

## Instructions for MLF Chromotography Kit

Stock Code: 94271

### Testing of Malic Acid in wines by chromatography

#### Operating Instructions:

- Take a sheet of chromatographic paper. In the longer side, draw a line with a pencil, parallel to the edge with a gap of about 3 cm.
- On this line, mark points about 3 cm from one to another and put a number below the line with a pencil.
- • With a micropipette, lay down on a point 3 mm<sup>3</sup> (1 mm<sup>3</sup> = 0,001 ml) of wine in several strokes (if the wine has a very low malic acid content, you can put 5 mm<sup>3</sup> or more).
- Proceed the same way on each point with a different wine.
- Lay down 1, 2 or 3 indicator with known contents (malic acid solution) to make subsequently estimations for the several samples.
- Let the patches completely dry up then roll up the sheet of paper in a cylinder, stapling together the edges, without contact between them. The edge with the patches is at the bottom, outside the cylinder.
- In the glass cylinder, mix 40 ml of butanol with bromophenol blue and 16 ml of acetic acid ½. It is possible to use volumes more important, but with the same ratio of 5 to 2 for these liquids.
- Pour the mixture in the flask. Place the rolled sheet standing on the centre and close the flask hermetically. These operations have to be done quickly, the stopper has to be greased before.
- The liquid must not be in direct contact with the wine patches, it goes up by capillarity action. When it arrives at about 1 cm from the upper edge (after 2h30 or 3h), remove the sheet of paper from the flask, without soiling it, and let it dry up hanged up in a dry and aired place, protected from acid or alcalin vapors. This step is long, you can use a ventilator to shorten it.
- When the paper is completely dry, some yellow patches due to the wine appear on the blue surface, upright the marked points.
  - at the bottom : tartaric acid
  - at the middle : malic acid
  - at the top : lactic and succinic acids
- Thanks to the indicator laid down before, you can locate and estimate the malic acid in the tested wines.

## Precautions

The flask has to be corked, except for the strict time needed to insert the paper.

- The mixture has to be changed every new testing.
- If the same pipette is used, it has to be rinsed with the wine to analyse.
- At the end of the testing, clean the pipette with distilled water and shake it (never blow inside).