Title
A SIMPLE AUTOMATIC VALVE FOR CONSTANT VOLUME COLLECTION IN COLUMN CHROMATOGRAPHY

Author
Nelson, Gary J.

Publication Date
1960-03-01

UCRL 9114

UNIVERSITY OF CALIFORNIA

Ernest O. Lawrence Radiation Laboratory

BERKELEY, CALIFORNIA

DISCLAIMER

For publication in Analytical Chemistry

UCRL-9114

UNIVERSITY OF CALIFORNIA

Lawrence Radiation Laboratory
Berkeley, California

Contract No. W-7405-eng-48

A SIMPLE AUTOMATIC VALVE FOR CONSTANT VOLUME COLLECTION IN COLUMN CHROMATOGRAPHY

Gary J. Nelson

March 1960

Short Communication

A Simple Automatic Valve for Constant Volume Collection in Column Chromatography*

By

Gary J. Nelson**

From the
Donner Laboratory of Biophysics
and
Medical Physics
University of California
Berkeley, California

In the course of work conducted on the chromatographic behavior of serum phospholipids on silicic acid columns[1,2] it was necessary to undertake extended chromatographic analysis with large columns and automatic fraction collectors. As has been mentioned by others[3,4] the standard methods of fraction cutting with automatic fraction collectors fail to give satisfactory results in column chromatography with organic solvents. In lipid chromatography a scheme that involves several changes in the elution solvents during a run is generally followed, and the number of drops per ml varies greatly over the entire elution scheme because of differences in the surface tension of the various solvents. Drop counting therefore will not yield fractions of constant volume. For essentially the same reason constant time interval fraction collecting is not practical, as the flow rate of the column cannot be maintained constant from one eluting solvent to another, or for that matter even in the period during which one solvent is eluting, and thus fractions of constant volume cannot be obtained.

The most obvious and simplest method of constant volume collecting when confronted with variable flow rates and variable drop size is automatic siphoning. This was tried and found unsatisfactory, for several reasons. First, the siphon available in this laboratory was not always dependable with various organic solvents.

The liquid level would reach the overflow tube, and instead of siphoning the solvent would slowly trickle down the tube. Second, when siphoning would occur, the amount left in the siphon varied between fractions and more so between solvents. Third, the amount remaining in a 10 ml siphon was often 0.5 ml or more, leading to considerable contamination between fractions. Fourth, the volume collected could be changed only by changing the siphon, and this was inconvenient during a run.

To overcome these difficulties a new type of automatic fraction cutter to be operated in conjunction with any automatic fraction collector was designed***. It operates with any solvent and delivers a constant volume regardless of flow rate of the column or surface tension of the eluent. The general principle is well known. It consists of a solenoid-operated valve which is activated by a photocell and relay. The operating scheme of the fraction cutter is shown in Figure 1.

A drop counting attachment was utilized with minor modifications for indexing the turntable. However, this valve system can be constructed and operated independently as long as a photocell and means of indexing the turntable are provided. The accuracy of the valve depends on the seal which a tapered Teflon plug, carefully ground and polished, makes with a ground glass seat of the same taper as

the plug. Details of construction are given in Figure 2. The valve stem is constructed entirely of Teflon and encloses a soft iron core which opens the valve when the coil is activated by the photocell. The spring is stainless steel and prevents the valve from lifting too high and obstructing the flow through the outlet. The eluent collecting tube is made of a constant bore glass tubing 1 cm O.D. diameter. Thus, the volume collected in each fraction is continuously variable simply by raising or lowering the position of the photocell along the tube. Figure 3 is an exploded photographic view of the components of the valve.

The sequence of operational events is as follows: When the meniscus reaches the level of the phototube, the light beam is interrupted. The turntable motor is activated and the turntable indexed, which takes 1.5 seconds in our instrument. Then the valve opens for 4 seconds, actuated by a time delay switch. Flow rates are adjusted so that complete drainage from the lower parts of the valve will occur between indexings of the turntable.

Gilson Medical Electronics, Madison, Wisconsin, manufactures a volumetric fraction collector, based on an analogous principle to that described here, which uses an all glass valve. However, their current model does not appear adequate for organic solvents because of leakage and evaporation problems. Microchemical Specialties, Berkeley, California, has also marketed an automatic valve but no test of its reliability or accuracy has been reported.

Factors which affect the volume collected are the length of time the valve is open, the flow rate of the column, the drain time of the collecting tube, the viscosity of the eluent and the retention of the eluent on the lower portion of the valve stem. Obviously, the valve must remain open long enough for the eluent collected to drain out. There will be some retention of eluent on the sides of the collection tube and, if the valve is operated as shown, solvent will continue to elute into the collector while the valve is open. This latter occurrence can be prevented by attaching a second identical valve to the top of the collector which is closed when the lower valve is open. Practically, this is not necessary when the flow rate is slow and valve is open for less than 5 seconds in which case the controlling factor becomes the drainage rate off the walls of the collecting tube. We have found that with a flow rate of 1 ml per minute, a time constant of 4 seconds for the valve to be open, and 10 ml collection volume, the variability in volume collected is less than one tenth of a milliliter per fraction, or one percent.

