Steel balls cleaning method for RNA and DNA extraction protocol (Molecular Tree Breeding Lab-NCSU)

Our group reused steel balls by following a simple cleaning protocol and an appropriate handling. **Tip**: Steel balls can be cleaned at the end of the extraction protocol but we recommend cleaning the beads as soon as possible taking advantage of the waiting-time between steps. Some of the reagents diluted in the lysis buffers are corrosive and we have noticed that steel balls can be rusted easily after a couple of hours of exposition to these reagents. **Tip:** wear gloves while handling the beads before and after cleaning.

- Place a metallic mesh strainer into a plastic container with some water on it.
- Remove the steel balls from the bottom of the 1.1 ml tubes by simple shaking the strip tubes against the metallic mesh strainer.
- Rinse the beads by changing the water in the plastic container 1-3 times. **Tip:** we have noticed that this step reduces the amount of water required to clean the steel balls that is usually used when is rinsed under running tap water. It also helps to keep the contaminated water into a container and can be discarded with other guanidine residues in an appropriate manner.
- Transfer the beads (with any plant material attached) to a 50 ml falcon tube.
- Incubate steel balls in 3.0% (w/v) sodium hypochlorite (bleach) for 3 minutes at room temperature (15–25°C).
- Rinse the steel balls thoroughly with distilled water to remove the sodium hypochlorite.
- Prepare a tray with a paper towel on it and disperse the steel balls on the tray. Use heat resistance plastic trays or metal trays.
- Dry beads at 50°C in an oven before use.