to be the most likely explanation for the discrepancy.

Chromosome f makes up linkage group and chromosome 2 according to Burnham (1956) in which the factor pair Vv (non-six-rowed vs. six-rowed) has been identified and found to be linked with the "male sterile dwarf" ms_3 with 28.34± 1.42 per cent crossing-over, calculated from the average of 6 F_2 progenies study. The same factor pair Vv has been found by Honecker (1945) to be associated with the Pa pa factor (resistance vs. susceptibility to *Puccinia anomala*). In linkage group I the gene for earliness has been reported by Ramage and Suneson (1958) to be associated with chromosome f. It was found to be linked with the male sterile with 4.6±2.05 per cent cross-over units as reported by Kasha and Walker (1958). Day and Dickson (1957) reported the factor pair Gg (toothed vs. untoothed lemma) to be associated with chromosome f. The No no factor (High nitrogen vs. low nitrogen) was reported by Day and Dickson (1957) to be in linkage group I.

Data on linkage studies in barley have been summarized by Smith (1951), and all factor pairs located in the seven linkage groups of barley have been brought up to date by Robertson *et al* (1955).

CONCLUSIONS

1. The genetics of reaction to leaf rust race 4 was investigated in the varieties Sudan (C.I.6489) and Reka 1 (C.I.5051) by crossing them with nine translocation stocks.
2. Analysis of the classification of the reaction of the F_2 plants to leaf rust, race 4, with X^2 for 3:1 ratio and P values for each cross of the varieties Sudan and Reka with the translocation stocks has been calculated.
3. A summary of the classification of the reaction of the F_2 plants from crosses of Reka and Sudan with the translocation stocks to *Puccinia hordei*, race 4, and for partial sterility *vs*. fertility, with X^2 values for 3:3:1:1 and P values for goodness of fit to indicated ratio has been presented.
4. X^2 values and P values for deviation from the expected recombination ratios based on actual frequencies for F_2 data from each cross of the varieties Sudan and Reka with the translocation stocks have been calculated.
5. Results obtained from the F_2 data of the crosses studied indicated that reaction to leaf rust is conditioned by genes independent of each other in the two varieties.
6. The chromosomes that carry those genes have been identified. It was found that chromosome e carries the gene for resistance in Sudan while chromosome f carries the gene for resistance in Reka 1.

SUMMARY

It has been determined previously that resistance to leaf rust of barley (Puccinia hordei Otth.) race 4 is controlled by single gene pairs in the varieties Sudan and Reka and these genes assort independently of one another. To gain further information about these genes, reciprocal translocations were utilized as a means for identifying the chromosomes which carry them. This was done by analyzing the F_2 data from crosses between the resistant varieties and translocation stocks.

Nine translocation stocks were crossed with Sudan and Reka. Each of these stocks was homozygous for a reciprocal translocation between two of the seven chromosomes. Among the nine stocks each chromosome was involved in at least two translocations. The F_1 plants from crosses of Sudan with all translocations and Reka with all translocations were tested for rust reaction and found to be resistant. Examination of the mature pollen showed that the F_1's were partially sterile. Cytological examination of the partially sterile F_1 plants revealed a ring of four plus five pairs of chromosomes indicating heterozygosity for the translocations. Three F_2 segregation patterns were found. A ratio of 3 resistant partially sterile: 3 resistant fertile: 1 susceptible partially sterile: 1 susceptible fertile was found in those instances where the gene for resistance was independent of the chromosomes involved in the translocation. A ratio of 2 resistant partially sterile:

1 resistant fertile: 1 susceptible fertile was indicative of complete linkage. If linkage was incomplete, some susceptible, partially sterile plants were produced in lower frequency than one-eighth.

In the crosses involving the variety Sudan with translocation stocks having interchanges of chromosomes a with e and c with e, the F_2 data indicated complete linkage. The common chromosome e must carry the gene for resistance. This is substantiated by crosses involving translocations of a with other chromosomes and c with others which showed independent assortment.

In the crosses involving the variety Reka with translocation stocks having interchanges of chromosomes b with f and e with f, the F_2 data indicate that in Reka the gene for resistance is located on chromosome f. This is substantiated by crosses involving translocations of b with other chromosomes and e with others which showed independent assortment.

The results obtained from the crosses studied and from the F_2 data agree with previous conclusions that the factors for resistance in Sudan and Reka are independent.

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ABSTRACT

The objective of this investigation was to assign the genes for resistance to leaf rust Puccinia hordei Otth. race 4 in two varieties of barley to the correct chromosome pair by means of crossing them with nine translocation stocks.

The varieties Sudan (C.I.6489) and Reka 1 (C.I.5051) were crossed with the following translocation stocks: 5051-2 (a-b), 5059-5 (a-e), 5030-2 (b-d), 5034-2 (b-f), 5062-3 (b-g), 5038-2 (c-d), 5056-2 (c-e), 5053-6 (e-f), and 5057-1 (f-g). The F_1 plants from crosses of Sudan with all translocations and Reka with all translocations were tested for rust reaction and found to be resistant. Examination of the mature pollen showed that the F_1's were partially sterile. In the crosses involving the variety Sudan with translocation stocks having interchanges of chromosomes a with e and c with e, the F_2 data indicated complete linkage. The common chromosome e must carry the gene for resistance. This is substantiated by crosses involving translocations of a with other chromosomes and c with others, which showed independent assortment. In the crosses involving the variety Reka with translocation stocks having interchanges of chromosomes b with f and e with f, the F_2 data indicate that in Reka the gene for resistance is linked with chromosome f. This is substantiated by crosses involving translocations of b with other chromosomes and e with others, which showed independent assortment.

The results obtained from the F_2 data of the crosses studied indicated that in the varieties Sudan and Reka reaction to leaf rust is conditioned by genes that are independent of each other.