საქართველოს მეცნიერებათა ეროვნული აკადემიის მოამბე, ტ. 4, № 2, 2010
BULLETIN OF THE GEORGIAN NATIONAL ACADEMY OF SCIENCES, vol. 4, no. 2, 2010

Genetics and Selection

Revealing Genes of Hybrid Necrosis and Red Hybrid Chlorosis in Crosses of Varieties of Georgian Wheat and the Importance of These Genetic Phenomena for Selection and Theoretical Research

Petre Naskidashvili*, Maka Naskidashvili, Ia Naskidashvili**, Nikoloz Gakharia****

* *Academy Member, Georgian State Agrarian University, Tbilisi*
** *Georgian State Agrarian University, Tbilisi*

ABSTRACT. It has been shown that aboriginal cultivar-populations of Georgian soft wheat and selection cultivars carry in their genotype the dominant genes Ne_1 or Ne_2 responsible for hybrid necrosis. None of the tested varieties were free of these genes. Genotypes of all tetraploid wheat species and hexaploid species (*T. macha, T. zhukovsky* and *T. compactum*) contain the gene Ne_1. Genotypes of all Georgian cultivars of soft wheat carry the dominant gene responsible for red hybrid chlorosis Ch_2. Genotypes of all tetraploid and hexaploid wheats except the species *T. macha* are carriers of the same gene. Almost all varieties of *T. macha* and the variety of emmer wheat *T. dicoccoides* var. arabicum are carriers of a very rare gene Ch_1. The possibility of avoiding the negative effect of these complementary genes is proposed. The great importance of these genes from the viewpoint of phylogeny, theory and selection is outlined.

The work is dedicated to the 80th Anniversary of the discovery of genes underlying the phenomenon of hybrid necrosis by our prominent teacher, Corresponding Member of the Georgian Academy of Sciences, Professor Leonard Dekaprelevich. *© 2010 Bull. Georg. Natl. Acad. Sci.*

Key words: *hybrid necrosis, red hybrid chlorosis, hybrid dwarfness, genotype.*

Introduction. The phenomenon of underdevelopment or death of wheat plants, obtained as a result of remote and close crosses in the first and successive generations, was described for the first time by Leonard Dekaprelevich.

In his presentation made at the first All-Union Congress of Geneticists and Breeders L. Dekaprelevich (1929) proposed that early death or underdevelopment of plants of F1 generation depends on the interaction of two dominant genes. He also established the symptoms of manifestation of this genetic phenomenon in plant [1].

As a result of study of necrotic F_1 combination I.A. Kostyuchenko (1936) [2] has established segregation into necrotic and normal plants at a ratio 9:7. This confirmed the statement of L. Dekaprelevich that hybrid necrosis in wheat is based on the interaction of two dominant complepmentary genes. In 1963 this phenomenon was given the name "hybrid necrosis" by the Dutch scientist J. G. Hermsen [3].

Further study of manifestations of necrosis phenomenon was continued in the 1940s [4, 5]. Interest in the study of this phenomenon has especially increased from the 1960s in the Netherlands and Japan [6-15].

The Dutch scientist J. G. Hermsen made a serious contribution to the study of hybrid necrosis [3, 6-8]. On the basis of vast experimental material he substantiated the statements made by L. Dekaprelevich and I. Kostyuchenko and at the same time established that the degree of manifestation of hybrid necrosis depends on the number of alleles of each necrosis gene. He revealed eight degrees of necrosis genes Ne_1 and Ne_2. Despite this he singled out three groups of necrosis (strong, moderate and weak). Hermsen revealed another genetic system and called it red hybrid chlorosis (Ch_1+Ch_2) [3].

the Japanese scientist K. Tsunewaki [9-15] made great contribution to the study of the geographic localization of genes determining hybrid necrosis and red hybrid necrosis.

Tsunewaki and Hamada [14] have revealed a new type of hybrid chlorosis, different from red hybrid chlorosis and gave it the name "chlorosis of the second type". Chlorosis of this type is conditioned by interaction of two dominant complementary genes Chl_1 and Chl_2. Chlorosis of this type was revealed as a result of the cross of the cultivar Nokida of *T. dicoccum* wheat with the line Ng of the var. typicum of *T. timopheevi*.

A bit earlier Japanese scientist Nikishava [18] established that along with common and widely distributed genetic system of necrosis in wheat there exists the second genetic system of necrosis and denominated this system as Net_1-Net_2.

