Title
USE OF ALUMINA COLUMNS IN CHROMATOGRAPHY

Authors
Kritchevsky, D.
Jorgensen, E.C.

Publication Date
1950-05-22

UNIVERSITY OF CALIFORNIA

Radiation Laboratory

BERKELEY, CALIFORNIA

DISCLAIMER

COPY 2

UCRL 712

Unclassified Distribution

UNIVERSITY OF CALIFORNIA

Radiation Laboratory

Contract No. W-7405-eng-48

USE OF ALUMINA COLUMNS IN CHROMATOGRAPHY

David Kritchevsky and Eugene C. Jorgensen

May 22, 1950

Berkeley, California

INSTALLATION:	No. of Copies
Argonne National Laboratory	8
Armed Forces Special Weapons Project	1
Atomic Energy Commission, Washington	2
Battelle Memorial Institute	1
Brush Beryllium Company	1
Brookhaven National Laboratory	8
Bureau of Medicine and Surgery	1
Bureau of Ships	1
Carbide and Carbon Chemicals Div., Union Carbide and Carbon Corp. (K-25 Plant)	4
Carbide and Carbon Chemicals Div., Union Carbide and Carbon Corp. (Y-12 Plant)	4
Chicago Operations Office	1
Cleveland Area Office, AEC	1
Columbia University (J. R. Dunning)	2
Columbia University (G. Failla)	1
Dow Chemical Company	1
H. K. Ferguson Company	1
General Electric Company, Richland	3
Harshaw Chemical Corporation	1
Idaho Operations Office	1
Iowa State College	2
Kansas City Operations Branch	1
Kellex Corporation	2
Knolls Atomic Power Laboratory	4
Los Alamos Scientific Laboratory	3
Mallinckrodt Chemical Works	1
Massachusetts Institute of Technology (A. Gaudin)	1
Massachusetts Institute of Technology (A. R. Kaufmann)	1
Mound Laboratory	3
National Advisory Committee for Aeronautics	2
National Bureau of Standards	2
Naval Radiological Defense Laboratory	2
New Brunswick Laboratory	1
New York Operations Office	5
North American Aviation, Inc.	1
Oak Ridge National Laboratory	8
Patent Branch, Washington	1
Rand Corporation	1
Sandia Laboratory	1
Santa Fe Operations Office	1
Sylvania Electric Products, Inc.	1
Technical Information Division, Oak Ridge	15
USAF, Air Surgeon (R. H. Blount)	1
USAF, Director of Armament (C. I. Browne)	1
USAF, Director of Plans and Operations (R. L. Applegate)	1
USAF, Director of Research and Development (F. W. Bruner, and R. J. Mason)	2
USAF, Eglin Air Force Base (A. C. Field)	1

INSTALLATION:	No. of Copies
USAF, Kirtland Air Force Base (M. F. Cooper)	1
USAF, Maxwell Air Force Base (F. N. Moyers)	1
USAF, NEPA Office	2
USAF, Office of Atomic Energy (A. A. Fickel, H. C. Donnelly)	2
USAF, Offutt Air Force Base (H. R. Sullivan, Jr.)	1
USAF, Wright-Patterson Air Force Base (Rodney Nudenberg)	1
U. S. Army, Atomic Energy Branch (A. W. Betts)	1
U. S. Army, Army Field Forces (James Kerr)	1
U. S. Army, Commanding General, Chemical Corps Technical Command (J. A. MacLaughlin thru Mrs. G. Benjamin)	1
U. S. Army, Chief of Ordnance (A. R. Del Campo)	1
U. S. Army, Commanding Officer Watertown Arsenal (C. H. Deitrick)	1
U. S. Army, Director of Operations Research (Ellis Johnson)	1
U. S. Army, Office of Engineers (Allen O'Leary)	1
U. S. Army, Office of the Chief Signal Officer (Curtis T. Clayton thru G. C. Hunt)	1
U. S. Army, Office of the Surgeon General (W. S. Stone)	1
U. S. Geological Survey (T. B. Nolan)	1
U. S. Public Health Service	1
University of California at Los Angeles	1
University of California Radiation Laboratory	5
University of Rochester	2
University of Washington	1
Western Reserve University	2
Westinghouse Electric Company	4
Univ. of Rochester (R. E. Marshak)	1
Total	143

Information Division
Radiation Laboratory
University of California
Berkeley, California

USE OF ALUMINA COLUMNS IN CHROMATOGRAPHY

David Kritchevsky and Eugene C. Jorgensen

Radiation Laboratory and Department of Chemistry,
University of California, Berkeley*

May 22, 1950

ABSTRACT

A general technique for preparation, grading and use of alumina in chromatography is outlined.

