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Natural Pigments

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I. INTRODUCTION

Thin-layer chromatography (TLC) remains a popular method for the analysis of natural pigments. The readily available equipment is easy to use, and operating costs are low. The progress of separation can be followed throughout the separation, the time required for completion of the process is usually short, and the results are immediately visible. Together with the robust nature of most of the systems that have been developed, these facts ensure that TLC will continue to enjoy popularity where rapid qualitative analysis required. Good separations of pigments obtained from these TLC systems are important to demonstrate chromatography to students of chemistry and biology, but the systems are also suitable for field work in biology and agriculture. It should be noted that the quantities of pigments present in most natural sources are such that TLC is often the method of choice for preparative separation. Provided that suitable precautions are taken, good quantitative analysis can often be obtained.

II. TLC IN GENERAL

A. Stationary Phase

A wide variety of TLC plates are commercially available, and many of these have found applications in the analysis of pigments. In this chapter, individual sorbents are discussed under specific pigment groups. It is also possible and cheaper to prepare TLC plates in the laboratory. Such laboratory plates can, with practice, provide excellent results and thus allow access to stationary phases or phase mixtures that are not otherwise available. The descriptions given below should allow preparation of a 0.25 mm layer with a surface area of about 2 m × 20 cm.

1. Preparation of Layers

a. Silica Layers. Silica gel G (30 g) is mixed with water (60 mL), and the slurry is transferred to the TLC plates with a commercial spreader. The plates are allowed to dry for 30 min followed by activation at 120°C for 1–2 h.

b. MgO Layers. MgO (10 g) and kieselguhr G (10 g) are passed through a 60 mesh sieve followed by mixing with 80 mL of distilled water. The slurry is transferred to the plates with a commercial spreader. The plates are allowed to dry for 12 h.

c. Cellulose Layers. Cellulose powder (15 g) is mixed with distilled water (100 mL) and homogenized in a blender for 30 s. The slurry is transferred to the plates with a commercial spreader unit. The plates are allowed to dry for 6 h.

d. Sucrose Layers. Sucrose (65 g) is passed through a 60 mesh sieve followed by mixing with 100 mL of petroleum ether (60–80°C). The slurry is transferred to the plates with commercial spreading equipment. The plates are allowed to dry for 30 min.