On the basis of the above-mentioned investigations it became known that four genetic systems are responsible for early death of hybrid plants: 1) two systems of necrosis Ne_1-Ne_2 and Net_1-Net_2; 2) two systems of chlorosis: red hybrid chlorosis Ch_1-Ch_2 and hybrid chlorosis of the second type Chl_1-Chl_2. After discovery of these systems a third system of chlorosis "white spotted chlorosis" was reported from Armenia [24]. To these four genetic systems "hybrid dwarfness" or "grass clump" should be added, which was first described by McMillan [17]. Later the Dutch scientist Hermsen [19] studied in detail the genetic nature of the latter system and divided it into three types according to the degree of manifestation. Phenomenon of "hybrid dwarfness" is based on interaction of two dominant complementary genes D_1 and D_2 and the latter genetic system is strengthened by the additional effect of D_3 gene [20-22]. In parallel to the evidence on the existence of two dominant genes there are data on the interaction of three genes [25].

Genes of chlosrosis and necrosis may be present in plants simultaneously and at the same time an epistatic effect of chlorosis on necrosis may take place. In this case chlorosis may suppress necrsosis [20-23].

In the former Soviet Union, in particular Georgia, investigations of chlorosis were started again from 1960 [20-22] and in Russia and Armenia from 1969 [24-30].

The degree of manifestation of chlorosis is influenced by the so-called effect of gene doses, or the number of Ne_1 and Ne_2 genes with different alleles (weak, moderate and strong) [20-22].

Material and methods. In order to obtain interspecific hybrids from aboriginal Georgian cultivar-populations 13 cultivars have been chosen for crosses (Ahaltsikhis Tsiteli Dolispuri, Dzalisura, Tsiteli Ipkli, Kartlis Tsiteli Dolispuri, all belonging to the var. ferrugineum; Kakhuri Dolispuri, Tianetis Tetri Dolispuri, Tetri Ipkli, Korboulis Dolispuri – var. aestivum, Lagodekhis Grdzeltavtava (long- spiked wheat), Khulugo, Gomborula – var. lutescens and 5 selection cultivars (Dolispuri 35-4, Dolispuri 18-46, Kakhi 8, Motsinave and Tbilisuri 5 – var. aestivum. The mentioned cultivars were crossed with tetraploid (*T. durum, T. turgidum, T. polonicum, T. georgicum, T, timopheevi, T. dicoccum, T. carthlicum, T. dicoccoides*) and hexaploid endemic species (*T. macha, T. zhukovskyi*), allooctaploid species (*T. fungicidum*) and autooctaploid species (*T. timonovum*). Crossing was performed using the free-limited method. Each species of soft wheat was crossed with 12 species and the total of 270 reciprocal combinations were obtained. Hybrids were obtained and grown in conditions of watering during the period of 1990-1992 and F_1-F_2 generations were studied during the period of 1991-1995. Hybrids were grown in conditions of high agrophone. Observations, records and processing of obtained experimental material were made according to accepted technique. Hybrids were obtained and studied on the experimental plot of the department of genetics and seed farming of Mukhrani study-research farm of the Georgian State Agrarian University (village of Mukhrani, Mtskheta district, East Georgia).

Experiment and discussion. 18 aboriginal and selection cultivars of Georgian soft wheat where crossed directly and reciprocally with a single variety of each tetraploid, hexaploid and octaploid wheat species. Three main varieties of *T. carthlicum* (var. stramineum, var, fuliginosum and var. rubiginosum) were used in crosses. Two varieties of hard wheat - var. coerulescens and var. reichenbachii – participated in crosses.

Genotypes of all tetraploid species used in crosses were carriers of the necrosis gene Ne_1. The gene Ne_1 contained in the genotpe of the variety of hard wheat var. coerulescens is especially strong. That's why this variety of hard wheat was used to test the presence of genes Ne_1 and Ne_2.

Study of 220 plants obtained as a result of crossing 5 cultivars of Kartlian ecotype of soft wheat with var. coerulescens of hard wheat has shown that in none of the plants the symptom of necrosis was manifested. The symptom of necrosis was not revelaed in crosses with var. reichenbachii of hard wheat and other tetraploid species.

The obtained results allow to conclude that genotypes of cultivars of Kartlian ecotype of soft wheat (Dolispuri 35-4, Dolispuri 18-46, Kartlis Tetri Dolispuri, Kartlis Tsiteli Dolispuri, Dzalisura) carry the gene Ne_1 of hybrid necrosis.

