PURPOSE: The Buoy-Deployed Seeding System (BuDS) (Figure 1) was developed to take advantage of the natural ability of mature reproductive shoots of eelgrass (Zostera marina) to release seeds over a period of weeks, and offers a low-cost alternative method of planting eelgrass using seed that can replace or compliment broadcast seeding efforts (Pickerell et al. 2005). This system may offer several advantages over current seeding methods including: 1) it may more closely mimic the natural process of seed release with regard to timing and distribution; 2) seed dispersal is targeted to preferred habitats where seedling development can be easily monitored, 3) equipment is inexpensive and can be reused, 4) the infrastructure (i.e., storage facilities and staff) for reproductive shoot and seed storage is not required; and 5) it can employ nontechnical personnel, allowing for community-based participation in restoration activities. Although this system was initially developed for use with eelgrass, it is being adapted for use in the restoration of other species of submerged aquatic vegetation (SAV) within the Chesapeake Bay region.

BACKGROUND: SAV meadows are known to be crucial habitats for numerous species of fish and shellfish of ecological and commercial importance. In addition to habitat support, SAV meadows perform other important ecological functions such as shoreline stabilization and water quality improvement. Given the continuing decline in SAV abundance over the last several decades, there has been an increased interest in development of cost-effective restoration techniques for SAV. The most commonly utilized restoration method for eelgrass has been the use of whole shoots removed from donor beds and relocated to transplant sites (Fonseca et al. 1998). Despite the relative success of this method, transplantation of adult shoots is extremely labor-intensive and costly and is not likely to be practical for large-scale restoration efforts. Potential impacts to donor sites associated with the removal and transplantation of wild plant stock are also a concern. Mechanization of the planting process has the potential to increase efficiency and lower costs, but development of this equipment is still in its infancy, and has met with limited success in some areas (e.g., Fishman et al. (2004)). A potential cost-effective alternative to the use of whole plants is the harvest and dispersal of seeds for use in SAV restoration (Ailstock and Shafer 2004). Early work by Addy (1947a, 1947b); Lamounette (1977); and Churchill and Riner (1978) first identified the potential of seeds for use in the restoration of...
eelgrass beds, and this approach has recently become the subject of renewed interest. Detailed methodologies for harvesting, processing, and dispersing seeds have already been developed (Orth et al. 1994; Ifuku and Hayashi 1998; Harwell and Orth 1999; Granger et al. 1996, 2002; Orth et al. 2003; Ailstock and Shafer 2004). To date, the most successful large-scale eelgrass seeding efforts have taken place in the Chesapeake Bay region involving the use of broadcast seeding (Orth 2003). Although there are obvious logistical and genetic advantages to using seeds, one drawback of the broadcast method is that large amounts of mature reproductive shoots must be transported to a specialized holding facility and held for up to 5 months until they can be planted in the fall. Seed handling and storage facilities are simple in principle, but require a number of large tanks equipped with flowing seawater and the infrastructure to support them. In addition to these requirements, there are also labor and energy costs associated with the operation and maintenance of these facilities. Therefore, this may not be a viable option for smaller, community-based restoration programs.

The BuDS system was developed as an alternative to broadcast seeding that mimics the ability of detached, floating reproductive shoots to transport viable seeds over great distances from the original eelgrass meadow, which was suggested by McRoy (1968) and demonstrated by Harwell and Orth (2002). Seeds in the floating reproductive shoots can continue developing after pollination until they mature and are released. Reproductive shoots continue to release mature seeds for a period of approximately 4 weeks (Churchill and Riner 1978, DeCock 1980, Harwell and Orth 2002). Unlike natural methods of seed dispersal by wind and currents where seeds are often distributed to areas incapable of supporting seedling establishment, the use of deployment buoys restricts dispersal to preferred habitats thereby maximizing reproductive success beyond that likely to be achieved in nature.

