



THE BEVERAGE PEOPLE

Malolactic Chromatography Set and Reagents

Vertical Style Kit

Vertical Chromatography Set Contains:

One gallon wide mouth jar and lid

Funnel

100 Capillary Pipets

Chromatography Paper - 6 Sheets (Store in dark)

Chromatography Solvent (n-butyl alcohol, formic acid and bromcresol green) - 4 oz.

Acid Standards Set - 3% solution (Malic, Lactic and Tartaric)

What the Kit Does

The Malolactic Chromatography Set and Reagents are designed to detect the organic acids, Tartaric, Malic and Lactic in grape juice, must or wine. The test uses a special paper that allows the solvent to rise through it, while attracting the acids to the solvent and to the paper, thus leaving a marker for each acid. Each acid has a unique attraction to the solvent and paper and will leave a mark in a consistent location.

The test consists of spotting standard acid solutions and wine or juice samples on chromatography paper, then resting the bottom edge of paper into the solvent solution and waiting until the rising solvent line reaches the top edge of the paper. This will take about 8 hours. Then the paper is removed and allowed to dry, which in drying allows the formic acid to evaporate and yellow spots appear on the blue-green background. These spots may then be identified by comparing their location against the acid standard's location.

Preparation of Chromatogram

Place one sheet of paper on a clean, dry work surface, handling it by the edges.

Using a pencil, draw a line all the way across the long side of the sheet and one inch up from the bottom of the paper. Mark a small "x" on the line for each acid standard and then each juice or wine sample. To allow for space between each mark, do not mark more than six spots. Label each "x" with the name of the acid or sample in pencil below the line.

Dip the clear end of one glass capillary pipette into one standard and fill about 1/4 of the tube. Drop the liquid onto the paper over the appropriate "x". Make the spot as compact as possible. Continue with a new pipette for each acid and each sample. When all spots are made, allow the paper to dry for 10 minutes. (You can speed this up with a hairdryer.) Repeat spotting the paper with the same acids over the same spots. Repeat drying, leaving until fully dry.

Pour solvent into the jar until it is about 1/2 " deep.

Staple the short ends of the paper together to form a cylinder. Staple once near each end. Do not overlap the edges of paper.

Lower the paper into the jar and cover with the lid. The bottom 1/4 " of paper must be immersed in solvent, but the level must still remain below the ruled line of spots.(See diagram 1)

Leave the jar sealed until the solvent moving up the paper reaches the top edge, approximately 5- 6 hours. (You may safely leave it in solvent overnight.)

Open the jar and lift out the paper, carefully handling only a corner of the paper, and hang it with a clothesline pin or a hook from a convenient, ventilated place to dry. (Tip: if you place an open bottle of plain ammonia near the drying paper, you will have a better definition of color between the background and the spots after the paper dries.)

Return the remaining solvent to the original container via the funnel. Solvent remains strong for at least a year, and maybe longer depending on how many tests are run. Allow 8 hours for drying.

When dry, the paper will have a blue-green background with yellow spots of acids. Finish the drying process with a warm hairdryer if the spots are weak in color. This will dehumidify the paper and bring up the color. Sometimes waiting a full day and then viewing gives a brighter definition to the spots.

Identify the acids present in your juice or wine sample by comparing the locations of the yellow spots. For example, the tartaric spot of acid will mark about 2" up from your tartaric acid "x". If tartaric is present in your sample, then 2" above the sample "x" there will appear a yellow spot. The same will follow for each acid. The malic spot is 2" higher than the tartaric and the lactic spot is another 2" higher. (See diagram 2)

How to interpret the results.

Bright yellow in the malic location and an absence of color in the lactic location means the sample juice or wine has not begun to do malolactic fermentation. Incomplete malolactic is indicated by yellow spots appearing in both the lactic and the malic locations. Completed malolactic fermentation will show an absence of a yellow spot in the malic position, but a bright yellow spot present in the lactic position.

A persistent yellow spot will remain in a wine that has utilized acid blend or malic acid to raise acidity, because a portion of this type of malic is not fermentable. As long as a strong lactic spot develops in such a wine, you can be sure that the fermentation has taken place. To be absolutely sure that the wine is finished, a wine laboratory would have to run an enzymatic M/L test to determine if more malic was fermentable or not.

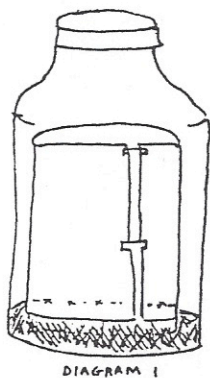


DIAGRAM 1

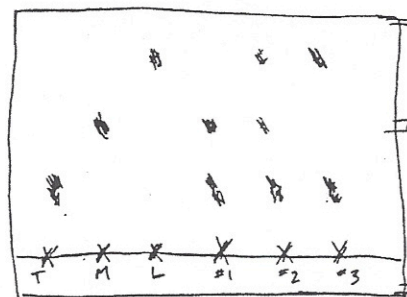


DIAGRAM 2