Completely different results were obtained in reciprocal combinations of crosses of cultivars of Kakhetian ecotype of soft wheat (Kakhuri Dolispuri, Lagodekhis Grdzeltavtava (Lagodekhian long spike wheat), Rachula, Kakhi 8, Gomborula) with tetraploid species (*T. durum, T. turgidum, T. polonicum, T. carthlicum, T. georgicum, T. timopheevii, T. dicoccum, T. dicoccoides*). In the generation F_1 both necrotic and normal plants were found. 313 plants were necrotic and 122 plants were normal. After earing necrotic plants developed small germinable grains. In F2 segregation into necrotic and normal plants took place. 418 plants were necrotic and 340 were normal - thus the ratio of necrotic plants to normal ones made 9:7. The theoreticaly anticipated ratio should have been 429:321. The obtained results have shown that genotype of cultivars of Kakhetian ecotype carry the necrosis gene Ne_2 with strong allele.

In the first generation of hybrids obtained as a result of crossing cultivars of soft wheat from Western Georgia (Tetri Ipkli, Tsiteli Ipkli, Khulugo, Korboulis Dolispuri) with tetraploid (*T. carthlicum, T. durum, T. turgidum, T. polonicum, T. georgicum, T, timopheevi, T. dicoccoides*) and with hexaploid wheat species (*T. zhukovskyi*, *T. macha, T. compactum*) part of plants manifested signs of hybrid necrosis. In plants of the second generation grown from seeds collected from these plants (F_1) segregation into necrotic and normal plants took place. The ratio of segregation was close to the theoretically expected ratio. Our results allowed to conclude that genotype of cultivars of soft wheat from Western Georgia carry the gene of necrosis N_2 with weak allele. Similar genotype is characteristic of the forest-mountain cultivar Akhaltsikhis Tsiteli Dolispuri.

Study of reciprocal hybrid combinations of F1 and F2 generations obtained as a result of crossing the oldest and selection cultivars of Georgian soft wheat with tetraploid and hexaploid species, except the combinations obtained from crosses with *T. macha* and wild emer wheat, did not reveal the phenomenon of red hybrid chlorosis. This made clear that cultivars of soft wheat participating in crosses and all genotypes of all cultivated tetraploid species carry in their genotypes the dominant gene of hybrid chlorosis Ch_2.

According to the Japanese scientists K. Tsunewaki and J. Hamada [14] var. imereticum of *T. macha* carries a very rare dominant gene Ch_1 of red hybrid chlorosis. Results of these researchers indicate that the var. imereticum of macha wheat carries in the genotype the genes $Ne_1ne_2Ch_1ch_2$. Due to this the species is a good object to test the presence of genes of red hybrid chlorosis. In the first generation of all combinations got from crosses with participation of this variety red hybrid chlorosis was revealed. Genotype of the var. imereticum of *T. macha* contains $Ne_1ne_2Ch_1ch_2$ [9].

Accordingly, the genotype of Kartlian ecotypes of Georgian soft wheat can be expressed as follows: $Ne_1ne_2Ch_2ch_1$, while the genotype of cultivars of Kakhetian ecotype can be expressed as $Ne_2ne_2Ch_2ch_1$. Similar results were obtained as a result of using in crosses the var. arabicum of wild emmer wheat *T. dicoccoides*.

Thus it has been established that var. arabicum of wild emmer wheat as well as var. imereticum of *T. macha* carry in their genotypes a very rare gene of hybrid chlorosis Ch_1. Due to this the var. arabicum can be used for testing the presence of genes of red hybrid chlorosis.

As a result of detection of complementary dominant genes responsible for red hybrid chlorosis in Georgian endemic wheat species and other varieties of wheat and also in aboriginal and selection cultivars of soft wheat the regularity of distribution of these genes in Georgia has been established. These results are reflected in the map illustrating distribution of genes of necrosis and chlorosis in Georgian wheats (Figure).

Thus thorough investigation of 270 reciprocal combinations of wheat has shown that 60 were completely viable, 90 half-viable, and 120 combinations turned out lethal. The cause of partial or complete loss of some interspecific hybrids was not phylogenetic remoteness of initial forms but joint action of two dominant complementary genes Ne_1 and Ne_2 (genetic system Ne_1+Ne_2). Sublethal combinations were obtained when the initial form carried weak alleles of chlorosis genes. Lethal combinations were got when one of the parents carried the necrosis gene with strong allele and another not less effective moderate allele. Despite the mentioned facts in 56 combinations a very rare genetic system (Ch_1+Ch_2) of red hybrid chlorosis was revealed, which also caused death or declined development of plants in the first and subsequent generations.

