Separation Science and Technology
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713708471

Programmed Multiple Development: The Solvent Advance and Removal Mechanisms, Isolated and Compared

John A. Perry[a]; Thomas H. Jupille[b]; Anni Curtice[b]
[a] CONSULTANT, CHICAGO, ILLINOIS [b] REGIS CHEMICAL COMPANY, MORTON GROVE, ILLINOIS

To cite this Article Perry, John A. , Jupille, Thomas H. and Curtice, Anni(1975) 'Programmed Multiple Development: The Solvent Advance and Removal Mechanisms, Isolated and Compared', Separation Science and Technology, 10: 5, 571 — 591
To link to this Article: DOI: 10.1080/00372367508058041
URL: http://dx.doi.org/10.1080/00372367508058041

SEPARATION SCIENCE, 10(5), pp. 571–591, 1975

Programmed Multiple Development: The Solvent Advance and Removal Mechanisms, Isolated and Compared

JOHN A. PERRY

CONSULTANT
CHICAGO, ILLINOIS 60616

THOMAS H. JUPILLE* and ANNI CURTICE

REGIS CHEMICAL COMPANY
MORTON GROVE, ILLINOIS 60053

Abstract

Throughout each programmed multiple development (PMD) in thin-layer chromatography (TLC), the thin-layer plate remains in contact with the solvent. An important result is a spot reconcentration that narrows spots of low as well as high R_F. Spot reconcentration occurs as an interaction between the spot molecules and the solvent front, during both solvent advance and solvent removal. When solvent is removed by heating in PMD, two separate mechanisms for spot reconcentration exist: (a) solvent removal by heating, and (b) solvent advance that follows solvent removal by heating. These mechanisms have been experimentally isolated and studied quantitatively. A chromatographic quality index $Q_{MD} = RR_W$ was used for expression of the results. Here R is resolution—the center-to-center separation of the spots divided by the average width of the spots—and R_W is the maximum ratio of the spot top-to-bottom widths in the chromatogram. To make the measurements of the developed spots more precise and clear, all spots were formed into rings before development. The measurements show that if solvent is removed conventionally—by air-drying the plate away from the solvent reservoir between developments—it matters little whether the successive solvent advances are uniform ($Q_{MD} = 2.1$) or successively more extended ($Q_{MD} = 1.5$). Both compare with straight PMD, for which $Q_{MD} = 10.0$. The chromatographic quality resulting from isolated solvent advances that follow solvent removal by heating is best for gentler, more prolonged heating ($Q_{MD} = 3.0$), but better for strong, brief heating ($Q_{MD} = 2.3$) than for no heat-

*To whom requests for reprints should be sent.

ing at all ($Q_{MD} = 1.5$). The chromatographic quality resulting from solvent removal is better than that resulting from solvent advance whether the heating is—as is preferable—gentler and more prolonged ($Q_{MD\text{-removal}} = 4.3$, $Q_{MD\text{-advance}} = 3.0$), or strong and brief ($Q_{MD\text{-removal}} = 3.4$, $Q_{MD\text{-advance}} = 2.3$). The full chromatographic quality of straight PMD ($Q_{MD} = 10.0$) is obtained because the solvent advance and solvent removal mechanisms complement each other and are supplemented by preheat—an initial, brief continuous development at the origin.

Programmed multiple development (PMD) (*1–9*) is a form of thin-layer chromatography (TLC) (*10, 11*). In PMD, the thin-layer plate remains at all times in contact with the solvent. Therefore throughout each PMD the solvent moves by capillary action toward the solvent front. The location of the solvent front is governed by the rate of solvent evaporation.

The rate of solvent evaporation is varied automatically, in accordance with the PMD program chosen by the operator. In consequence, the solvent front is caused to move up and down the thin layer. The solvent front moves farther up the plate with each solvent advance, but returns to the point of spot deposition with each solvent removal.

