

MOLECULAR MARKERS EFFICIENCY FOR ASSESSMENT OF GENETIC STRUCTURE IN BARLEY ACCESSIONS

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Barley is the fourth most important grain crop in the world which is well-adapted to marginal and stress-prone environments, and hence a more reliable plant than wheat or rice in regions located in colder or higher altitude. Genetic diversity is a key component in breeding programs. We have analyzed the genetic diversity of 106 barley accessions using RAPD and ISSR molecular marker systems. The accessions were composed of wild and domesticated barley representing genotypes from fifteen different countries. A total of 40 polymorphic primers (20 RAPD and 20 ISSR) were used. The RAPD primers generated 331 amplification products and the ISSR primers produced 295 DNA fragments, out of which 225 and 247 were polymorphic, respectively. By RAPD markers the polymorphic banding patterns with the number of amplified fragments varied from 10 (AF-14 and OPL-09) to 26 (OPA-04). The percentage of polymorphism ranged from 53.84 % (BY-15) to 100 % (OFG-14, OPF-03 and W-07) with an average of 77.47 %. Contrary to this, the number of bands amplified per primer by ISSR markers varied between 10 (UBC-872, ISSR-4, 6 and 7) and 21 (UBC-845) and the percentage of polymorphism ranged from 57.14 % (UBC-814) to 100 % (ISSR-5 and 7) with 83.97 % polymorphism. Dendrogram was constructed based on RAPD and ISSR polymorphism dividing the barley accessions into eleven however by combined RAPD+ISSR all samples were grouped into ten main distinct clusters. High correlation between the RAPD and ISSR marker systems was shown using a Mantel test ($r = 0.852$). The data obtained confirmed and indicated that Azerbaijan cultivated barley accessions distinctly diverged from wild types.

Key words: wild and cultivated barley, genetic diversity, polymorphism, molecular markers

ЕФЕКТИВНІСТЬ МОЛЕКУЛЯРНИХ МАРКЕРІВ
ДЛЯ ОЦІНКИ ГЕНЕТИЧНОЇ СТРУКТУРИ
ІЗОЛЯТИВ ЯЧМЕНЮ

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Ячмінь — четверта за важливістю зернова культура у світі, яка добре адаптована до маргінального середовища з можливими факторами стресу, а отже надійніша рослина, ніж пшениця чи рис, для регіонів, розташованих у холодніших чи вищих висотах. Генетичне різноманіття є ключовим компонентом селекційних програм. Ми проаналізували генетичне різноманіття 106 ізолятів ячменю за допомогою систем молекулярних маркерів RAPD та ISSR. Було використано ізоляти як диких, так і акліматизованих рослин, які представили генотипи з п'ятнадцяти різних країн. Загалом було використано 40 поліморфних праймерів (20 RAPD та 20 ISSR). Праймери RAPD згенерували 331 продукт ампліфікації, а праймери ISSR створили 295 фрагментів ДНК, із яких 225 і 247 були поліморфними, відповідно. За маркерами RAPD схеми поліморфних смуг з кількістю ампліфікованих фрагментів були у діапазоні від 10 (AF-14 та OPL-09) до 26 (OPA-04). Відсоток поліморфізму був від 53,84 % (BY-15) до 100 % (OFG-14, OPF-03 та W-07) при середньому значенні в 77,47 %. На відміну від цього, кількість смуг, ампліфікованих на один праймер за допомогою маркерів ISSR, була у діапазоні від 10 (UBC-872, ISSR-4, 6 та 7) до 21 (UBC-845), а відсоток поліморфізму складав від 57,14 % (UBC-814) до 100 % (ISSR-5 та 7) при поліморфізмі на рівні 83,97 %. Було створено дендрограму на основі поліморфізму RAPD та ISSR, яка поділила ізоляти ячменю на одинадцять груп, однак за поєднанням RAPD+ISSR всі зразки було згруповано у десять основних чітких кластерів. Було показано високий рівень кореляції між системами маркерів RAPD та ISSR за допомогою тесту Мантела ($r = 0,852$). Отримані дані підтвердили і продемонстрували, що ізоляти ячменю, вирощені в Азербайджані, чітко відрізняються від дикого ячменю.

Ключові слова: дикий і акліматизований ячмінь, генетичне різноманіття, поліморфізм, молекулярні маркери.

REFERENCES

- Abdellaoui R, Kadri K, Ben Naceur M, Ben Kaab LB (2010) Genetic diversity in some Tunisian barley landraces based on RAPD markers. *Pak J Bot* 42(6):3775–3782.
- Akar T, Avci M, Dusunceli F (2004) Post-harvest operations. Food and Agriculture Organization of the United Nations.
- Aliyoun-Nazari S, Zamani Z, Khadivi-Khub A, Fatahi R (2014) Molecular characterization of wild accessions of Marmareh (*Prunus incana* Pall.) revealed by simple and inter-simple sequence repeats. *Europ J Hort Sci* 79(2):70–75.

