

Gas Output Varied Reproducibility in GDS-80

INTRODUCTION

As GDS-80 is so simple and convenience in gene transfer experiment, each condition setting of GDS-80 is very important. In the previous article mentioned that setting of GDS-80 is quiet important and closely related to the deliver efficiency. Including the pressure setting, gas output intensity, and gas flow rate adjusting, GDS-80 should be set at the most optimal condition prior to perform the bombardment. When it comes to adjust the gas output intensity, rounds setting in the back of the GDS-80 main body should be adjusted to have most even spread sample distribution. Why is it so important to adjust the proper gas output intensity at proper condition in GDS-80?

EQUIPMENTS AND MATERIALS

- Whole set of GDS-80 with 4.5 mm barrel, including pressure regulator and hose assembling (Wealtec)
- Laminar flow
- 3 cm target spacer (Wealtec)
- Gas cylinder with Helium gas (99.999%) over than 1000 psi.
- Samples: Onion epidermis.

PROCEDURES

1. Setup the GDS-80 system according to the standard procedure in the manual.
2. Sterilize whole equipments and materials with proper treatment prior to the experiment.
3. Assemble the barrel and sample loading sleeve with main body of GDS-80 inside the laminar flow and connect the whole GDS-80 system.
4. Set the deliver pressure at 50/60 psi and the gas flow rate around 10~15 L/min.
5. Change the rounds setting in the back of the GDS-80 main body to have 0~4 rounds.
6. Prepare the plasmid DNA/ gold particle solutions prior to perform the bombardment.

(1 μ g DNA/ 0.6 mg Gold)

7. Cut the onion epidermis in proper size (3 cm x 3 cm x 2 mm) and sterilized before use.
8. Aliquot 10 μ L DNA sample solution into the sample loading hole.
9. Perform the bombardment toward the onion with the help of 3 cm target spacers.
10. Incubate the sample for 2 days and observe the result under the microscope.

RESULTS

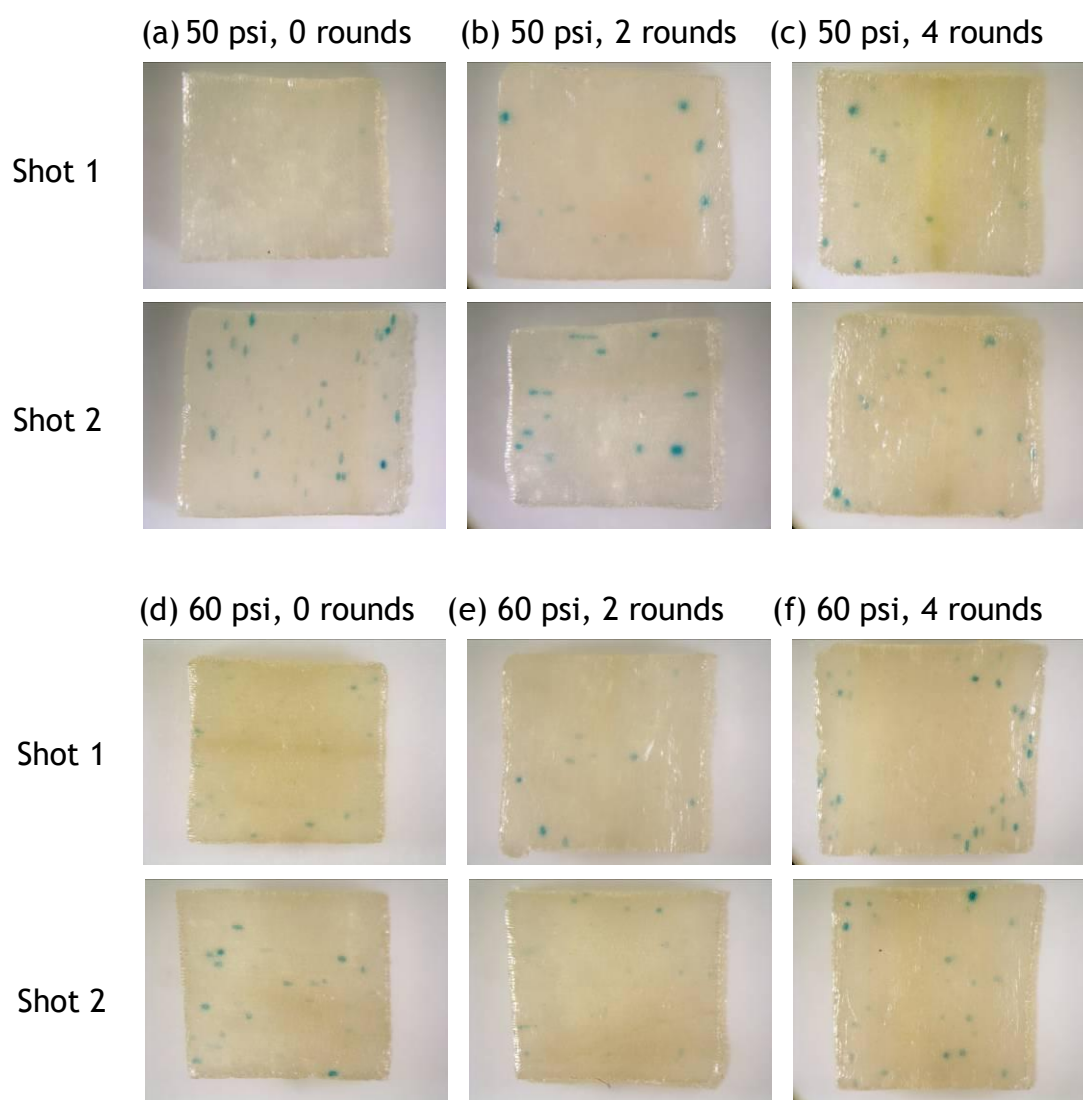


Figure 1. *Twice bombardments result of onion samples delivered with different round and pressure settings. (a)~(c) 50 psi and (d)~(f) 60 psi.*

DISCUSSION

The results in the *fig. 1* were repeated at least six times for each setting. As in the result, when the setting is different from 0 rounds to 4 rounds, all setting conditions of GDS-80 can get transfection result. However, the reproducibility of each setting was so different. While setting with 0 to 2 rounds under 50 or 60 psi, the GDS-80 gas output is not strong enough so that the transfer efficiency in *fig. 1(a)(b)(d)(e)* were different from shots to shots. Using of rounds setting like this, users will not get the repeated data under the same condition. As the round setting getting higher, the gas output becomes more stable and the reproducibility gets better. When the rounds' setting gets higher, GDS-80 can perform with stronger gas output and more stable transfer result. Although adjust the GDS-80 with more rounds will enhance the deliver efficiency when transfer gene onto samples with harder surface. Users will have the transfer result lean toward aside on the samples because of setting with the gas output intensity under uneven spread condition of the GDS-80. Users should set with proper rounds setting according to the instruction manual to have most even spread delivery and most reproducible transfection to ensure have same condition at each shot.