Each time the solvent front moves up or down across a spot, the spot becomes reconcentrated. During solvent advance, molecules behind the front move toward molecules not yet reached. During solvent removal, molecules behind the front move toward molecules that have been deposited by the receding solvent front. Thus each spot is reconcentrated twice per PMD cycle.

For solvent advance under isothermal conditions, spot reconcentration is well described by the Jupille equation (*4, 6*):

$$X_f = (1 - R_F)^n X_i$$

Where X_i is the initial distance, taken along the line of chromatographic development, between two of the same R_F molecules that act as statistical aggregates, n is the number of solvent advances, R_F is the spot-to-solvent velocity ratio (usually measured as the spot migration distance divided by the origin-to-front solvent migration distance), and X_f is the final distance between these molecules.

One form of multiple development in conventional TLC is called undimensional chromatography (UMC) (*12*). In UMC, a given thin-layer plate is repeatedly developed in the same direction for the same distance. The Jupille equation applies to both UMC and PMD isothermal solvent advance, given that the PMD solvent removal is instantaneous.

If PMD solvent removal is not instantaneous but lasts a fixed time—

perhaps a minute—with each cycle, and is done under approximately isothermal conditions at ambient temperature, then the resultant spot reconcentration by solvent removal has been shown to be about as effective as that of solvent advance for a spot of R_F 0.4 (*6*).

In PMD the solvent is removed from the plate by a gas sweep, or IR radiation, or both. Therefore the thin-layer plate temperature during solvent removal can be below, at, or above ambient.

If the solvent is removed solely by heating the plate with the IR radiator that is part of the PMD developer, the plate remains cool to the touch where wet, but becomes warm or hot where dry.

At the end of the time allotted to solvent removal by heating, the power to the IR radiator is usually (though not necessarily) turned off. The temperatures of the radiator and radiator housing then slowly return toward ambient. As the thin-layer plate cools, the solvent front again begins to advance into the thin layer. The thin-layer plate undergoes marked local cooling at the advancing solvent front as the solvent that has reached the front evaporates and carries away the heat of evaporation.

The degree of spot reconcentration that is associated with solvent removal by heating differs markedly from that found with isothermal, ambient-temperature conditions.

We have isolated, demonstrated, and measured examples of two spot reconcentration mechanisms associated with solvent removal by heating. One of these two mechanisms involves solvent advance into a plate that is slowly cooling after having just been heated sufficiently, in the PMD developer environment, to drive the solvent front back to the spot origin. The other involves solvent removal, by heating, from a plate that is cool when solvent removal begins. We also isolate and demonstrate, but do not measure, a third mechanism, called preheat, that involves a brief, powerful continuous development onto a front at or near the origin.

This paper reports the isolation and study of these spot reconcentration mechanisms.

EXPERIMENTAL

Equipment

Thin-layer Plates. The thin layer was Silica Gel G. The thin-layer plates used were precoated, obtained from Camag, New Berlin, Wisconsin 53151. The 5 cm × 10 cm plates used in this work were cut from the 20 cm × 20 cm plates received.

Pipettes. Camag 5 μl pipettes were used. These bear markings showing 1 μl volumes up to 5 μl.

Sample. The spots were made from Baker TLC Reagent, a solution in benzene of Butter Yellow, Sudan Red, and Indophenol Blue. Benzene development of these dyes on Silica Gel G yields approximately the following R_F's: Butter Yellow, 0.40; Sudan Red, 0.15; and Indophenol Blue, 0.08.

Solvent. Benzene, reagent grade.

PMD. The PMD Model 2000 Programmer and Model 222 Developer, both obtainable from the Regis Chemical Co., Morton Grove, Illinois 60053, were used for the PMD developments.

Procedure

Plate Preparation. Channels I mm wide were cut from the thin layer parallel to each 10 cm edge and about 5 mm in. For development, a 3-mm-thick, U-shaped, Teflon spacer along the edges and top of the plate separated the thin-layer plate from a glass facing plate, also 5 cm × 10 cm. The three were clamped together and the open bottom set into the solvent reservoir.