(*) The work described in this paper was sponsored by the Atomic Energy Commission.

USE OF ALUMINA COLUMNS IN CHROMATOGRAPHY

David Kritchevsky and Eugene C. Jorgensen

Radiation Laboratory and Department of Chemistry,
University of California, Berkeley, Calif. *

The theory and general practice of adsorption chromatography has been thoroughly covered in several books (1,2). The exact procedure which is to be followed in alumina chromatography has never been outlined. For persons beginning to use alumina columns, the procedure to be followed is usually arrived at after much trial and error and considerable expenditure of time. This report, while it contains nothing new, is an attempt to systematize the most popular current techniques for preparation of alumina, testing and chromatographic procedures.

A. Preparation and Grading of Alumina: - The adsorptive capacity of alumina increases as the water adsorbed decreases. Thus, the adsorptive capacity can be increased by heating the material to high temperatures. The following conditions have been developed to give alumina of varying adsorptive strengths (3).

I - Heat small portions to glowing in an iron pot. Shake vigorously throughout heating. Cool in desiccator.

(*) The work described in this paper was sponsored by the Atomic Energy Commission.

(1) Strain, "Chromatographic Adsorption Analysis," Interscience Publishers, Inc., New York, New York, 1945.

(2) Zechmeister and Cholnoky, "Principles and Practice of Chromatography," John Wiley and Sons, Inc., New York, New York, 1943.

(3) Brockmann and Schodder, Ber., 74, 73 (1941).

II - Cover bottom of iron vessel with alumina. Heat 4-6 hours with shaking. Cool. Expose to air, in a thin layer, for 30 minutes.

III - As II. Expose to air for an hour or shake in moist air.

IV and V - As II. Shake in moist air.

It might be pointed out that commercially available alumina (Merck or Alcoa) is entirely satisfactory, so long as the material is of uniform adsorptive strength. Generally, aluminas of strength II-III are adequate for most work.

The alumina is graded by measuring the strength of adsorption of various dyes. The dye solutions are prepared by dissolving small amounts of the dye in ether-benzene 4:1. A column of alumina 10 x 1.5 cm. is used. Ten ml. of the dye solution is added and eluted with 20 ml. of the solvent mixture. The following table summarizes the results:

Activity	I	II		III		IV		V
Solution No.	1	1	2	2	3	3	4	5
Position on Column	A B		C A	A	D C	D	E D	F E
Filtrate		B		A		C		

A = p-Methoxyazobenzene

B = Azobenzene

C = Sudan Yellow

D = Sudan Red

E = p-Aminoazobenzene

F = p-Hydroxyazobenzene

B. Size of Column: - Generally, a column which has a length to width ratio of 15:1 is desirable. The weight ratio of alumina to substance to be chromatographed should be about 30:1. These ratios are empirical.

C. Column Preparation : - It is desirable to construct glass columns having a stopcock at the bottom for ease in packing and to enable the operator to stop the flow of eluent whenever necessary.

A convenient manner of preparation of the column is the following: A wad of glass wool, which has previously been soaked in the first solvent to be used, is packed into the bottom of the column. The solvent is poured in to more than the desired height and the dry alumina is slowly poured in. During alumina addition, the column is constantly tapped (a rubber stopper at the end of a rod is very convenient) to insure uniform settling of the alumina. After the required amount of alumina has been added another wad of glass wool or cotton is used to cover the alumina; this helps to keep the column moist.

D. Table of Solvent Strengths: - In order of decreasing eluting power (4).

Water
Methanol
Ethanol
Propanol
Acetone
Ethyl Acetate
Ethyl Ether
Chloroform
Methylene Chloride
Benzene
Toluene
Trichloroethylene
Carbon Tetrachloride
Cyclohexane
Hexane

E. Useful Procedure: - The substance to be chromatographed is dissolved in a small amount of the initial solvent (the least polar is preferable), placed on the column and eluted. The common solvents are petroleum ether, hexane, benzene, ethyl ether, ethyl acetate, chloroform and methanol. These should be prepared in pure,

---

(4) Claesson, Ark. Kem. Mineral. och. Geol., 23A, No. 1, 1946.

anhydrous state before using. Generally, one does not change abruptly from one solvent to another, but proceeds gradually through solutions of the two, e.g., petroleum ether, then petroluem ether-benzene 9:1, 8:2.....1:1....benzene, benzene-ether 4:1, etc. The material being chromatographed and the amount being eluted dictate which mixtures to use.

The size of cuts may vary with each experiment; usually with 0.5-2.0 gm. of substance, it is advisable to take 25-50 ml. cuts into tared flasks. After evaporation of the solvent the flask is weighed. When the amount eluted falls below 10% of the amount placed on the column it is customary to change solvents.

