DEVELOPMENT OF EST-SSR MARKERS BASED ON TRANSCRIPTOME SEQUENCING IN KODO MILLET (*PASPALUM SCROBICULATUM* L.)

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Kodo millet (Paspalum scrobiculatum L.) (2n = 4x = 40) mostly found in dry and semi-arid parts of Africa and Asia, also known as miraculous millets, is a staple food and a nutrient-security crop. It's commonly known that kodo millet has the strongest tolerance to drought. Little study was done on high throughout genomic studies in relation to the creation of millets improved by genomics. In the present investigation, 21,080,571 total reads of sequence data were included in the 6.55 gigabytes of raw sequence that was produced following the genome sequencing run. After utilizing FastQC to verify the raw data quality, a total sequence of 1,087,611 and 57,81,23,572 bp was obtained. Three RNA samples from mature, blooming, and vegetative specimens were gathered, and the high-throughput Ion torrent sequencing method was applied to the sequencing process. In the vegetative stage, there were 1, 34, 40, and 824 raw readings; in the reproductive and mature stages, there were 1, 66, 35, 843 reads and 97, 43, 629 bp reads, respectively. At three distinct stages, the average read length was 131.6 bp, 137.6 bp, and 148.8 bp, in that order. The interface modules BatchPrimer3 (version 1.0) were used to construct the SSR and EST-SSR primers. Four FASTA files including contigs from the transcriptome and genome were utilized to identify SSRs. A total of thirty SSRs were found, with GC% and Tm ranges of 40–70 and 58–62 °C, respectively. Of the 30 SSR and EST-SSR primers that were used for validation, only 17 were able to amplify the DNA from 27 distinct types of Kodo millet. 24 bands were obtained from the PCR amplification of 27 distinct types. We think that these findings may be applied in the future as possible candidates to enhance this significant staple crop.

Key words: *Kodo millet, Genome, Transcriptome, RT-PCR, cDNA library, EST-SSR.*

РОЗРОБКА EST-SSR МАРКЕРІВ НА ОСНОВІ ТРАНСКРИПТОМНОГО СЕКВЕНУВАННЯ ПРОСА КОДО (*PASPALUM SCROBICULATUM L.*)

Просо кодо (*Paspalum scrobiculatum* L.) (2n = 4x = = 40) переважно росте в сухих і напіваридних регіонах Африки та Азії. Це продовольча культура, багата на поживні речовини, яку також називають чудодійним просом. Загальновідомо, що просо кодо має найвищу стійкість до посухи. Було проведено небагато високопродуктивних геномних досліджень, пов'язаних зі створенням проса, поліпшеного за допомогою геноміки. У цьому дослідженні 21 080 571 загальних зчитувань даних послідовності було включено в 6,55 гігабайт необробленої послідовності, яка була отримана після секвенування геному. Після використання FastQC для верифікації якості необроблених даних було отримано загальну послідовність 1 087 611 і 57 81 23 572 пн. Було зібрано три зразки РНК зі зрілих, квітучих і вегетативних зразків, а для процесу секвенування було застосовано високопродуктивний метод секвенування Ion torrent. На вегетативній стадії було 1, 34, 40 та 824 необроблених зчитування; на репродуктивній та зрілій стадіях було відповідно 1, 66, 35, 843 зчитування та 97, 43, 629 пн зчитування. На трьох різних стадіях середня довжина зчитування становила 131,6 пн, 137,6 пн та 148,8 пн відповідно. Для побудови праймерів SSR та EST-SSR використовували інтерфейсні модулі BatchPrimer3 (версія 1.0). Для ідентифікації SSR було використано чотири файли FASTA, що містили контиги з транскриптому та геному. Всього було виявлено тридцять SSR з діапазонами GC% та Tm, відповідно 40–70 та 58–62 °C. З 30 праймерів SSR та EST-SSR, які були використані для валідації, лише 17 змогли ампліфікувати ДНК з 27 різних типів проса кодо. В результаті ПЛР-ампліфікації 27 різних типів було отримано 24 смуги. Ми вважаємо, що ці результати можуть бути застосовані в майбутньому, як можливі кандидати для поліпшення цієї важливої продовольчої культури.

Ключові слова: просо кодо, геном, транскриптом, ЗТ-ПЛР, бібліотека кДНК, EST-SSR.

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