

TLC METHODS

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(This is one of several files of miscellaneous thoughts on TLC methods, which will eventually be merged and rearranged)

TLC Annotations

Annotation slips briefly indicating the chemical contents of were placed in the packets of all borrowed specimens that were sampled for chemical analysis. The primary purposes of these annotations were: 1) to indicate that a sample had been used for TLC, to avoid unnecessary depletion of the specimen by further removal of fragments, and 2) To provide myself and other workers at least some idea of the chemical constituents, as a basis for further investigation... Because of the large number of specimens that were analyzed and annotated, the numerous unknown substances, and many changes in techniques and interpretations over the course of the study, it was not feasible to spend an excessive amount of time in preparing or revising these annotations. In many cases, these annotations were preliminary and incomplete (especially with regard to most terpenoids, fatty acids and unknown substances).

The Rf classes and other characteristics given for the unknowns were often only quick estimates, and were sometimes erroneous for various reasons. Descriptions of postcharring fluorescence and various other characteristics of the unknowns were often not given. Work on identification of the unknowns has been a gradual process, and is still continuing.

The annotation slips cite the three solvent systems as being the "A", "B" and "C" of Culberson (1970, 1972). However, it should be pointed out that in all of my TLC, toluene was substituted for benzene in Solvent A (as recommended by _____, 19__), and methyltert.butyl ether was substituted for diethyl ether (as recommended by _____, 19__). These changes, made for safety reasons, do have some affects on the Rf values of some substances. Elix (1987) refers to the modified B solution as B'.

In several cases the names given for the substances on the annotations are older descriptive synonyms rather than the correct chemical name (e.g., "consporomic acid")

=2'0demethylpsoromic acid).

Usually the only indication of relative quantity of the substances given was the qualifier "trace" used when the presence of a substance was indicated only by a very faint spot.

More complete and updated descriptions of the unknown substances, and a certain amount of additional and revised information on the known and unknown constituents of many of the individual specimens (especially types) will be given in my publications. As further progress is made in identifying or at least better characterizing the unknowns, information on the chemical constituents of particular specimens will be made available to workers with a special interest in lecanoras or their chemistry.

Record Keeping

When large numbers of plates are being run (especially in critical where there are many unknowns), it is necessary to assign a unique, unambiguous number to each spot sample. In my study I have used a combination of a "set" letter (A through Z, AA through ZZ) marked on each of a group of plates, and consecutive numbers (to 3 digits) for each spot in that set. This kind of combination can be continued using AB, AC, etc., BA, BC, etc., and can be convenient for keeping and updating records, and for locating and reexamining stored plates. These "Sets" of four to seven plates were often run developed and (partially) interpreted together on a single day and thus should be relatively comparable.

Solvent Systems

Problems with Hygroscopic Plates

Fluorescence in Longwave Ultraviolet

To clearly see and distinguish the different colors of fluorescence of spots in longwave ultraviolet light (before or after charring), three things were found essential: 1) using a sufficiently intense uv light source (give specs. of one in Dennis's lab), 2) working inside a booth kept completely dark by black curtains, and 3) wearing greenish tinted goggles (give specs), which also protect the eyes.

I learned the above rather late in my studies...(how do I phrase that?).

Fluorescence After Charring

The color and intensity of the postcharring fluorescence of many substances change over time, and are affected by a number of variables in the running and development of the plates...