This design is quite flexible and can be scaled up or down to suit any given chromatographic requirement without difficulty. The unit described herein is adequate for volumes in range from 1 to 20 ml as it stands, or 0.5 ml to 100 ml by modification of the collection tube design. At present a microvalve to work in the 0.1 to 1.0 ml range is being designed.

Footnotes

* This work was supported in part by the United States Atomic Energy Commission.

** United States Public Health Service Postdoctoral Research Fellow of the National Heart Institute.

*** A commercial instrument enbodying this design should be available soon from Research Specialities, Inc., Richmond, California.

References

1. G. J. Nelson and N. K. Freeman, J. Biol. Chem., 234, 1375 (1959).

2. G. J. Nelson and N. K. Freeman, J. Biol. Chem., 235, 615 (1960).

3. N. S. Radin, in D. Glick, ed., Methods of Biochemical Analysis, Vol. 6, Interscience Publishers, Inc., New York, 1958, pp. 163-189.

4. W. H. Bancroft, Jr., Anal. Chem., 31, 1447 (1959).

Figure Captions

Figure 1. Circuitry and operating scheme of the solenoid valve in conjunction with an automatic fraction collector and modified drop counter. The only significant additions to the original circuit are the time delay switch, A, and coil, B.

Figure 2. Working drawing of the solenoid valve. The ground glass valve seat is indicated at A, the Teflon plug at B, and the soft iron core at C.

Figure 3. An exploded photographic view of the components of the solenoid valve.

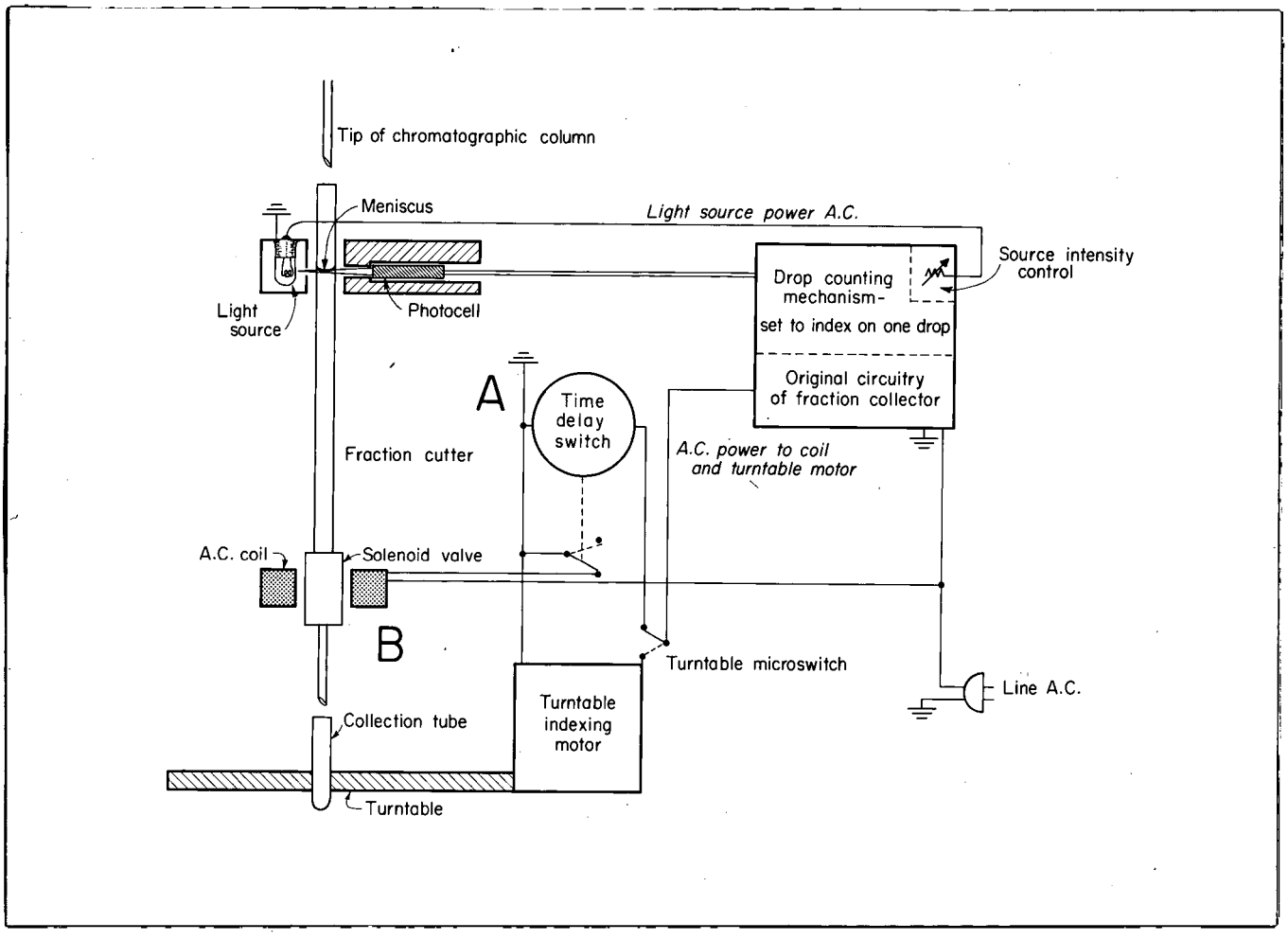
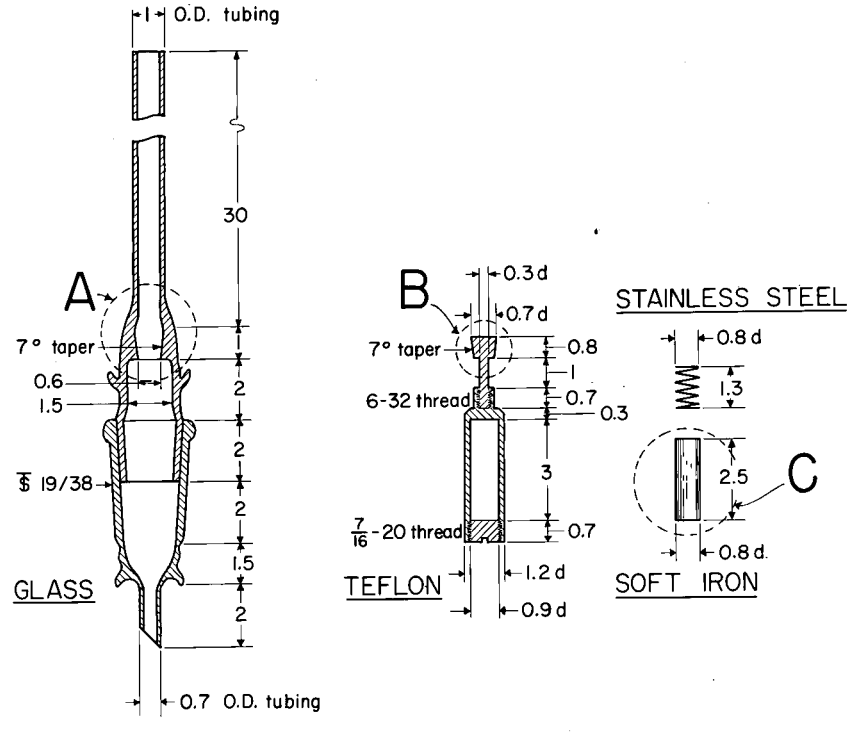

