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Biotechnological investigations on the natural rubber producer *Taraxacum koksaghyz* concerning the herbicide target AHAS

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The Russian dandelion *Taraxacum* koksaahyz (Tks) is one of the most promising candidates as an alternative for the rubber tree Hevea brasiliensis. This crop is currently the sole commercial source for natural rubber, saving the worldwide needs for producing tires and other goods which cannot be replaced by synthetic analogs. Presently, the global population of this tree is threatened by the fungus Microcyclus ulei that leads to dieback of trees. Tks is excellently appropriate to build up new resources concerning the physical and chemical characteristics of the rubber. Until now the cultivation and harvest of Tks is elaborative and the yield is not yet comparable to the one H. brasiliensis. Further biotechnological and agricultural optimization is needed to achieve a crop that can substantially contribute to the world rubber market. Tks grows under sparse conditions in the region of the Tian Shan Mountains and is therefore very eligible for being grown on marginal acreages in the European climate zone. As Tks has a very slow early growth phase, weed is overgrowing the young plant resulting in reduced size and rubber yield. In addition to other projects focusing on improvement of field performance by e.g. breeding or fertilization, our part of the "EVITA" project aims to develop an imidazolinone resistance for a sustainable weed control during Tks seed production. By screening the gene sequence of the herbicide (acetohydroxyacid synthase, AHAS) of wild type and EMS-mutagenized plants which are imidazolinone tolerant, we get information about the natural sequence variability and induced mutations. So far we could not find herbicide resistance endowing SNPs - suggesting that the investigated AHAS1 gene might not play the prominent role in this herbicide resistance mechanism. By using degenerated primer as well as the RACE method we are searching for other AHAS genes in the Tks genome which are relevant for imidazolinone resistance. Further we want to use the recently developed but well known CRISPR/Cas9 nuclease system for introducing specific mutations in a targeted manner to provoke imidazolinone resistance. As a proof of concept this will be done first with the already known AHAS1 gene.

As this work is generating a lot of sequencing data, it is necessary to have an easy, fast and reliable tool for their analysis. In addition to manual evaluation with the CLC Main Workbench, usage of the web-based workflow management system Galaxy proved to be efficient for detecting SNPs in hundreds of reads. By defining workflows with parameters and error rates, analysis is simplified and less afflicted to subjective errors.