

Seed Inoculation: Master Farmer Training Practical

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Purpose. To demonstrate the preparation of stickers, methods of coating seeds with inoculant and a seed pelleting technique.

Conceptual Background. Sticker materials are recommended to bind the rhizobia to the seed. The stickers used in the following demonstrations are gum arabic and sugar, and are compared to water. Both of these adhesives must be dissolved in water before use. Two seed coating methods are used. The *slurry method* and the *two-step method*. In the *slurry method*, inoculant is first mixed with the sticker. The resulting slurry is then applied to the seeds. The *two-step method* requires seed coating in two stages. First, the seeds are coated with the sticker. The inoculant is then added and coated onto the sticky seeds. Note that the amounts of sticker used for each method vary with seed size (Table 1) and in this practical soybean seeds are used.

Under certain conditions, it is advisable to *pellet inoculated seeds* with a protective layer of powdered calcium carbonate or rock phosphate. This treatment is most commonly done with seeds of pasture legumes, but may also be practiced with grain legumes, particularly where grown in highly weathered and nutrient depleted soils. The pellet is applied after seed coating by either the slurry method or the two-step method. The seeds are rolled in the pelleting material immediately after inoculation while they are still wet and sticky.

Table 1. The amounts of sticker, inoculant and mineral coating required for selected grain legumes.

legume seed	seed weight g/seed	----- slurry method -----		----- two-step method -----		----- two-step pelleting -----		
		sticker ml/kg seed	inoculant g/kg seed	sticker ml/kg seed	inoculant g/kg seed	sticker ml/kg seed	inoculant g/kg seed	coating g/kg seed
soybean	0.14	30	10	20	10	40	10	200
bush bean	0.42	22	10	19	10	33	10	160
climbing bean	0.45	20	10	18	10	30	10	150
groundnut	0.50	18	10	16	10	25	10	100
cowpea	0.14	30	10	25	10	40	10	200

Materials. The amounts of materials needed should be gauged according to the number of participants in the exercise. The list of materials below is based on 15 to 21 participants divided into three groups.

1. 500 ml bottled water (x9)
2. Tablespoon for measuring (x3)
3. Teaspoon for measuring (x3)
4. Two liter plastic bags (x12)
5. Wooden stirring spoon (x3)
6. Small plastic funnel (x3)
7. Marking pen (x3)
8. Plastic buckets, 3 liter capacity (x3)
9. Plastic bucket, 20 liter capacity with lid
10. Gum Arabic, granular (3 x 200 g)
11. Sugar, granular (3 x 100 g)
12. Agricultural lime (calcium carbonate), finely powdered (3 x 200 g)
13. Soybean inoculant (3 packs x 100 g)
14. Soybean seed (12 kg in 1 kg bags)
15. Paper sheets (x27)

Note that measurements are provided in grams, liters and milliliters. In the field it is more practical to convert these volumes and measurements into more convenient units. One level teaspoon holds five ml of sticker and one heaped teaspoon of inoculant contains five grams. Three teaspoons make one tablespoon.

1. Preparing the sticker

Gum arabic. Heat 500 ml water in plastic containers by placing them in the sun (or on the dashboard of a auto) for 1 hour prior to the demonstration. Open bottle, remove 200 ml of water, add 200 g of gum arabic (or 5 teaspoons) using the plastic funnel and shake until dissolved. Set aside to cool. This procedure results in a 40% gum arabic solution. Mark the plastic bottle as containing gum arabic solution. If the weather is cloudy and cool, it may be necessary to warm the water over a stove to dissolve the gum arabic, and replace it into the plastic bottle using the funnel.

Sugar. Remove about 100 ml of water from a 500 ml water bottle. Add 100 grams of sugar using the plastic funnel. Shake until dissolved. This procedure results in a 20% sugar solution. Mark the plastic bottle as containing sugar solution. It is not necessary to warm the water before dissolving the sugar into it.

