CYTOPLASMIC MALE STERILE AMARANTHUS PROGRESS REPORT 2020. David M. Brenner

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Male sterility is useful for plant breeders to cross-pollinate between plants, especially plants with many small flowers. Cytoplasmic male sterility (CMS) is a way of managing male sterility to get populations of 100% male sterile plants to use as females in plant crossing. In many crops especially sunflowers, and sorghum, most of the cultivars are F_1 hybrids made with CMS to benefit from hybrid vigor (heterosis) and thereby improve performance over open pollinated cultivars (Kim and Zhang, 2018). CMS may someday be used to produce improved seeds for amaranths as is already done with those crops (Peters and Jain, 1987; Brenner, 2020). We in the United States National Plant Germplasm System are assembling useful germplasm and information for crop improvement including CMS amaranths (GRIN, 2020a).

There is progress to report since the 2019 (Brenner) publication on male sterility which describes a publicly available *Amaranthus* accession with CMS (PI 686465), and accessions that both maintain male sterility (PI 568179) and restore male fertility (PI 538323) in crosses with that CMS accession. Unfortunately, the above CMS and maintainer accessions are agronomically poor and need to be replaced in a new CMS system by better grain producing parents. A series of crosses was initiated in 2019 to classify additional accessions as restorers or maintainers by their performance as males in crosses with the CMS accession. The results of these crosses are posted online in the GRIN database, with eleven maintainers and five restorers to date (GRIN, 2020b). The tentative interpretation is that accessions of *A. caudatus* and *A. cruentus* maintain CMS and accessions of *A. hypochondriacus* restore male fertility. There are potentially many accessions that either restore or maintain. This is like the situation in sorghum where many accessions are classified as restorers or maintainers by their crossing outcomes posted in GRIN (2020c).

There was an unexpected problem with seedlings, death after germination. Crosses of the CMS accession (PI 686465) with two maintaining males (PI 599338 and PI 608791) resulted in normal in CMS populations and were then crossed with the male fertility restorer (PI 538323) to confirm male fertility restoration. But instead of the healthy fertility restored populations, these two populations have an unusual lethal condition in 100% of the observed seedlings. The seeds germinate well with healthy roots and cotyledons, but the hypocotyls are short and weak, and the apex does not grow beyond the cotyledons. The seedlings die within three weeks after germinating in greenhouse soil. Two other maintainer males (PI 689685 and PI 642741) resulted in healthy populations after similar crosses including the same restorer, and a third (PI 538255) had a mixture of viable and not viable seedlings. The lethal trait could be a result of small parent populations with rare abnormalities, or genes that are more common. After observing these lethal-trait seedlings, other lethal-trait seedlings were observed at a low frequency among the CMS accession (PI 686465) seedlings, which may be the source of this lethal trait.

In the fall of 2020, an effort was started to backcross the CMS cytoplasm into seven diverse advanced backgrounds of accessions that are expected to maintain CMS (PI 538255, PI 538320, PI 628779, PI 636182, PI 642741, PI 647848, PI 674253). The intention is to create CMS versions of successful amaranth types. Eventually these backcross populations can be crossed with diverse accessions that restore male fertility and be tested for beneficial heterosis.

Plant breeders can receive seeds of 'PI' accessions via the GRIN online database (GRIN, 2020d). Use the "New User" click-on to start a germplasm request. It is also possible for plant breeders to develop additional male sterile populations from their own sources. Some of our male sterile populations are from an *A. hypochondriacus* cultivar, 'Plainsman'. Seed lots of Plainsman were searched for rare off-type translucent and dark seed-coat seeds, which are probably from a heritage of crossing with weeds. In 1992 a sample of these off-type seeds were planted in rows and were found to segregate for male sterility (Brenner, 1993). The same seed-selection method may work to find male sterility in other *Amaranthus* cultivars.

Conclusion

The grain amaranths are suited to CMS hybrid seed production. More progress is needed before F_1 hybrids can be produced from adapted parents and be ready for yield testing in field conditions.

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