In a series of cuts having the same melting point, if a mixed melting point of the first and last cut shows no depression, the fractions may be combined. In some instances it is necessary to rechromatograph a fraction. In such a case, the cuts and solvent ratios must be more closely regulated.

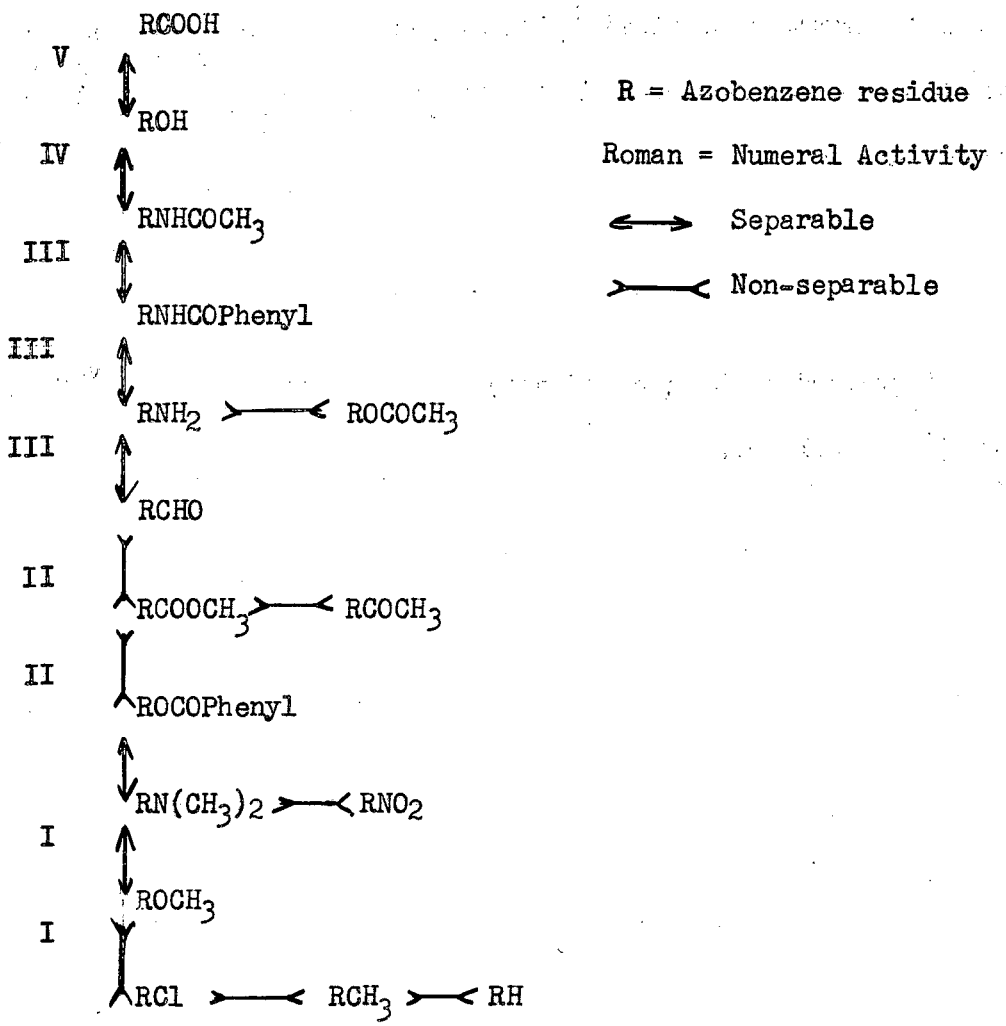
Pressure (gentle) may be used to hasten the flow, but it must be carefully regulated and used throughout.

The column must never be allowed to become dry in the course of any single experiment.

F. Separation of Organic Compounds: - The following scheme is indicative of the type of separation one may expect, considering the functional groups involved and the activity of the alumina used (5).

---

(5) Brockmann, Angew. Chem., 59, 199 (1947).



G. Possible Applications to Paper: - The preceding techniques might be easily adapted to a more rapid analytical method using alumina impregnated filter paper (6,7). Whatman No. 54 filter paper is cut into strips and dipped into a solution of aluminum sulfate, 65 g/L, and drained. The paper is then soaked in 2 N aqueous ammonia and washed under a running tap for 5 hours. It may be dried

---

(6) Datta, Overell and Stack-Dunne, Nature, 164, 673 (1949).

(7) Datta and Overell, Biochem. J., 44, xliii (1949).

at room temperature or at 140° C; the adsorptive capacity is increased by storing over phosphorus pentoxide. The standard Brockmann dyes may be used to measure the adsorptive strength.

SUMMARY

A general technique for preparation, grading and use of alumina in chromatography is outlined.