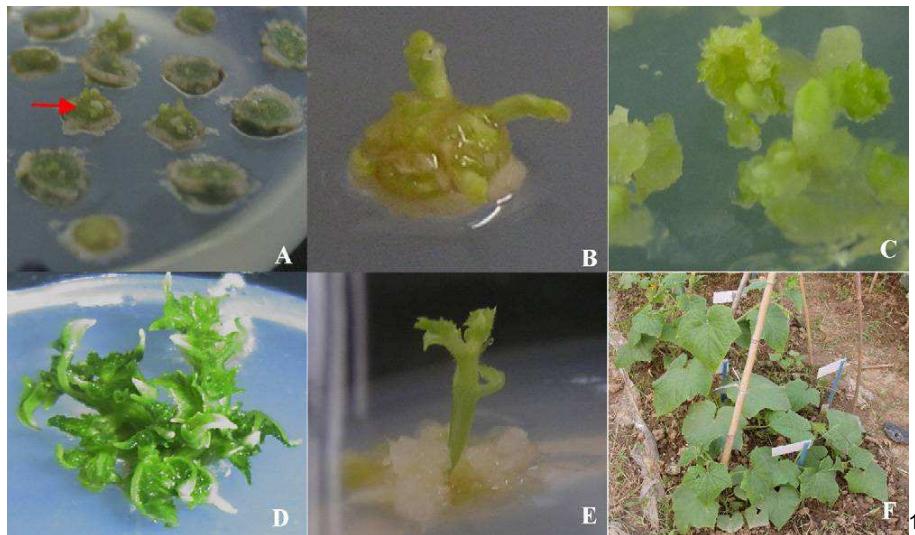


## Study on In vitro production of haploid vegetables



Syngenta is one of the world's leading companies with more than 25,000 employees. "Bringing plant potential to life" is the purpose of the company. Syngenta has two main business lines, crop protection and seeds production.

Innovation on plant breeding is helping to improve the quality of the crops and obtain new varieties. Producing double haploids is a relatively new technology used for the company especially producing double haploids vegetable crops. Inducing haploids is used for the development of commercial vegetable hybrids. The importance of pure lines not only presents in increasing efficiency of selecting certain features also marker development. Ovary culture , anther culture have been main approaches which used in commercially producing homozygous plants and genetic studies.

Working with pure lines is important for the breeding in order to gain strength in the market for the company syngenta, and important in marker assisted breeding. It takes about ten years to get parental lines needed for hybrid breeding in the way of traditional inbreeding. However haploid induction through in vitro gynogenesis in cucumbers makes it much more efficient of getting pure lines. DH populations are indispensable for marker development. Furthermore, DH line is important for developing multi-genetic traits, such as one genotype with combined of more than one or two resistance genes on disease.

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<sup>1</sup> \*The picture used from Diao et al., 2009: Embryogenesis and regeneration of plantlets of cucumber.

The company started to produce DH cucumbers 2 years ago, but not successful of low efficiency of shoots obtained. Cucumber (*Cucumis sativus* var. *sativus* L) is a member of family Cucurbitaceae. The project focus on testing effect of different combinations of hormones on explants shoots obtained, and effect of different concentrations of Jasmonic acid. Testing Jasmonic acid is because it has been found that in low concentrations (0.01 – 1 µMol) jasmonic acid can have a hormonal function and stimulates cell division and micro calli development. (Toro *et al.*, 2002). The experiments on cucumbers used ovary culture, cutting un-fertilized cucumbers in small sections then put them in induction medium. The result showed unexpected shoots formation from all hormone combinations, only few combinations gave shoots but with low efficiency. The result showed the Miller medium is better to be used as basic medium compared with MS medium. The results found that Jasmonic acid seems have effects on the shoots formation in the lower concentration (0.1µmol and 0.01µmol), but more research have to be done later to make a certain. For further experiments it is necessary to search for other/or extra stress-treatments, in order to trigger the right response of the ovules. JA can be tested on a larger scale and not only in the induction medium but also upon early transfer of cucumber explants 1-2 weeks after initiation of the cultures.

Unlike cucumber, the DH production protocol of *Brassica oleracea* has already been established. However the production of DH on some *Brassica* genotype is not efficient. Here, jasmonic acid may be an improvement for recalcitrant genotypes. The research tested the effect of different concentration of jasmonic acid on two genotypes of *Brassica*, white cabbage and cauliflower. Anther culture was used to produce double haploid *Brassica*. By comparison to the results rising from different JA concentrations, 1 µmol JA gave best shoots/anther frequency during the embryogenesis in the genotype of white cabbage, 0.05 µmol JA gave highest score of shoots/anther in the genotype of cauliflower. The shoot formation from embryos was counted after 1<sup>st</sup> subculture, showing the highest shoot formation frequency from embryo presents in the 0.01µmol JA in white cabbage and 0.05 µmol in cauliflower, respectively. Jasmonic acid may have effect on embryo formation of shoots formation; however more tests have to be done later. The further research using wider JA concentration range and more genotypes is needed to reveal JA's role on DH production.