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Lev Elkonin, Adv Crop Sci Tech 2022, Volume 10

Improvement of nutritional value of sorghum grain using site-directed mutagenesis of kafirin genes

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Sorghum (Sorghum bicolor (L.) Moench) is one of the most important crops of world agricultural production, a reliable source of fodder and food grains in the arid regions. However, unlike other cereals, sorghum grain has a lower nutritional value, the main reason for which is the resistance of its storage proteins (kafirins) to protease digestion. Modern biotechnology has an arsenal of methods for solving this problem, in particular, RNA interference and genome editing technologies. Using RNA interference technology, we previously obtained mutants with improved digestibility of kafirins and increased lysine content in two varieties of grain sorghum. Currently, to create sorghum lines with improved digestibility of kafirins we are using an approach based on genome editing technology. For this purpose, we have created a series of binary vectors for site-directed mutagenesis of the k1C5 and gKAF1 genes encoding 22 kDa α- and γ-kafirins, respectively.

Nucleotide sequences encoding the signal polypeptides of these proteins responsible for their packaging into the protein bodies of endosperm cells were chosen as targets. In total, four vectors were created: p1C and p2C – for editing k1C5 gene, and p3C and p4C – for editing the gKAF1 gene; each of these vectors contained the cas9 endonuclease gene and genomic target motifs (23 bp sequences). Using Agrobacterium-mediated genetic transformation these vectors were introduced into genome of the grain sorghum variety Avans. In total, 22 transgenic plants carrying cas9 gene were obtained. Among the regenerants obtained from experiments with the p2C, the plants were identified in which the kernels had a modified endosperm texture with a disturbed development of the vitreous endosperm, as well as the plants with a reduced grain size. SDS-electrophoresis of kernel proteins revealed a number of mutants with a significantly higher level of kafirin digestibility (up to 87%) compared to the original cv. Avans (60%). DNA sequencing of the k1C5 gene sequence, encoding the signal polypeptide of 22 kDa α-kafirin, in one of the mutants with improved digestibility of kafirins from T1 generation revealed the presence of a deletion of the third nucleotide of the target, which may have led to a frameshift and a change in the amino acid composition of the polypeptide. Among the plants obtained from experiment with the p3C, two mutants with a deletion and a substitution in the nucleotide sequence of the gKAF1 gene, encoding the γ-kafirin signal polypeptide, were identified. Thus, based on the genome editing technology, we obtained the mutants with significantly improved digestibility of kafirins, which can be used in sorghum breeding.

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Biography

Dr. Lev Elkonin is professor at department of biotechnology, federal center of agriculture research of the south-east region, saratov, 410010, Russia

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Macdex Mutema, Adv Crop Sci Tech 2022, Volume 10

Irri drop report: Framework for quantifying and reporting canal water losses

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Canal use for conveying water is as old as irrigation development. Canals remain in use at many irrigation schemes today because they transport bulk water cheaply. However, canals are associated with high water losses through, mainly, seepage, leakage, and evaporation. While the water losses are known to exist, their quantification is a daunting task due to several challenges. Firstly, canal water flow is turbulent and unsteady. Secondly, the canal water losses vary with microclimates, environmental conditions, and physical characteristics of the canal network. Thirdly, separating seepage and leakage in the field cannot be practically done, which makes modeling to understand their character and dynamics as individuals impossible. Yet, it is important to quantify the different canal water loss types at different spatial-temporal scales and to understand their main drivers with a view to generating strategies for corrective measures. As a first step, a literature study was performed to elucidate the effects of canal water flow and soil characteristics on water losses. Data 1388 canal sections from 45 peer reviewed papers published across the world were used, with the results showing that evaporation is generally negligibly smaller than both seepage and leakage losses. The results confirmed that water losses decline with increasing soil clay content regardless of lining material. Canal shape, wetted perimeter and wetted area also showed significant effects on water losses. In addition, methods for assessing the canal water losses had effects on the quantitative values of the losses. The physical mechanisms of the dominant water loss types together with dominant driving factors need to be considered in future field research. The impact of physical features such as embankment height and freeboard depth on water losses also need consideration. The study results, informing on future field research on canal water losses, should excite water scientists, engineers and practitioners.

Biography

Macdex Mutema is a Senior Research Engineer at the Agricultural Research Council-Natural Resources and Engineering, South Africa, in the Irrigation and Agricultural Infrastructural Engineering division. He has expertise in assessing the condition and performance of irrigation infrastructure for smallholder and large-scale irrigation systems. The Irri Drop Report is an attempt to develop a framework for capturing and reporting on the major water loss types from irrigation canals in South Africa. The framework aims to eventually evolve into a model that report on the spatial and temporal variability of the different major water loss types for lined and unlined canals, and also for different lining materials and canal shapes. The proposed framework borrows from the Water Use Efficiency Accounting Report (WUEAR) developed for the Water Administration System (Water Admin, https://wateradmin.co.za) developed by NB Systems, who are partners in development of the Irri Drop Report.

Neha Sharma, Adv Crop Sci Tech 2022, Volume 10

Serological Identification, In silico comparison and Validation of Partial Coat Protein (CP) Gene of Zucchini Yellow Mosaic Virus (ZYMV) infecting summer squash in India

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Zucchini yellow mosaic virus (ZYMV) belonging to Potyvirus family is responsible for causing major crop losses in summer squash in India. By Direct Antigen Coating Enzyme-linked immunosorbent assay (DAC ELISA) the presence of potyvirus was confirmed and ZYMV in particular was authenticated by Double antibody Sandwich ELISA (DAS ELISA). RNA isolation of test virus cDNA showed a 700 bp amplification by using specific primers and the PCR product was outsourced for sequencing and finally a sequence of154 bp was obtained with 50.65 % G+C content and 49.35 % A+T content. The test sequence proved 91 % alignment with D13914 (ZYMV isolate from USA) on BLASTN analysis. Expasy tools were further used for translating the test sequence into protein sequence and it showed 75.9% similarity on BLASTP analysis. To further understand evolutionary status, on phylogenetic tree construction with other already submitted sequences at National Center for Biotechnology Information (NCBI) it clearly proved a close relation of the Indian isolate of ZYMV with ZYMV isolate of USA. 22 different restriction sites were found in In silico restriction digestion in the test ZYMV sequence via using NEB cutter. On secondary structure prediction dominance of alpha helix was reported. The present study aimed to enhance the knowledge about ZYMV and hence prevent crop losses by controlling the disease. The study would also help plant virologists and pathological researchers to work on coat protein-mediated virus resistance against ZYMV.

Biography

Dr. Neha Sharma is currently working as a assistant professor, biotechnology, IILM College of Engineering and Technology, Greater Noida, India

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