Spotting. On spot deposition, all spots were carefully formed into rings as follows. One microliter of the dye solution was drawn into the 5-μl pipette. This was not then spotted, but four more microliters of (reagent-grade) methanol were drawn into the same pipette, bringing the total liquid volume in the pipette to 5 μl. The pipette was then inverted and the spot applied. Thus, in one continuous operation, 1 μl of the dye solution was immediately followed into the thin layer by 4 μl of methanol. The pipette was then again filled with another 5 μl of methanol, which was then applied with care to the center of the dye spot. All the methanol was then removed from the thin layer by a stream of nitrogen applied for a full minute.

On each plate, one such spot, centered laterally on the plate, was applied.

Development Conditions

PMD Model

As the point of departure, the multiple developments were based on a 5-cycle Mode 1 PMD with 100-sec unit times for solvent advance and removal segments.

In such a PMD, the successive solvent advance times would be 100, 200, 300, 400, and 500 sec.

The solvent removal times would depend on the unit time—which was set at 100 sec—and on whether the removal time was fixed or scheduled.

In fixed time, each solvent removal would take 100 sec. For these, the power was set at 10. This power setting yields 100% of the maximum power output from the heaters.

In scheduled time in Mode 1 with a 100-sec unit time, the successive solvent removal times would be 100, 200, 300, 400, and 500 sec. For these, the power was set at 4. This power setting yields 20% of maximum power. When scheduled times were used in this study, the PAUSE was also used.

When the PMD Programmer PAUSE button is pushed, the programmer completes that cycle but interrupts the program on completion of solvent removal and applies INTERIM power if it is called for. In this study the INTERIM power was set at 5. This power setting yields 50% of maximum power. PAUSE is terminated manually by pushing the PAUSE button a second time.

Thus the scheduled-time solvent removal in this study called for 20% power for periods that increased with each cycle, followed automatically by 50% power until the solvent front reached the origin. The periods at 50% decreased as the 20% periods lengthened. These scheduled-time solvent removals were terminated manually.

PMD Fixed-Time Model, Demonstrated

Figure 1 shows a plate developed by an uninterrupted, normal PMD program corresponding to the model, with fixed time. The origin was placed 2.5 cm from the plate edge—the normal PMD distance.

Solvent Advances, Isolated

The isolated solvent advances were made under three preexisting conditions. These conditions corresponded to (a) conventional solvent removal and to (b) PMD fixed and (c) PMD scheduled solvent removals.

Conventional solvent removal means that immediately following each solvent advance the thin-layer plate was removed from the solvent trough, disassembled from its spacer and facing plate, and dried in open air. The plate assembly and the developer remained at ambient temperature before and during the solvent advance.

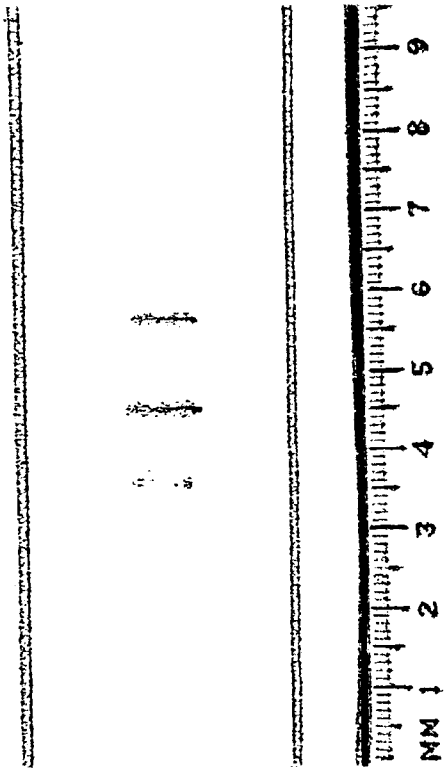


FIG. 1. A normal PMD. Five cycles, 100-sec unit times in Mode 1, fixed-time solvent removal.