Conclusions:

Fig. Distribution of Chlorosis and Necrosis Genes in Georgian Wheat

- Genotypes of cultivar-populations and selection cultivars of Kartlian ecotypes (Eastern Georgia) of soft wheat mainly are the carriers of the dominant gene of hybrid necrosis Ne_1 and dominant gene of red hybrid chlorosis Ch_2 with strong allele.
- In Kakheti cultivars of soft wheat dominate, which carry in the genotype the dominant gene Ne_2 of necrosis with strong allele and the dominant complementary gene Ch_2 of red hybrid chlorosis with strong allele.
- Genotypes of cultivars of soft wheat from the mountainous zone of Georgia carry dominant comlementary gene of necrosis Ne_2 with moderate allele and the dominant complementary gene of red hybrid chlorosis Ch_2 with strong allele.
- Genotypes of cultivar-populations of soft wheat from Western Georgia carry the dominant comlementary gene of necrosis Ne_2 with weak allele and the dominant complementary gene of red hybrid chlorosis Ch_2 with strong allele.
- Genotypes of all tetraploid species (*T. carthlicum, T. georgicum, T. timopheevi, T. durum, T. turgidum, T. polonicum,T. dicoccum , T. dicoccoides*) are the carriers of dominant complementary gene of necrosis Ne_1 with strong allele, (hexaploid species *T. zhukowski* is carrier of the same gene) and the dominant complementary gene of red hybrid chlorosis Ch_2 (except *T. dicoccoides*) with strong allele.
- Varieties of *T. macha* (total of 14 varieties) carry in their genotype the dominant complementary gene of necrosis Ne1 with strong allele and the majority of these varieties and the genotype of var. arabicum of the wild emmer wheat contain a very rare dominant complementary gene of red hybrid chlorosis Ch_1 with strong allele.
- Presence of lethal genes in wheat genotype has phylogenetic importance and at the same time these genes are the genetic barrier of a species or cultivar.
- Negative effect of lethal genes can be avoided or mitigated by means of the following: cross of such sorts, whose genotypes are carriers of weak alleles of these genes. Breeding of hybrids in conditions of good agrotechnical conditions and pollination with mixed pollen give good results.
- In the case of crossing cultivars which carry strong alleles of necrosis genes gamma irradiation (Co^{60}) of seeds of F_1 generation can be assessed as a more or less effective influencing method [31].
- Cross of plants of F_1 generation with non homozygous plants can be considered as prospective. In such crosses intervarietal hybrid should be used as female parent. In F_1 of such combination the number of surviving plants increases significantly [32].

გენეტიკა და სელექცია

საქართველოს ხორბლის სახეობათა შეჯვარებისას ჰიბრიდული ნეკროზის, წითელი ჰიბრიდული ქლოროზის გამოვლენა და გენეტიკური მოვლენების მნიშვნელობა სელექციურ მუშაობაში და თეორიულ გამოკვლევებში

პ. ნასყიდაშვილი*, მ. ნასყიდაშვილი**, ი. ნასყიდაშვილი**, ნ. გახარია**

* *აკადემიის წევრი, საქართველოს სახელმწიფო აგრარული უნივერსიტეტი*
** *საქართველოს სახელმწიფო აგრარული უნივერსიტეტი*

საქართველოს რბილი ხორბლის აბორიგენული ჯიშ-პოპულაციები და სელექციური ჯიშები გენოტიპში ატარებენ ჰიბრიდული ნეკროზის განმაპირობებელ დომინანტურ გენს Ne_1 ან Ne_2. ამ გენებისგან თავისუფალი ჯიშები არ გამოვლენილა. ხორბლის ყველა ტეტრაპლოიდური სახეობის და ჰექსაპლოიდური სახეობების (*T. macha, T. zhukovsky* და *T. compactum*) გენოტიპშია გენი Ne_1. საქართველოს რბილი ხორბლის ყველა ჯიშის გენოტიპი წითელი ჰიბრიდული ქლოროზის განმაპირობებელი დომონანტური გენის Ne_2-ის მატარებელია. ამავე გენის მატარებელია ყველა კულტურული ტეტრაპლოიდური და ჰექსაპლოიდური (გარდა ხორბალ მახასი) სახეობის გენოტიპი. ძალიან იშვიათი გენის Ch_1-ის შემცველია ხორბალ მახას თითქმის ყველა სახესხვაობა და წყვილმარცვალას *T. dicoccoides* სახესხვაობა var. arabicum. გარდა ამისა, ნაშრომში ნაჩვენებია, რომ შესაძლებელია ამ კომპლემენტარული გენების უარყოფითი მოქმედების თავიდან აცილება. მითითებულია, რომ ამ გენებს გარდა უარყოფითი მოქმედებისა, დიდი მნიშვნელობა აქვთ ფილოგენეტიკური, თეორიული და სელექციური თვალსაზრისით.

