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22. © Propagation protocol for ram's head lady's slipper, *Cypripedium arietinum*.
Steele, W. K. Native Plants Journal 8(1):58-64. 2007.

William K Steele

ABSTRACT

Adaptation of tissue culture methods yields vigorous ram's head lady's slipper (*Cypripedium arietinum* Ait. f. [Orchidaceae]) seedlings ready for removal from flasks after 11 to 12 mo. Following vernalization, these seedlings grow into robust green plants that typically bloom 2 to 3 y after outplanting.

Steele WK. 2007. Propagation protocol for ram's head lady's slipper (*Cypripedium arietinum*). Native Plants Journal 8(1):58–64.

KEY WORDS

micropropagation, seed propagation, tissue culture, Orchidaceae

NOMENCLATURE

USDA NRCS (2006)

EDITOR'S NOTE

Although NPJ shows metric and English units of measure in manuscripts, the amounts of chemicals used in micropropagation work are so small that trying to measure them in English units becomes unrealistic. Conversions are provided at the end.

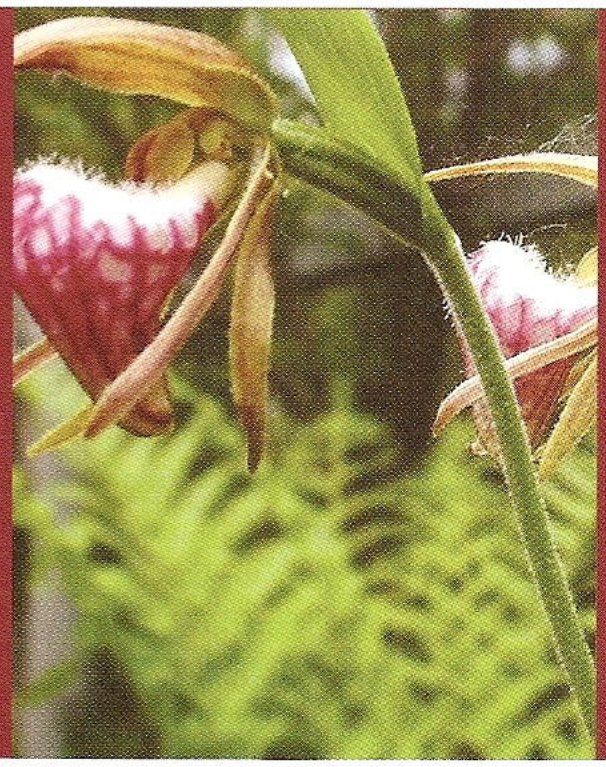
Figure 1. Flowering *C. arietinum* plants grown from seeds.

Photos by William K Steele

PROPAGATION PROTOCOL FOR

Ram's Head Lady's Slipper

CYPRIPEDIUM ARIETINUM



The ram's head lady's slipper (*Cypripedium arietinum* Ait. f. [Orchidaceae]), a rare terrestrial orchid, inhabits cool forests of the northeastern US from Maine to Minnesota and southeastern Canada from Nova Scotia to Saskatchewan. The species name derives from the fancied resemblance of the flower to the head of a goat (Figure 1). Every state and Canadian province in which the plant occurs lists it as Special Concern, and most of these jurisdictions consider it Threatened or Endangered, that is, S2 or S1 in rankings as used by The Nature Conservancy (Brzeskiewicz 2000). The vulnerability of this plant obliges conservation efforts including artificial propagation.

In nature, *Cypripedium* orchids reproduce by means of minute seeds that contain little in the way of food reserves. Germination of such seeds in the soil requires an external source of energy and mineral nutrients, and this source comes in the form of soil fungi. In the ground, fungal hyphae invade the seed, but the orchid embryo has the ability to digest these filaments to obtain sugars and other nutrients for growth.

The commonly used "asymbiotic" laboratory procedures for germination of orchid seeds use no fungus and instead supply all the nutrients the infant orchid requires in a liquid solution or gel medium. Different species of orchids, even within the same genus, generally require media of different composition, and an individual young plant commonly requires a somewhat different balance of nutrients at different stages of its life. One of the major considerations in artificial orchid propagation is to find the optimal medium composition for a given species both for germination and for continued growth of the young seedlings.

At Spangle Creek Labs, we propagate mostly native *Cypripedium* species but also some from Eurasia, for commercial and conservation purposes. Minnesota Department of Natural Resources regulations that protect Threatened species within the state do not permit us to sell *C. arietinum* seedlings or mature plants.

MICROPROPAGATION

Laboratory propagation of *Cypripedium* species employs the basic methods of plant tissue culture, but unlike true tissue culture, which uses explants cut from meristem regions of existing plants, we start with seeds. The use of seeds instead of explants has two main advantages for our purposes: seeds require less