- Bertini CH, Schuster I, Sedyama T, Barros EG, Moreira MA (2006) Characterization and genetic diversity analysis of cotton cultivar using microsatellite. *Genet Mol Biol* 29:321–329.
- Bhagyaawant SS, Gupta N, Gautam A, Chaturvedi SK, Shrivastava N (2015) Molecular diversity assessment in chickpea through RAPD and ISSR markers. *World J Agric Res* 3(6):192–197.
- Botstein D, White RL, Skolnick M, Davis RW (1980) Construction of a genetic linkage map in man using restriction fragment length polymorphisms. *Am J Hum Genet* 32:314–331.
- Catanã R, Mitoi M, Ion R (2013) The RAPD techniques used to assess the genetic diversity in *Draba dorneri*, a critically endangered plant species. *Adv Biosci Biotechnol* 4:164–169.
- Cheghamirza K, Zarei L, Zebarjadi AR, Honarmand SJ (2017) A study of the association between ISSR and RAPD markers and some agronomic traits in barley using a multiple regression analysis. *BioTechnologia. J Biotechnol Computat Biol Bionanotechnol* 98(1).
- Drine S, Guasmi F, ALI SB, Triki T, Boussorra F, Ferchichi A (2016) Genetic diversity analysis of different barley (*Hordeum vulgare* L.) genotypes from arid and humid regions using ISSR and RAPD markers. *J New Sci* 34.
- El Sayed AA, Ezzat SM, Mostafa SH, Zedan SZ, Abdel-Sattar E, El Tanbouly N (2018) Inter simple sequence repeat analysis of genetic diversity and relationship in four Egyptian flaxseed genotypes. *Pharmacognosy Res* 10(2):166–172.
- El-Awady MA, EL-Tarras AA, El-Assal S (2012) Genetic Diversity of Some Saudi Barley (*Hordeum vulgare* L.) Landraces Based on Two Types of Molecular Markers. *Amer J Appl Sci* 9(5):752–758.
- Eshghi R, Abrahimpour F, Ojaghi J, Salayeva S, Baraty M, Rahimi M (2012) Evaluation of genetic variability in naked barley (*Hordeum vulgare* L.). *Inter J Agric Crop Sci* 4(16):1166–1179.
- Eshghi R, Akhundova E (2010) Genetic diversity in hullless barley based on agromorphological traits and RAPD markers and comparison with storage protein analysis. *Afr J Agric Res* 5(1):97–107.
- FAO, 2018. <http://www.fao.org/faostat/en/#data/QC>
- Farsani TM, Etemadi N, Sayed-Tabatabaei EB, Talebi M (2012) Assessment of Genetic Diversity of Bermudagrass (*Cynodon dactylon*) Using ISSR Markers. *Int J Mol Sci* 13:383–392.
- Fernandez M, Figueiras A, Benito C (2002) The use of ISSR and RAPD markers for detecting DNA polymorphism, genotype identification and genetic diversity among barley cultivars with known origin. *Theor Appl Genet* 104(5):845–851.
- Gebremedhin W, Firew M, Tesfye B (2014) Stability analysis of food barley genotypes in northern Ethiopia. *Afr Crop Sci J* 22(2):145–153.
- Gonzalez A, Coulson M, Brettell R (2000) Development of DNA markers (ISSRs) in mango. *Acta Hort* 575:139–143.
- Gorji AM, Poczai P, Polgar Z et al (2011) Efficiency of arbitrarily amplified dominant markers (SCoT, ISSR and RAPD) for diagnostic fingerprinting in tetraploid potato. *Am J Potato Res* 88(3):226–237.
- Grewal S, Kharb P, Malik R, Jain S, Jain RK (2007) Assessment of genetic diversity among some Indian wheat cultivars using random amplified polymorphic DNA (RAPD) markers. *Indian J Biotechnol* 6:18–23.
- Guasmi F, Elfalleh W, Hannachi H et al (2012) The use of ISSR and RAPD markers for genetic diversity among south Tunisian barley. *Inter Scholarly Res Not*.
- Guasmi, F, Elfalleh, W, Hannachi H, Fiores K, Touil L, Marzougui N, Triki T, Ferchichi A (2012) The Use of ISSR and RAPD Markers for Genetic Diversity among South Tunisian Barley. *Inter Scholarly Res Network Agronom* (1):1–10.
- Guliyev N, Sharifova S, Ojaghi J, Abbasov M, Akparov Z (2018) Genetic diversity among melon (*Cucumis melo* L.) accessions revealed by morphological traits and ISSR markers. *Turk J Agric For*. 42:393–401.
- Hammer O, Harper DAT, Ryan PD (2001) PAST: paleontological statistics software package for education and data analysis. *Palaeontol Electron*. 4(9).
- Hou YC, Yan ZH, Wei YM, Zheng YL (2005) Genetic diversity in barley from west China based on RAPD and ISSR analysis. *Barley Genet Newsletter* 35:9–22.
- Izzatullayeva V, Akparov Z, Babayeva S, Ojaghi J, Abbasov M (2014) Efficiency of using RAPD and ISSR markers in evaluation of genetic diversity in sugar beet. *Turkish J Biol* 38:429–438.
- Kanbar A, Kondo K (2011) Efficiency of ISSR and RAPD Dominant Markers in Assessing Genetic Diversity among Japanese and Syrian Cultivars of Barley (*H. vulgare* L.). *Res J Agric Biol Sci* 7(1):4–10.
- Keilwagen J, Kilian B, Iizkan H, Babben S, Perovic D, Mayer KFX et al (2014) Separating the wheat from the chaff – a strategy to utilize plant genetic resources from *ex situ* gene banks. *Scientific Rep* 4:5231.
- Khan MK, Pandey A, Thomas G, Akkaya MS, Kayis SA, Ozsensoy Y, Hamurcu M, Gezgin S, Topal A, Hakki EE (2015) Genetic diversity and population structure of wheat in India and Turkey. *AoB PLANTS*. 7:1–14.
- Khatab IA, El-Mouhamady AA, Mariey SA, Elewa TA (2019) Assessment of water deficiency tolerance indices and their relation with ISSR markers in barley (*Hordeum vulgare* L.). *Cu Sci Inte* 8:83–100.

