

## Fraction Collector Modifications

Eugene A. Talley, Eastern Regional Research Laboratory, Agricultural Research Service, U. S. Department of Agriculture, Philadelphia 18, Pa.

THE TWO MODIFICATIONS in fraction collecting equipment described are useful during the ion exchange chromatography of amino acids (3).

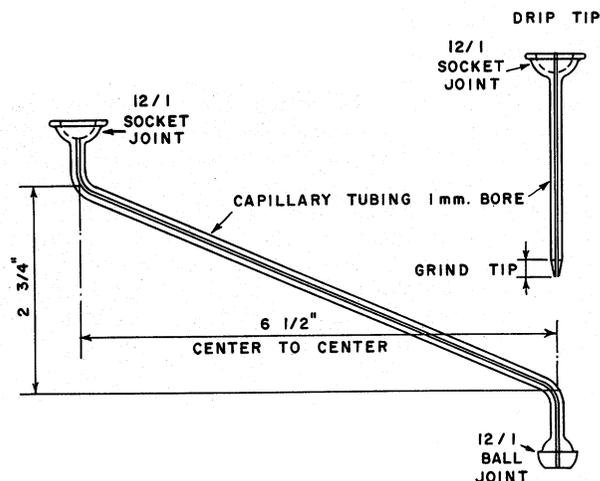
A capillary inflow arm (Figure 1) has been substituted for the wider tube frequently employed with drop-counting fraction collectors, such as the Technicon photoelectric fraction collector (the design shown in the figure is for the Technicon apparatus). The columns were fitted with No. 12 ball joints. The tubing between these joints and the resin support was of larger than capillary size, but should be reasonably short; separation of peaks on the effluent curve was improved if this tubing was air filled rather than liquid filled during elution.

The use of this capillary arm minimizes the uptake of ninhydrin-positive materials from the laboratory air and it improves the uniformity of dropping of the eluate. The bevel on the dropping tip can be omitted for large drops. (One of the reviewers suggests that the tip may be beveled by drawing it out while blowing into the capillary, thus preventing a significant reduction in bore while reducing the outside diameter of the tube.) With large diameter columns, two of the connecting tubes can be used in series, permitting a column to be disconnected for regeneration without changing the position of the column.

The lower joint can be held by a pinch clamp. The upper joint, however, must be free to rotate in order to permit the shift from one row of the collector to the next. Low viscosity grease and a screw-type clamp (No. C5155, Scientific Glass Apparatus Co.) with a rubber ring for cushioning, were used. Sections of split rubber tubing or tape may be necessary to make the clamps fit the capillary tubing.

The second modification is the use of an extra counter to effect a change in the temperature of the water bath used to control the temperature of the columns. In using the Moore and Stein ion exchange procedure for the determination of amino acids (3), the author found that increasing the flow rate considerably does not appreciably broaden the effluent peaks obtained. Recent developments (2, 4) also use increased flow rates. In the case of the 100-cm. columns, if the flow rate is doubled, about 200 fractions are collected in 24 hours, which is about the

Figure 1. Capillary inflow arm for use with the Technicon photoelectric fraction collector



capacity of the turntables of the fraction collectors used in this work. This is a convenient rate, except that the second temperature change is required at a time when one is normally absent from the laboratory.

An auxiliary counter has been used to switch the control contacts of the relay of the heating bath from one thermostat to another at any given fraction number. This counter switch is a duplicate of the Microflex reset counter used in the drop-count attachment of the Technicon fraction collector. Thus it also may be used as a spare counter in an emergency. It is Eagle Signal Corp. No. HZ40A6 with the standard operation sequence arrangement A242. It may be obtained as the open type for installation in a cabinet or as the enclosed type for use outside. This counter as obtained from the manufacturer contains a jumper across contacts L2 and 3 of its terminal block. This jumper must be removed when the counter is to be used in the Technicon drop-count attachment (the jumper across 3 and 4 must also be removed when the counter is used as a replacement drop counter) or for this application.

