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Poster Abstract – E.02

EXCISION OF A SELECTABLE MARKER IN TRANSGENIC WHEAT (*TRITICUM AESTIVUM*) USING A CHEMICALLY REGULATED Cre/loxP SYSTEM

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Selectable marker genes are required to ensure the selection of transgenic plants during plant transformation. However, once transformation is accomplished, the presence of a marker gene in transgenic plants becomes undesirable, particularly as most of the selectable marker genes currently used confer resistance to antibiotics and herbicides.

The removal of selectable marker genes will not only lead to the elimination of potential environmental and health-related risks as well as technical barriers, but also increase the consumer acceptance of GM crops and their products.

Several strategies have been employed to remove selectable markers from transgenic plants.

Recently, chemical-inducible Cre/loxP DNA recombination systems (CLX) have emerged and seem to provide a highly reliable method to generate marker-free transgenic *Arabidopsis* and rice plants in a single step transformation.

Wheat is a recalcitrant species and, in comparison with rice and maize, progress in this species has been slower. A robust method for the transformation of wheat has only recently been developed. Furthermore, in wheat, selection can be carried out only after the regeneration phase, otherwise no plants will regenerate. For this reason, it is not possible to obtain transgenic marker-free wheat plants by single step transformation - chemical induction needs to be carried out on the T1 generation after T0 selection.

We have transformed wheat plants with pX6-GFP, which contains the CLX system. Transgenic plants were grown in order to obtain seeds and the immature embryos were then collected and subcultured on callus induction medium for 10 days. The calli were then transferred onto fresh medium containing β-estradiol for 20 days.

Excision of the marker gene was monitored through the detection of GFP fluorescence.

Fluorescent calli were transferred onto regeneration medium in order to obtain marker-free plants.

These plants will be molecularly analysed in order to verify the excision and the efficiency of this chemically-inducible system.