- Koyuncu M, Ardiç O et al (2015) Identification of Genetic Divergence in Some Bread Wheat Varieties by RAPD and ISSR Analyses. *SDU J Faculty Agriculture/SDÜ Ziraat Fakültesi Dergisi* 10(2).
- Liu K, Muse SV (2005) PowerMarker: an integrated analysis environment for genetic marker analysis. *Bioinformatics* 21:2128–2129.
- Mahar KS, Rana TS, Ranade SA, Meena B (2011) Genetic variability and population structure in *Sapindus emarginatus* Vahl from India. *Gene* 485(1):32–39.
- Majeed Zargar S, Farhat S, Mahajan R, Bhakhri A, Sharma A (2016) Unraveling the efficiency of RAPD and SSR markers in diversity analysis and population structure estimation in common bean. *Saudi J Biol Sci* 23(1):139–149.
- Mantel N (1967) The detection of disease clustering and a generalized regression approach. *Cancer Res* 27:209–220.
- Mareiy SA, Farid MA, Karima AR (2018) Morphological and molecular characterization of some Egyptian barley cultivars under calcareous soil conditions. *Middle East J* 7(2):408–420.
- Nevo E (2006) Genome evolution of wild cereal diversity and prospects for crop improvement. *Plant Genet Res* 4:36–46.
- Ojaghi J, Akhundova E (2010) Genetic diversity in doubled haploids wheat based on morphological traits, gliadin protein patterns and RAPD markers. *Afr J Agric Res* 5:1701–1712.
- Ojaghi J, Akhundova E (2010) Genetic diversity in doubled haploids wheat based on morphological traits, gliadin protein patterns and RAPD markers. *African J Agric Res* 5:1701–1712.
- Rahimi M, Hervan I, Valizadeh V, Kajori F, Ebrahimipour F (2014) Genetic Diversity among wild and cultivated barley by ISSR Marker. *Bull Env Pharmacol Life Sci* 3(10):57–62.
- Sadigova S, Sadigov H, Eshghi R, Salayeva S, Ojaghi J (2014) Application of RAPD and ISSR markers to analyse molecular relationships in Azerbaijan wheat accessions (*Triticum aestivum* L.). *Bulgar J Agric Sci* 20(1):97–105.
- Salayeva S, Ojaghi J, Pashayeva A, Izzatullayeva V, Akhundova E, Akperov Z (2016) Genetic diversity of *Vitis vinifera* L. in Azerbaijan. *Rus J Genet* 52(4):391–397.
- Santos LF, Oliveira E, Silva AS, Carvalho FM, Costa JL, Padua JG (2011) ISSR Markers as a Tool for the Assessment of Genetic Diversity in Passiflora. *Biochem Genet* 49(7–8):540–545.
- Sharma R, Sharma S, Kumara S (2018) Pair-wise combinations of RAPD primers for diversity analysis with reference to protein and single primer RAPD in soybean. *Ann Agrar Sci* 16:243–249.
- Singh A, Dikshit HK, Jain N, Singh D, Yadav RN (2014) Efficiency of SSR, ISSR and RAPD markers in molecular characterization of mungbean and other Vigna species. *Indian J Biotechnol* 13:81–88.
- Systat, 2009. SYSTAT 13. Systat Software. San Jose CA.
- Tiwari JK, Rastogi NK, Chandrakar PK, Verulkar SB (2013) Fingerprinting of rice (*Oryza sativa*) cultivars using inter simple sequence repeat markers. *Curr Adv in Agric Sci* 5(2):250–253.
- Verma KS, Haq S, Kachhwaha S, Kothari SL (2017) RAPD and ISSR marker assessment of genetic diversity in *Citrullus colocynthis* (L.) Schrad: a unique source of germplasm highly adapted to drought and high-temperature stress. *Biotech* 7(288):1–24.

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