AMPure XP beads purification(Quail et al., 2009)

1. Let the AMPure XP beads come to room temperature before the experiment
2. Mix the beads well so that they appear homogeneous and consistent in color.
3. Add an equal volume of AMPure beads (e.g. 50 μl of AMPure XP to 50 μl of sheared DNA) to a new 1.5 ml Eppendorf tube.
4. Pipette DNA sample into this tube (e.g. 100 μl of sheared DNA). Mix well on a vortex mixer. Briefly centrifuge for 2 seconds to spin down residues on the side of the tube.
5. Incubate the DNA with the beads for 5 minutes.
6. Put the tube in the magnetic stand and wait for the solution to clear (~3 minutes).
7. Remove the supernatant from the tube and add 500 μl of 80% ethanol without disturbing the pellet.

*The original AMPure protocol recommended use of a 70% ethanol wash solution. This needed to be freshly made, as over time it becomes more dilute since it absorbs atmospheric water and ethanol evaporates. When you rinse with more dilute ethanol more DNA gets washed away. We have found that this can be avoided by using an 80% ethanol solution instead. This washes just as well but does not need to be freshly prepared each time*.

1. Repeat step 7.
2. Remove all ethanol, including residual droplets and dry the samples for 5 minutes in the magnetic stand on the bench with the tube lids open. Do not dry longer than 5 minutes. The beads get too dry and yield decreases.
3. Add 30 μl EB buffer, mix well on a vortex mixer, and incubate for 5 minutes at room temperature.
4. Put the tube in the magnetic stand and leave for 2 to 3 minutes.
5. Remove 30 μl of the supernatant to a fresh 1.5-ml tube. (Take care not to pipette any beads)
6. Proceed to template preparation (Basic Protocol 3).

Reference

Quail, M. A., Swerdlow, H., & Turner, D. J. (2009). Improved protocols for the illumina genome analyzer sequencing system. *Curr Protoc Hum Genet*, *Chapter 18*, Unit 18 12. <https://doi.org/10.1002/0471142905.hg1802s62>