A rapid method for making large numbers of crosses.
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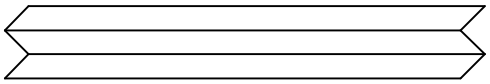
Often it is necessary to intercross large numbers of strains. A rapid way to do this, a modification of the standard crossing procedure, uses liquid medium in large (~6 inch) test tubes. Liquid Westergaard and Mitchell medium containing 0.2% sucrose is used. Approximately 5 ml is dispensed into each tube. Into each tube is dropped a 12 x 6 cm piece of filter paper pleated on its long axis in the following way:



If necessary push the paper to the bottom. This piece of paper wicks up medium and acts as a surface on which sexual development will take place. The special pleat prevents sagging and keeps the paper away from the walls of the tube. Rigid plastic or metal caps are ideal because they can be easily removed for inoculations.

When the tubes have been autoclaved and cooled, a drop or two of conidial suspensions of each parent is dripped down the filter paper. If the tubes are arranged in racks in the appropriate grids, many hundreds of crosses can be made by simply inoculating drops along rows and columns of a grid.

Ascospores shot onto the walls can be recovered in the usual way. For ascus analysis, the filter paper and its perithecia can be removed from the tube with ease. Linear asci are dissected out of perithecia, whereas nonlinear asci can be recovered by placing a cut-out section of paper carrying mature perithecia close to a fresh 4% agar collecting surface.

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