## PROTOCOL FOR LACCASE ACTIVITY DOSAGE USING BOTRYTEST

Preconditions: the analysis must be done on non-sulphited must or wine.

If necessary, the must/wine can be desulphited before analysis, by adding oxygenated water\*  $(150 \,\mu\text{L}\,\text{of solution at }30\%/100 \,\text{mL}\,\text{of must/wine})$ .

\*Oxygenated water is not provided in the kit.



- Using the pipette supplied, put 5 mL of must or wine to be analysed into the syringe.
- Put the syringe on top of the tube and leave to percolate until the resin contained in the syring is wet at its base (3 to 10 minutes). Then insert the plunger into the syringe and press lightly and slowly. Collect the first millimeter (up to the first marker) in the tube.
- Add 1.4 mL of buffer solution (up to the second marker) then 0.6 mL of "BOTRYTEST laccase" reagent (up to the third marker on the tube).

  Mix the content by shaking the tube.
- After exactly 3 minutes (use a stopwatch or timer), the laccase activity can be determined by comparing the color development with the color scale chart.