**EELGRASS SEED DEVELOPMENT AND FLOWER HARVEST:** Following pollination, several stages of seed development occur. These include: a) style erection, b) back bending of style, c) flowering of the half anthers, d) seed development, e) seed release (Figure 2), and f) withering (DeCock 1980). A detailed explanation of the flowering process is provided by DeCock (1980). After release, most seeds sink relatively quickly to the sea floor (Orth et al. 1994). A portion of the seed rain can be transported by gas bubbles formed during release from the ovary to a distance greater than 200 m (Tutin 1938, Churchill et al. 1985), but studies show that most seeds are deposited adjacent to the parent plant (Ruckelshaus 1996). Because seeds continue to mature in detached reproductive shoots, drifting shoots can disperse batches of seeds over great distances (Harwell and Orth 2002). The BuDS system was developed to incorporate natural aspects of seed dispersal which include wind and tide-induced water movement in the process of dispersal. As a result, the timing and movement of seeds is a random process that takes place over a 3- to 4-week period.

The methods employed for hand harvest of eelgrass flowering shoots have been well described by other authors (e.g., Granger et al. (2002)) and will not be covered in detail. Mature reproductive shoots are easily distinguished from the surrounding leaf canopy given their
different color, texture, size, and epiphytic fouling. Once the differences between vegetative and reproductive shoots are recognized, the process of collection is fairly simple. Although some areas may allow for hand collection while wading during low tide, the most efficient means of collection is usually during high tide using SCUBA. Under these conditions, the flowers are best identified from the surrounding background (Figure 3) and there is no need to wade through the meadow possibly damaging shoots.

Timing of flower collection is critical to the success of any seed-based restoration effort. The authors recommend collecting flowering shoots during the first or second week of the four weeks of seed release (Figure 4). Collection prior to the signs of initial seed release or after more than 10 percent of the seeds have been released will reduce potential seed yield (i.e., a larger proportion of immature seeds or fewer seeds because seed release has occurred). Determining the natural timing of flowering at the donor site is best understood by conducting regular monitoring of flower development. A review of the stages of flower development described by DeCock (1980) is important to understanding the process. Weekly observations of flower/seed development are usually sufficient during the early stages of development, but once stage IV (i.e., recognizable seeds) is reached, monitoring every two to three days may be necessary to effectively target collection. Collection should begin when seed release is first observed and no later than when 10 percent of the seeds have been lost. Any number of different mesh bags could be used to hold reproductive shoots during the collection process; the authors have found 1-mm mesh shellfish spat bags to be effective.

Once the collection window has been established for a given location, it can be used as a general guide for future collections at that site. However, since the maturation of seeds is related to water temperature, yearly temperature variations can cause the season to change by up to two weeks earlier or later than any previous year. For this reason, it is useful to track water temperatures at the collection site so that they can be compared to temperatures in subsequent years when collections are planned. Regardless of preparations made from one year to the next, it is always necessary to closely monitor the donor site when the seeds have reached stage IV.
BuDS LINE ASSEMBLY AND DEPLOYMENT:

Materials List: (as shown in Figure 5)

- Line - ¼-in. (6.4-mm) floating polypropylene line
- Cement block - ½ or full depending on exposure
- Buoys – 11-in. (28-cm) buoys or similar
- Pearl nets – square (36- x 36-cm); 6- or 9-mm mesh size
- Used garden hose
- Wire ties (≥25-lb capacity)

The following section describes the assembly of a single BuDS unit using the materials listed above.

Cut garden hose into 24-in. (61-cm) sections. Cut the line into 17-ft (5-m) lengths using a soldering iron or rope cutter. If using scissors or a knife, be sure to melt the ends of the line using a soldering iron or flame to prevent fraying.

**Buoy Attachment.** At one end of the line measure down 3 ft (0.9 m) and bend the line back on itself creating a bight. Tie a half hitch in the end of the bight, creating an eye (closed loop) with a 2-to 3-in. (5- to 7-cm) opening. Make sure to pull the knot tight. Slide a buoy, flat end first, up from the bottom of the line to the base of the knot. Wrap the loose end of the line around the buoy to the pointed end of the buoy and tie the two lines together using a bowline knot. Secure the bare end of the line that results from the knot by tucking it into the lay of the line running along the side of the buoy. This is done by slightly separating the strands in the line and pushing the loose end through the space (Figure 6).

