соответственно). Большая часть линий проявляли умеренную устойчивость (5–7 баллов). Для сорта Истра 1 характерна очень высокая степень устойчивости (9 баллов).

Вместе с тем в зависимости от комбинации скрещивания количество устойчивых к мучнистой росе линий несколько различалось. Так, среди линий F_4 комбинации скрещивания (Истра 1 / Одесская 16-*Vrn-B1a* // Истра 1) / Антоновка (всего 69 шт.) 66,6% характеризовались умеренной устойчивостью, 29% – относительной восприимчивостью к мучнистой росе и лишь 4,3% линий данной комбинации скрещивания были высокоустойчивыми (8 баллов) к возбудителю заболевания. При этом уровень устойчивости двух из трех родителей сорта Антоновка и почти изогенной линии Одесская 16-*Vrn-B1a* составлял по 6 баллов у каждого. В комбинации скрещивания (Истра 1 / Goudveld // Истра 1) / Антоновка подавляющее большинство из 44 линий F_4 поражались мучнистой росой в средней степени (балл 5–6), а 15 линий проявили сильную восприимчивость (балл 3–4). Устойчивость сорта Goudveld (донор гена *Vrn-B1a* ярового типа развития) составляла 5 баллов. В данной комбинации скрещивания не было выявлено устойчивых линий. В то же время в комбинации скрещивания (Истра 1 / Norin 29 // Истра 1) / Антоновка все без исключения линии проявляли устойчивость к мучнистой росе на уровне 8 баллов. Возможно, это обусловлено тем, что в отличие от других комбинаций скрещивания сорт Norin 29 (донор гена *Vrn-B1a* ярового типа развития) характеризовался устойчивостью к мучнистой росе (7,5 баллов) по сравнению с сортом Goudveld (5 баллов) и изогенной линией Одесская 16-*Vrn-B1a* (6 баллов).

Полученные результаты еще раз доказывают перспективность использования отдаленной гибридизации в качестве источника устойчивости к грибковым заболеванием и возможность создания исходного материала для селекции мягкой пшеницы.

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FREE PROLINE AND PROTEIN LEVELS IN CORN CELLULAR CULTIVARS GROWN UNDER OSMOTIC STRESS PRESSURE

L.E. Sergeeva, V.M. Kurchii, S.I. Mykhalska, L.I. Bronnikova
Institute of Plant Physiology and Genetics, National Academy of Scienses of Ukraine
e-mail: Zlenko_lora@ukr.net

The problem of plant osmotic stress tolerance is one of the most complicated. Wild type plants developed various protective mechanisms to mitigate detrimental effects of salt and water stresses. But the necessity of cultural plants with higher levels of stress tolerance becomes critical. Therefore, the investigation of tolerant genotypes (natural or experimentally obtained) makes a significant contribution to the detection of new tolerance determinants.

It is known, that free proline accumulation occurs in plant tissues during various osmotic stresses. This event is considered to be a common biological response to a wide range of biotic and abiotic stresses. Plants accumulate proline to mitigate stress detrimental effects. Proline lends increased viability to suffered plants. This amino acid is a cellular compatible osmolyte that protects enzymes against inactivation. It may serve as an energy supply for utilization during period of reparation. The activation of proline synthesis and inhibition of its oxidation retain proline level. There are two pathways of proline synthesis. The first one developed with the participation of Δ-pyrroline-5-carboxylate synthase (P5CS); the alternative pathway of proline biosynthesis is catalyzed by Orn-δ-aminotransferase (OAT). The proline degradation is the reverse process and catalyzed by Proline dehydrogenase (PrDH).

The enzyme Proline dehydrogenase oxidizes proline on mitochondrial membranes under normal conditions. Proline accumulation during osmotic stress occures due to increased synthesis and reduced degradation. Therefore the permanent inhibition of the proline oxidation (proline

accumulation) both at normal and stress conditions demonstrates certain changes of enzyme or its gene.

We obtained corn plants *via* Agrobacterium-mediated *in planta* transformation with LBA 4404 strain harboring pBi2E with double-stranded RNA-suppressor of the PrDH gene. Cellular cultivars were initiated from T1 and wild type corn plants. In our opinion it is better to study stress changes on cellular level. In this instance we ignore proline transport among mature plant organs (the first). We can estimate direct stress influence on cells that divide and grow (the second).

Corn cell cultivars were examined under salinity (2.5% sea water salts) or water stress (0.8M mannitol). Osmotic stresses were prolonged to 42 days. Such experimental conditions were used to simulate devastating stress attack on the environment. Free proline and protein levels were estimated in calli of those corn genotypes.

Under normal conditions free proline levels were not considerable in both transformed (T-) and control (C-) calli. At the same time proline levels of T-calli exceeded those parameters of C-calli more than 3 times. Such event was a result of the RNA-suppressor integration into plant genome. The protein levels were: T-calli – 6.1 ± 0.6 mg/g f. w.; C-calli – 2.6 ± 0.6 mg/g f. w. So those data reflect the normal versatile functions of any variant. Tested corn genotypes realized all stages of the cell development.

Hard osmotic stresses changed cells existence. On the 42-nd day of stress pressure we noted essential differences. Free proline levels increased in all tested cultivars. Proline contents of T-calli rose under salinity at 1.4 times, under mannitol pressure at 3.4 times; proline levels in ordinary cells (C-calli) increased under salinity at 2.6 times, under water stress at 9.3 times.

J. Liu and J.-K. Zhu (1997), tested sos1 mutant of *Arabidopsis thaliana* that was more than 20 times more sensitive to NaCl than wild type Arabidopsis. This mutant plant accumulated more proline than wild type. The lack of correlation between proline level and salt-stress tolerance in certain plant species has led to the conclusion that proline accumulation is merely a consequence of stress.

In our experiment we used lethal for cell cultures doses of salt and mannitol. Protein biosynthesis is an adequate marker of plant viability. It was strongly decreased in corn cultivars during osmotic stress. But protein levels, measured in T-calli, exceeded parameters of C-calli more than 1.4 times (sea water salts) and 1.3 times (mannitol). To our opinion, that protein biosynthesis restriction in T-calli was the reflection of metabolism special limitations (stress adaptations) not stress injuries.

Effects of genetic manipulation can be specific. The event of proline permanent level increase calls direct attention. There is a debate whether proline or its degradation product pyrroline-5-carboxylate (P5C) is the cause of toxicity (Mani et al. 2002). In our case the Agrobacterium-mediated transformation did not alter cell cycle of growth and development. Cell cultivars, obtained from transformed corn plants maintained optimal proline levels. In those cultivars the balance between proline biosynthesis and degradation was developed.

Corn cell cultivars, initiated from T1 plants challenged severe osmotic stresses. Their stress-tolerance was correlated with higher free proline levels.