Tip of chromatographic column
Meniscus
Light source power A.C.
Source intensity control
Drop counting mechanism-
set to index on one drop
Original circuitry of fraction collector
Light source
Photocell
A
Time delay switch
Fraction cutter
A.C. power to coil and turntable motor
A.C. coil
Solenoid valve
B
Turntable microswitch
Line A.C.
Turntable indexing motor
Collection tube
Turntable

Fig. 1.



Note : All measurements are in centimeters

Fig. 2

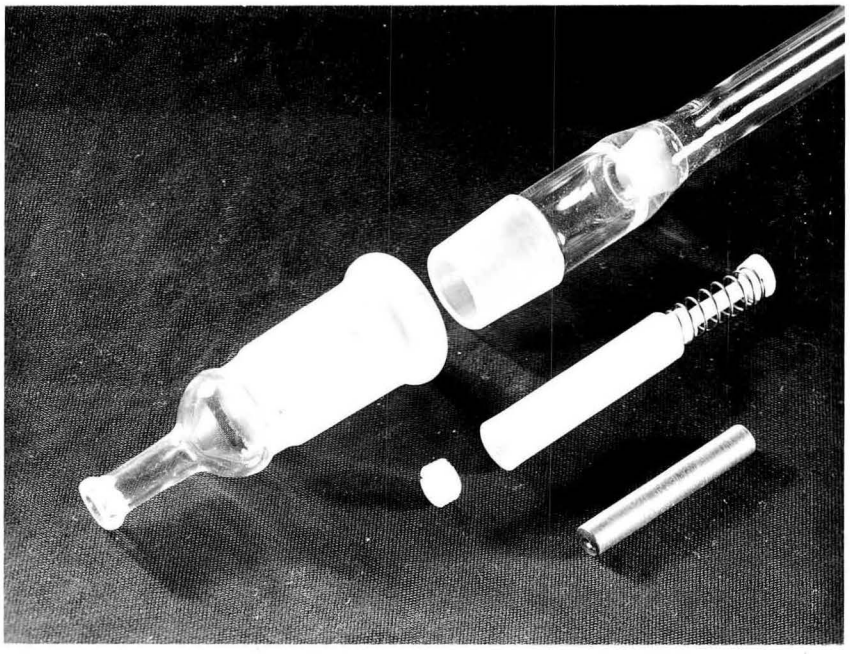


Fig. 3.