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**POSTOVOITOVA A. S., RABOKON A. M., BILONozhko YU. O., KALAFAT L. O., PIRKO YA. V.,  
BLUME YA. B.**

**Institute of Food Biotechnology and Genomics of the NAS of Ukraine, Osipovskoho st., 2a, Kyiv, 04123,  
Ukraine**

e-mail nastya.postovoytova@gmail.com, тел. +38(044)434-37-77

## **DEVELOPMENT OF *TRITICUM DICOCCUM* INTRON LENGTH POLYMORPHIC (ILP) MARKERS FOR USING IN CEREAL BREEDING**

Emmer wheat (*Triticum dicoccum* (Schuebl.) Schrank) is one of the wheat species that belongs to one of the largest higher plant families – the family *Poaceae*. This tetraploid wheat is formed by hybridization of two diploid wild grasses, *Triticum urartu* and yet unidentified *Aegilops*. *Tr. dicoccum* has a number of useful characteristics, including unpretentiousness to soils, drought and cold resistance, rust resistance etc. Due to this, this type of cereal is used in wheat breeding and is an interesting object for the creation of new improved breeding lines and varieties using modern molecular genetic and breeding approaches.

Today the vast majority of known DNA markers have been tested on various types of cereals, including IRAP, REMAP (Campbell et al., 2011; Carvalho et al., 2010), SSR (Song et al., 2005), ISSR (Khaled et al., 2015) and others. Undoubtedly, one of the effective approaches is Intron Length Polymorphism (ILP), in particular the TBP method (Rabokon et al., 2015). In view of this, the development of novel DNA markers that are polymorphic and more cross-transferable is prerequisite for evaluating the existing diversity of cereal varieties, molecular breeding, systematic maintenance and authenticity testing of commercially important varieties and hybrids.

Therefore, the aim of this work was to develop ILP markers for *Tr. dicoccum* for their further use in the selection of this species and other cereals.

EST sequences *Tr. dicoccum* were downloaded from the EST database available at the National Center for Biotechnology Information (NCBI) (<https://www.ncbi.nlm.nih.gov/>). Due to the probable possibility of having the same

EST sequences in the database, in order to eliminate their redundancy, the EST was assembled using the online tool EGassembler (Masoudi-Nejad et al., 2006). All unique EST sequences *Tr. dicoccum* were analyzed in the PIP database (<http://ibi.zju.edu.cn/pgl/pip/>) (Yang et al. 2007) and the probable positions of the introns were identified by comparison with the homologous gene sequences of *Oryza sativa*. In addition, ILP markers for *Tr. dicoccum* were developed using the PIP database. Namely, the selection of primers to exon regions was carried out in order to further amplify introns and evaluate their polymorphism.

Today 105 EST sequences *Tr. dicoccum* are available in the EST database. In order to exclude the EST sequence redundancy, the analysis was performed using the online tool EGassembler and 90 unique sequences (78 singletons and 12 contigs) were established. Using the PIP database in 90 EST *Tr. dicoccum* found 165 intron positions and developed 79 ILP markers. Each pair of ILP primers was named Trd\_##, where ## was the ILP marker number.

As a result of bioinformatics analysis, 79 ILP markers were developed, which will be used for differentiation of *Tr. dicoccum* lines/varieties. In addition, it is planned to evaluate the possibility of their use for genotyping of closely related cereal species, both cultivated and wild, as well as interspecific hybrids, as high cross-species transferability rate will increase our understanding of intra- and interspecies gene flow, genetic structure, and evolutionary relationships among cultivated and wild relatives of *Poaceae*.

**Keywords:** *Intron Length Polymorphism, ILP marker, Triticum dicoccum, genotyping.*