**Anchor Attachment.** The garden hose is used to prevent fraying and breakage of the line where it is attached to the concrete block anchor. Slide the free end of the line (the end not attached to the buoy) through one hose section so that 12 in. (30.5 cm) hangs out of one side. Then, slide this hose section through the center hole of a cement block. Pull the ends of the hose together above the block and tie the bare end to the main line using a bowline knot. Tie a half hitch in the bare end around the line running through the hose and tuck the remaining line into the open end of the hose closest to the base of the knot (Figure 7).
Stocking. Stocking rates are site-specific depending on flower size, seeds per flower, and availability of reproductive shoots. Depending on the purpose of the deployment it may or may not be necessary to standardize the stocking rate, but the authors recommend standardizing stocking such that roughly the same number of seeds are present in each net and an approximate seeding rate can be determined per unit area. After collection, transfer the reproductive shoots from the mesh collection bags to large fish totes filled with seawater on board the collection vessel or onshore depending on the type of deployment. Then pack the reproductive shoots loosely into containers of known volume (e.g., ½ gallon (1.9L) plastic pot) (Figure 8) and transfer to the pearl nets (Figure 9). The authors commonly stock between 100 and 125 reproductive shoots per net, resulting in a potential seed yield ranging from 3,000 to 5,000 seeds. After stocking, sew the net closed with the polypropylene thread and secure with a single half hitch. Suspend filled nets from the collection vessel or set in shallow water near the shore for a deployment from shore by passing a line through the loops on the nets.

Transport and Deployment. When underway by vessel or vehicle, stocked nets are stacked on edge in fish totes or similar containers filled partly with cool seawater and covered with damp burlap (Figure 10). Prior to attaching the nets, assembled lines can be transported in stacks on the vessel or vehicle. Take care that the line and buoys do not become entangled. This is best accomplished by wrapping the line around the block and setting the buoy to one side. Upon arriving at the planting site, nets are individually attached to the eye at the end of each buoy line using the wire ties (Figure 11) and set in place. Buoy spacing is determined by the net stocking density, length of the line used, and desired planting density. If complete coverage is desired, it is recommended that buoys be placed so that adjacent buoy radii overlap.
Retrieval. BuDS lines can be retrieved 4 to 5 weeks following deployment or after all of the seeds have been released. Depending on the monitoring requirements, it may be necessary to mark the location of the blocks so that the contribution of each line can be assessed. In this case bring along separate half cement blocks, bricks, or similar markers to place on the bottom when the cement blocks and lines are removed. Following retrieval, the lines and nets should be left in the sun to dry and rinsed to remove algal growth. In most cases the nets should be cut from the lines to allow for separate storage and handling.

MONITORING: The success of an SAV restoration program cannot be properly assessed without implementing a monitoring program. Given that BuDS is a seed-based technique, monitoring for seedlings is the most logical and effective alternative. Depending on the region, seedlings normally emerge from the bottom sediments during late winter and early spring. Given the circular pattern of buoy movement, seedling distribution typically follows a broken or continuous arc at a distance from the anchor approximately equal to the line length depending on depth, tidal amplitude, wind conditions, and other factors (Figure 12). Young seedlings, less than 2 in. (5 cm) tall, are exceedingly difficult to locate in situ and it is usually necessary to wait until the seedlings have reached a height of about 3-4 in. (7.5-10 cm) to facilitate monitoring. However, delaying monitoring until mid- to late summer may render it nearly impossible to distinguish between seedlings and adult shoots.

IMPORTANT CONSIDERATIONS FOR PLANNING: As with any other eelgrass restoration method, site selection is critical to the success of the effort. In many areas, it may be necessary to obtain a permit to collect flowers and/or plant eelgrass. Be sure to check with local municipalities and natural resource managers during the early planning stages. Local experts will
likely know the best locations to attempt a restoration project based on historic occurrence and current environmental conditions. Typically, restoration sites should not be closer than 300 m to an existing meadow. Medium- to low-energy sites with sandy to sandy/mud bottom have proven the best areas for BuDS deployment. Deploying large numbers of buoys may also raise a navigational or legal concern. Collaboration with and education of nearby waterfront homeowners and local boaters is recommended to avoid additional conflicts. Avoiding areas of high boat traffic, commercial or recreational fishing, or other human use is also recommended to avoid the attractive nuisance issue.