The count coil of the counter must be supplied with a suitable electrical pulse each time a tube is changed, if it is to be used to switch thermostats or other electrically operated apparatus when a definite number of tubes is filled. The Technicon drop-counting attachment is fitted with a two-wire Twistlock midget flush base (the direct reading counter outlet shown in the schematic diagram of the Technicon drop-count control) which furnishes a pulse every time a

drop is registered. By changing the internal wiring slightly, this can be used to furnish a pulse each time a tube is changed. Two wires (one to the multiplier socket and the other to the photoelectric relay chassis) are attached to one terminal of the direct reading counter outlet. These wires are removed and connected together. The vacant terminal is then connected to terminal 1 of the counter in the drop-count attachment. This can be done rather simply by the use of a jumper from the vacated outlet terminal to contact 24 on the terminal block in the bottom of the drop-counting cabinet. After these changes are made, the multiplier socket is still operative, but the direct reading counter outlet now furnishes an electrical pulse only when the tubes are changed under the dropping tip instead of each time a drop is registered. Then a midget size two wire Twistlock cap such as Hubbell No. 7428, can be installed on a cord and used to connect to the count coil of the extra counter when it is being used.

Several changes are made also in the auxiliary counter when it is used with the Technicon drop counter. For this application, it is safer to separate the clutch coil wiring from that of the count coil winding as described below. Then the clutch coil can be plugged directly into the line without danger of a short circuit. As furnished, both the count and the clutch coils are connected to terminal L1 of the terminal block of the counter. If the jumper between terminals A and B is removed, the wire from the count coil to L1 can be connected to terminal B. Then the switch inside the counter and connected across

terminals *B* and *L2* will disconnect the count coil when the count is completed. The count coil is connected to the source of electric pulses (the direct reading counter outlet of the modified Technicon drop count attachment) through terminals *M* and *L2*. The clutch coil is connected through terminals *L1* and *A* to the power line in such a way that it may be disconnected easily. The switch in the counter connected across terminals 4 and 2 is closed during the counting period and opens when the count is completed. The switch connected across terminals 1 and 3 has the opposite action. The items to be operated are connected accordingly. For example, for switching from 50° to 75° C. after 75 tubes are filled, the counter is set for 75 (the count will be correct as set for this application), the 50° C. thermostat is connected to terminal 2 and the 75° C. thermostat to

terminal 1. Terminals 3 and 4, connected by a jumper, are connected to one relay control terminal. The other side of the relay control line is connected to each of the thermostats.

The counter as furnished is conditioned to reset itself on low voltage. This means that the counter will reset itself to zero count if there is a power interruption and then the switching action will take place later than intended when action resumes. The counter is easily conditioned for non-reset on low voltage (*I*). When this change is made, the counter will not reset on power interruption and change the effective count. When starting, the clutch coil is energized only while the counter is being set and until the count is to begin and then is de-energized until one wishes to reset the counter again to zero. If the counter is conditioned for reset on low voltage, the

clutch coil must be energized throughout the entire counting period.

To condition the counter for use as a replacement for the counter in the Technicon drop-counter attachment, the entire procedure described in the last paragraph above and the first half of the previous paragraph is reversed.

#### LITERATURE CITED

- (1) Eagle Signal Corp., Moline, Ill., Microflex Bull. 110C, 720. (Direct bulletin requests to E. F. Havey.)
- (2) Hamilton, P. B., ANAL. CHEM. 30, 914-19 (1958).
- (3) Moore, S., Stein, W. H., *J. Biol. Chem.* 192, 663-81 (1951).
- (4) Spackman, D. H., Stein, W. H., Moore, S., *Federation Proc.* 15, 358 (1956).

MENTION of trade names or companies in this paper does not imply endorsement by the Department of Agriculture over others of similar nature not mentioned.