შრომა ეძღვნება ჩვენი დიდი მასწავლებლის, საქართველოს მეცნიერებათა ეროვნული აკადემიის წევრ-კორესპონდენტის, პროფესორ ლეონარდე ლეონარდის ძე დეკაპრელევიჩის მიერ ჰიბრიდული ნეკროზის გენეტიკური მოვლენის განმაპირობებელი გენების აღმოჩენიდან 80 წლისთავს.

REFERENCES

1. *L.L. Dekaprelevich* (1930), In: Trudy Vsesoyuznogo syezda po genetike, selektsii, semenovodstvu i zhivotnovodstvu, **2**: 231, Leningrad (in Russian).
2. *I.A. Kostyuchenko* (1936), Zh. Sotsialisticheskoe rastenievodstvo, **19**: 128 (in Russian).
3. *J.G. Hermsen* 1963), In: Proceedings of the Second International Wheat Genetics Symposium. Lund: 439-452.
4. *R.M. Caldwell, L.E. Compton* (1943), J. Heredity, **34**, 3: 64-70.
5. *E.G. Heyne, G.A. Wiebe, R.H. Painter* (1943), J.Heredity, **34**, 8: 243.
6. *J.G. Hermsen* (1960), Euphytica, **9**, 1: 141.
7. *J.G. Hermsen* (1963), Euphytica, **12**, 1: 126-129.
8. *J.G. Hermsen* (1963), Genetics, **33**: 245-287.
9. *K. Tsunewaki* (1969), Wheat Information Service, 28: 1-4.
10. *K. Tsunewaki, J. Nakai* (1967), Canadian J. Genet. and Cytol., **9**, 1: 69.
11. *K. Tsunewaki, J. Nakai* (1967), Canadian J. Genet. and Cytol., **9**, 1: 75.
12. *K. Tsunewaki. J. Nakai* (1967), Canadian J. Genet. and Cytol., **9**, 1: 385.

13. *K. Tsunewaki, J. Hort* (1967), Japan. J. Genetics, **42**, 4: 245.
14. *K. Tsunewaki, J. Hamada* (1968), Japan. J. Genetics, **43**, 4: 279-288.
15. *K. Tsunewaki* (1971), DAN. Biologiya, **l24**, 9: 1251 (in Russian).
16. *A.C. Zeven* (1971), Euphytica, **20**: 239-254.
17. *J.R.A. McMillan* (1937), Counc. Sci. Ind. Res. Bull., **104**, 1: 69.
18. *K. Nischikawa* (1962), Gene analysis of type 2 necrosis, Siken, Ziho, 14: 45.
19. *J.D. G. Hermsen* (1967), Euphytica, **2**: 134.
20. *L.L. Dekaprelevich, P.P. Naskidashvili* (1971), Genetika, **7**, 3: 19 (in Russian).
21. *P.P. Naskidashvili* (1984), Mezhvidovaya gibridizatsiya pshenitsy. Monografiya. M., 256 p. (in Russian).
22. *P.P. Naskidashvili, M. Sikharulidze, E. Chernysh (*1983), Khorblis selektsia sakartveloshi [Wheat Selection in Georgia], Tbilisi, 340 p. (in Georgian).
23. *V.F. Dorofeev, A. F. Merezhko* (1969), Genetika, **5**, 4: 161 (in Russian).
24. *G.A. Babajanyan* (1970), Biologicheskii Zhurnal Armenii, **23**, 11: 69 (in Russian).
25. *A.F. Merezhko* (1970), Genetika, **6**, 4: 112 (in Russian).
26. *V.A. Pukhal'skii, A.V. Kozlenko* (1970), Sel'skokhozyaistvennaya biblioteka, **4**, 1: 44 (in Russian).
27. *V.A. Pukhal'skii, A.V. Kozlenko* (1970), Genetika, **6**, 9: 16 (in Russian).
28. *N. Sarkisyan, G. Babajanyan, A. Mkrtychyan* (1971), Biologicheskii Zhurnal Armenii, **24**, 8: 19 (in Russian).
29. *A. Mkrtychyan* (1971), Biologicheskii Zhurnal Armenii, **24**, 10: 44 (in Russian).
30. *V. M. Pilnev, I.A. Piranovskii* (1971), Scientific-Technical Bulletin of the All Union Institute of Genetics and Selection, **16**: 31 (in Russian).
31. *N.V. Turbin, T. S. Shilko* (1970), In: Otdalennaya gibridizatsiya. M.: 143-148 (in Russian).
32. *D. Sharma* (1966), Euphytica, **18**, 1: 66.

Received October, 2009