

Establishing optimal parameters for gene editing in sweetpotato through protoplast isolation and culture

Sweetpotato is a highly nutritious root rich in starch, and while it very much looks similar to the common potato on the surface, they are only distantly related. The nutritional value however, as well as the appetizing taste of the sweetpotato has in recent years made it an attractive candidate crop for increasingly widespread cultivation in the Scandinavian peninsula. This is supported by local media, reporting an increase in consumption of sweetpotato exceeding 1600% between 2010 and 2018, with comparable numbers originating from Norway. Despite this, the vast majority of sweetpotato on the market today are imported, and not for the lack of farmers trying to cultivate these delicacies locally.

Unfortunately, this root vegetable is ill equipped to survive sustained cultivation in the relatively low temperatures, or the long summer days of the local cultivation season, instead preferring a climate that by Scandinavian standards could be described as sweltering. As the tubers are originally native to west Africa, this is not very surprising at all, but on the other hand this hasn't stopped both individuals as well as larger government sponsored institutions from attempting to adapt the sweetpotato to a colder climate.

With this in mind a genetically modified sweetpotato with regards to higher chilling stress tolerance and adaptation to longer days could prove a promising solution to this otherwise diverse and complicated dilemma. However, while extensive research has been conducted to establish techniques and protocols to allow for this in recent years, barely any have focused specifically on sweetpotato, instead placing more emphasis on the common potato.

And so the first step to locally cultivated sweet potato will by necessity be establishing how to best introduce new and useful genes. For this purpose using protoplasts is arguably the most attractive option. But what are protoplasts? And why is working with them the most convenient alternative?

Well, put simply, protoplasts are individual plant cells that lack a cell wall. This makes them quite vulnerable, but also far easier to alter genetically as there is one less layer that protects their DNA from outside influences. You might think of it as how it would be quite tricky for a mechanic to work on a car's engine without first opening the hood. The same principle applies here, and while there are other techniques that indeed do allow for gene editing without first removing the cell wall, it seems like the most obvious first step. Not only does it make future work exceedingly simple, but it's also a quicker and far cheaper alternative.

With this in mind, an extensive literary review of existing protocols, using a wide selection of parts such as leaves and stem tissue from sweetpotato, but also closely related species was

conducted, and the data analyzed through multiple advanced statistical models. This has in turn allowed us to investigate possible and even probable relationships between different treatments and their success rate at removing the cell wall through enzymatic digestion, and then achieving this without also killing the cells.

Armed with this theoretical knowledge, our task has then been to validate these findings in practical experiments and ultimately establish optimal parameters for gene editing in sweetpotato. And it is our hope that this project will lay the foundation for a future where sweetpotato could be made to be a hardier crop, not only adapted for colder climates as found in Sweden, but worldwide.