2. Inoculating legume seeds using the slurry method

Preparing the slurry. For coating soybean seed, slurry consisting of 1 part of inoculant and 3 parts sticker is recommended. For demonstration and practice of this procedure, only a small amount of seed will be coated. Remove 10 g of BIOFIX inoculant (two heaping teaspoons) from the packet and place it into a 300 ml container. Add 30 ml of water (or two level tablespoons). Mix the inoculant and the water until uniform mixture is achieved

Slurry inoculation. Place one kg of soybean seeds (about 1200 ml or 2½ 500 ml mugs and place them into the 3 liter bucket. Add 40 ml of the slurry. Stir the seeds with a wooden spoon until they are uniformly coated with the inoculant slurry. After coating, spread the seeds onto clean paper and allow them to dry. Mark the paper sheet as holding a slurry-water preparation. Repeat the seed coating procedure with slurries made from other sticker solutions to achieve the treatments as summarized below:

- 1 kg of soybean seed coated with 40 ml of a slurry prepared by mixing 10 g of BIOFIX inoculant with 30 ml of 40% gum arabic solution. Mark the paper sheet as holding a slurry-gum arabic preparation.
- 1 kg of soybean coated with 10 ml of a slurry prepared by mixing 10 g BIOFIX inoculant with 30 ml of sugar solution. Mark the paper sheet as holding a slurry-sugar solution preparation.

After coating compare the three different slurry preparations, inspect them for evenness of coating and for adhesion quality. The best coating is usually achieved with gum arabic. Sugar should be second best. Water as an adhesive appears good initially but the inoculant tends to flake off the seed after drying. *Conclusion, whenever possible, a gum arabic sticker should be used for seed coating.* The slurry method of legume seed inoculation described in this section is presented in Illustration 1.



Illustration 1. The slurry technique first mixes the inoculant and adhesive and then combines them with the legume seed.

3. Inoculating seeds using the two-step method

Place 1 kg of soybean seeds into a plastic bag. Add 20 ml of water (1 level teaspoon plus 1 level teaspoon). Inflate the bag and twist it shut in such a way that the walls of the bag are rigid. Shake the bag vigorously for about one minute until the seeds are uniformly coated. Open the bag and add 10 g of BIOFIX inoculant (two heaping teaspoons). Close the bag as before and shake again, but more gently for one minute. Note that too vigorous or prolonged shaking may dislodge the inoculant from the seeds. Immediately after coating, spread the seeds on paper and allow them to dry in a shady place. Mark the paper sheet as holding a 2-step-water preparation. Repeat the coating procedure with the following treatments:

- 1 kg of soybean seed wetted with 20 ml of the 40% gum arabic solution and then coat with 10 g of BIOFIX inoculant. Immediately after coating, spread the seeds on paper and allow them to dry in a shady place. Mark the paper sheet as holding a 2-step-gum arabic preparation This procedure is described in Illustration 2.
- 1 kg of soybean seed wetted with 20 ml of 20% sugar solution and then coat with 10 g of BIOFIX inoculant. Immediately after coating, spread the seeds on paper and allow them to dry in a shady place. Mark the paper sheet as holding a 2-step-sugar solution preparation.

There should now be six different preparations of inoculated seed spread on marked paper sheets. Compare the three different two-step inoculated seeds to one another and the slurry inoculations (Table 2). When we compare the two-step and slurry treatments, the seeds from some of the preparations appear darker in color. This indicates that more inoculant was applied to each seed by this method. Rank the six preparations by appearance on a scale of 1 (no inoculant on seed) to 5 (darkest appearance).



Illustration 2. The two-step procedure first combines legume seed and adhesive, and then mixes them with the rhizobial inoculant.

