CRISPR/CAS9 MEDIATED GENOME EDITING AND ITS POTENTIAL APPLICATIONS IN GENETIC IMPROVEMENT OF HEVEA BRASILIENSIS

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Biotechnology is now entering a new era, equipped with tools, which are quick and more precise than the conventional methods of genetic engineering. Genome editing with the help of engineered nucleases is the latest technology, where precise manipulation of specific genomic sequences is possible by knocking out undesirable genes or modifying genes to gain new functions. This technique was developed based on a naturally occurring bacterial immune mechanism and relies up on sequence specific endonucleases which are capable of generating DNA double strand breaks at specific locations within the genome. With the help of error-prone natural endogenous DNA repairing mechanism in the cell, site-specific mutations can be introduced. The nucleases can be programmed theoretically in such a way that precise editing of any gene in an organism would be possible to gain desirable phenotypes. Among the various genome editing platforms, CRISPR/Cas9 mediated genome editing is the most popular technology owing to its simplicity and versatility. Since variations generated through these techniques are precise and more similar to natural variations and are more acceptable than conventional GMOs where the possibility of retaining undesirable gene sequences and unforeseen ill effects are relatively high. This paper aims to give an overview to this new technology which holdsmajor implications in different areas of life science. The concept, history, mechanism, applications and thelimitations of this technology are discussed in detail. The potential application of the technology in genetic improvement of Hevea brasiliensis and its implications are also discussed.

Key words: CRISPR/Cas9, Gene editing, Guide RNA, Gene knockout, DNA free editing, Hevea

INTRODUCTION

Creation, identification and utilization of genetic variation is the base of any crop improvement programme. Twentieth century witnessed the green revolution mainly due to development of high yielding varieties by conventional breeding techniques producing hybrids that responded to application of inorganic fertilizers. Conventional breeding strategies

are not sufficient enough to meet the demand of increasing global population in the 21st century even as the extent of arable land is declining and several external factors such as climate change, land degradation *etc.* adversely affect productivity. In conventional breeding, breeder has absolutely no control over the process of recombination of genes during sexual reproduction except for the selection of