

Routine Preparation of Silica Gel Media Using Silicate Solutions of Varying pH^1

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Existing methods for the preparation of silica gel media are either time consuming or produce gels of unfavorable chemical and physical properties. Ion-exchange column chromatography can be used to obtain silicic acid from sodium silicate solutions (K. T. Wieringa, Antonie van Leeuwenhoek, *J. Microbiol. Serol.* **32**:183, 1966). But because of rapid gelation, pH and nutrient adjustments must be made immediately after collection of the silicic acid, and sterilization must be achieved by boiling prior to pouring the plates. Another improvement is the use of sterile phosphoric acid to cause rapid gelation of sterile nutrient-amended potassium silicate solutions (B. Funk and T. A. Krulwich, *J. Bacteriol.* **80**:1200, 1964). However, the amount of phosphoric acid required for neutralization is such that the final gel possesses an abnormally high salt content (35 to 40 g of phosphate per liter).

The possibility arose that the essential features of the two preceding methods could be combined to facilitate the production of more versatile silica gel media. This communication describes the preparation and gelation properties of silicate solutions varying in pH between silicic acid ($pH < 3.0$) and potassium silicate ($pH > 12.5$).

A potassium silicate solution prepared as described by Funk and Krulwich (1964) was passed through a 50×3 cm column of H-saturated cation exchange resin (Dowex 50 W-X8). The first 25 to 30 ml of eluate was discarded and the silicic acid was then collected with constant stirring in beakers containing potassium silicate to obtain a set of silicate solutions in the desired pH range. The solutions were autoclaved (15 min at 120 C) immediately after preparation. Only silicate solutions of $pH \geq 8.0$ were stable to autoclaving, and these formed gels on addition of sufficient phosphoric acid to effect neutralization. The

amount of 6 N phosphoric acid necessary for gelation of 10 ml of potassium silicate ($pH > 12.5$) was about 4 ml; in contrast, comparable silicate solutions of pH 9 to 10 required <0.5 ml. The rate of gelation after neutralization and the firmness of the gels increased with increasing pH of the silicate solution and increasing silicate concentration of the final nutrient-amended medium. Gelation rate was particularly sensitive to temperature; for example, poured plates of a pH 9.0 silicate solution neutralized with phosphoric acid required 1 to 2 days to gel at room temperature, but <3 hr at 60 C.

Batch equilibration can be used instead of column chromatography to prepare the silicate solutions. Potassium silicate solution is equilibrated with sufficient H-saturated resin to produce a silicate solution of pH 6 to 8, the resin is removed by filtration, and the silicate solution is then titrated with potassium silicate to the appropriate pH . The exact procedure for media preparation will depend on the desired chemical and physical properties of the final medium. The following procedure is amenable to bulk preparation of media for pour, spread, and streak purposes, since gelation does not occur for 2 to 3 hr after neutralization. A sterile pH 10.0 silicate solution (100 ml) is mixed aseptically with sterile, double-strength nutrient solution (100 ml) and sterile 6 N phosphoric acid (4 to 5 ml); the resultant medium is sterile, clear, and firm, has negligible syneresis water and a phosphate content of <5 g/liter. Substitution of a pH 8.0 silicate solution for the pH 10.0 solution will lower the phosphate content to <1.5 g/liter, but the medium will be softer and will require heat to effect gelation within a reasonable time period. Poured plates of this medium will gel within 4 hr when placed in an oven at 60 C.

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