Comment. The two-step method allows for more inoculant to be applied to the seed, especially when gum arabic is employed as an adhesive. If we used for instance, 30 ml of the sticker, we could coat as much as 100 g of inoculant onto 1 kg seeds, which results in 10 million rhizobia per seed if the inoculant contains one billion rhizobia per gram. Such a rate is, however, excessive as it is not cost effective for farmers under normal conditions. To apply more than this amount of sticker is not practical because the seeds would clump if more than 30 ml of sticker per kg of soybean seeds is applied

Table 2. A template for comparing the results of different seed inoculation procedures.

Adhesive	Inoculation procedure	
	Slurry	Two-step
	----- ranking (1 to 5) -----	
Water		
Gum Arabic solution (40%)		
Sugar solution (10%)		

4. Inoculating larger amounts of seed

The upper limit for inoculating seed using plastic bags is about five kg using the two-step method, otherwise the risk of puncturing the bag and spilling seed and inoculant grows too great. A more useful container for larger amounts of seed (e.g. 10 kg batches) is a 20 liter plastic basket with a lid. In this case, place 10 kg of seed into the plastic bucket and add 200



Illustration 3. Seed coating with limestone may be performed following inoculation with rhizobia but requires that additional adhesive be applied.

ml of 40% gum arabic solution. Close the lid and shake for one minute. Open the container and inspect to assure that the seeds are evenly coated, not clumped together and that no sticker is clinging to the walls. Add 100 g of inoculant (or an entire packet of BIOFIX inoculant) and again close the lid. This time shake more gently for one minute, open the lid and inspect seeds for uniformity coating. If coating is not complete, immediately continue shaking for 30 seconds. After coating, spread the seeds out on a clean canvas. After the seeds have dried, place them back into the bucket and store under cool, shaded conditions until sowing as soon as possible. Even larger amounts of seed (e.g. 20 to 40 kg) may be inoculated using a large plastic or canvas sheet, mixing the seed and adhesives and inoculants by rolling.

5. Pelleting Seeds

Pelleting after slurry application. Make a slurry from 40 ml of gum arabic solution and 10 g of inoculant. Place one kg of soybean seeds in a 3 liter plastic bucket and add the slurry. Stir the mixture until uniformly covered. Spread the seeds on a clean paper sheet and add 200 g of finely ground limestone (or rock phosphate). Roll the seeds on the paper sheet until they are evenly pelleted. Spread the seeds across the paper sheet and allow them to dry (see Illustration 3).

Pelleting after the two step method of inoculation. Place one kg of soybean seeds into a plastic bag and add 40 ml of gum arabic sugar sticker. Close bag and shake until the adhesive evenly coats the seed. Add 10 g of inoculant and shake gently for one minute. Open the bag and add 200 g of limestone and again shake gently until all seeds are uniformly coated. Spread pelleted seeds on paper and allow to dry.

Compare the two preparations for evenness of coating, firmness of pellet and amount of calcium carbonate adhering to the seed. Note that to accommodate the pelleting material, more sticker must be applied. Water alone is unsuitable for pelleting because it does not produce a firm, evenly coated pellet.

Amplifying farmer training. Each of the Master Farmers is provided a set of materials and instructions so that they may repeat the inoculation demonstration within their own associations. To do this, a package is prepared that contains the following materials:

1. 500 ml bottled water (x3)
2. Tablespoon for measuring
3. Teaspoon for measuring
4. Two liter plastic bags (x4)
5. Wooden stirring spoon
6. Small plastic funnel
7. Marking pen
8. Plastic bucket, 3 liter capacity
9. Plastic bucket, 20 liter capacity with lid
10. Gum Arabic, granular (200 g)
11. Sugar, granular (100 g)
12. Agricultural lime (calcium carbonate), finely powdered (3 x 200 g)
13. BIOFIX Soybean inoculant (1 package x 100 g)
14. Soybean seed (9 kg in 1 kg bags)
15. Paper sheets (x9)
16. Inoculation protocol

The seed inoculation is conducted as a demonstration by the Master Farmer and records are kept concerning the time, place, number of participants and overall impressions of the training activity. These results are reported to the N2Africa Inoculant Delivery Specialists through the Farm Liaison Officer.