Selecting an appropriate donor site is just as critical as the planting site. In addition to the need for an abundance of reproductive shoots, it is also necessary to choose a site that is logistically convenient for boat or land access. Before engaging in large-scale seed collection, it is important to understand the contribution of seeds to the maintenance of the donor meadow. Observations of heavy seedling recruitment may indicate that seeds are a primary mechanism for maintenance of the meadow, making the site a poor candidate for seed collection. In many areas, seeds appear to be important to the survival of meadows that have experienced lowered shoot density to physical disturbance or other factors. Meadows that are under stress from anthropogenic or other disturbance should be avoided. Although even intense collection efforts can have little effect on the meadow-wide density of reproductive shoots, more than 50 percent of the flowers in one area should never be harvested.

The BuDS system can be modified to accommodate a range of environmental conditions. Line length can be adjusted according to depth at low water and tidal amplitude, as well as the desired coverage for each buoy. In areas of extreme tidal amplitude, subsurface buoys can be deployed using a line length shorter than the depth at mean lower low water. Such systems may require the use of a screw anchor and require less lifting capacity than the buoys listed above. Subsurface buoys can be fabricated by cutting the larger ones into quarters.

Depending on the seeding rate required, additional reproductive shoots can be stocked into each net, or additional nets can be attached to single buoys. Depending on the coverage desired, buoy spacing can be increased so that adjacent radii do not overlap; however, given the recent evidence that distances of 3 m between flowering shoots can lead to pollen limitation and reduced reproductive output (Reusch 2003), a close spacing may be warranted. Conversely, a test planting consisting of a single buoy can be used to assess the potential effectiveness of seeding at candidate restoration sites before large-scale efforts are undertaken.

Availability of pearl nets can be a problem depending on the time of year and local suppliers. It is important to contact an aquaculture supplier to place an order for nets months prior to expected deployment. Failure to do so may result in the inability to obtain nets. Although a specific net type and mesh size are recommended, smaller mesh sizes are available and can be used for BuDS. The authors have successfully used the 6-mm and 4-mm mesh sizes in previous deployments. Mesh sizes smaller than 4 mm should be avoided as they will more easily foul and may also prevent efficient seed release. Round pearl nets are also available and may be used in place of the square ones described here. Although effective as substitutes, they do not have the same internal structure as the square nets and may not be as easily handled and transported once stocked.
POTENTIAL FOR FURTHER USE: BuDS, as a modular system, can be adapted to fit a multitude of needs. Anchors can vary from the simple cement blocks described above or heavier anchors could be substituted to provide maximum anchorage in exposed sites. The mesh size of the nets can be changed to accommodate plants with larger seeds than *Z. marina* (e.g., *Posidonia*). Other species that may be effectively seeded using the BuDS system include *Potamogeton pectinatus*, *P. pusillus*, *P. perfoliatus*, and *Ruppia maritima*. For other species, such as *Posidonia oceanica* and *Thalassia testudinum*, however, experimental trials may be advisable to develop appropriate adaptations.

COSTS: The simplicity of our method, with highly repetitive setup and deployment, is conducive to volunteer labor and the costs of storage and handling facilities are avoided. Each buoy/net/anchor setup costs only about $6.50, so the total cost of materials per ¼ acre would be approximately $100, depending on desired buoy spacing. The costs of deployment and monitoring would be dependent on dive time (if SCUBA is necessary) and travel time (if boat, etc. is needed to access planting site). Additional materials used for transport (fish totes, burlap, etc.) could be substituted with whatever materials are available and serve the same purpose.

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POINTS OF CONTACT: For more information, contact Chris Pickerell (cp26@cornell.edu), Habitat Restoration Specialist with Cornell Cooperative Extension, Marine Program at the Cornell Marine Lab, 3690 Cedar Beach Rd., Southold, NY 11971, or Deborah Shafer (Deborah.J.Shafer@erdc.usace.army.mil), Engineering Research and Development Center, 3909 Halls Ferry Road, Vicksburg, MS, 39180. This technical note should be cited as follows:


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