Preconditions (b) and (c) involved heating. The heatings closely simulated the relevant solvent removal that would have preceded the solvent advances. The thin-layer plate assembly was held at the normal distance from the radiator, above the trough support, and in the normal orientation—the back of the thin-layer plate receiving radiation directly from the radiator. In short, the only difference was that the plate assembly was held outside the solvent trough during the heating. At the end of the indicated time, the plate assembly was placed into the solvent trough for the relevant solvent advance time.

Precondition (a): Conventional Solvent Removal. Two plates were made with only conventional solvent removal preceding the solvent

advances. One was developed by five UMC solvent advances, each lasting 500 sec; this is shown in Fig. 2.

The other was developed with the PMD scheduling already described, namely, by successive solvent advances lasting, in turn, 100, 200, 300, 400, and 500 sec; this is shown in Fig. 3.

Precondition (b): PMD Solvent Removal, Fixed Time. Immediately preceding each solvent advance, the dry plate (in its assembly) and the developer were heated at 100% of available power for 100 sec. After each solvent advance, the plate was dried by conventional solvent removal.

Three plates were made under precondition (b) with solvent advances timed according to the PMD Model described earlier. These plates

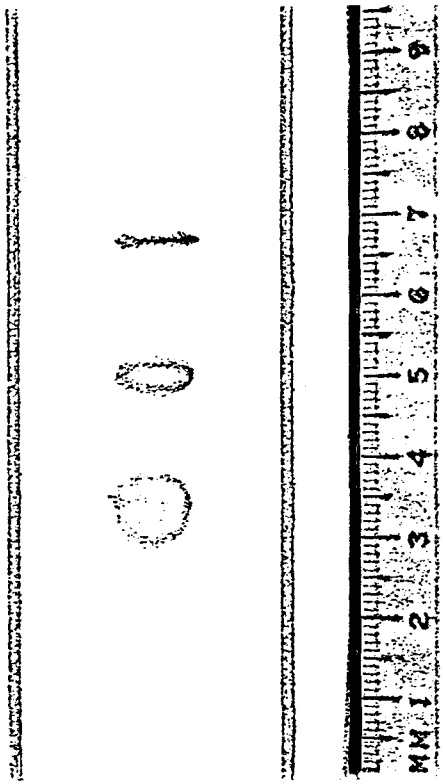


FIG. 2. UMC. Five 500-sec solvent advances.

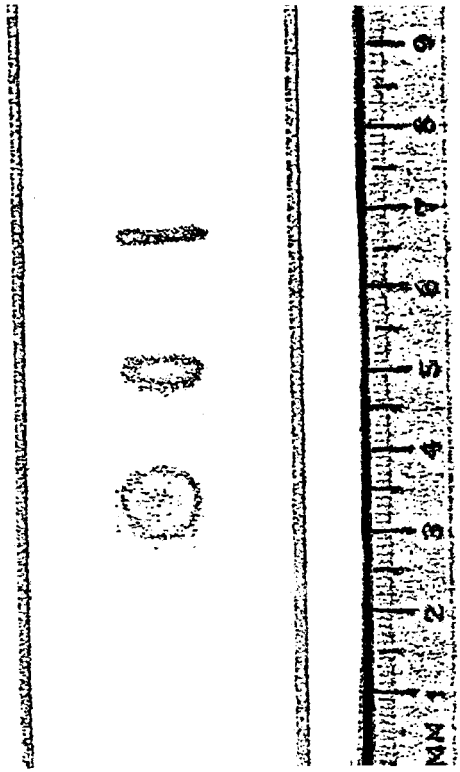


FIG. 3. Five PMD-scheduled solvent advances corresponding to Mode 1, 100-sec unit time. Conventional solvent removal between advances.

differed in the distance of the origin from the edge of the plate: Figure 4a, 2.5 cm; Figure 4b, 3.0 cm; and Figure 4c, 3.5 cm.

Precondition (c): PMD Solvent Removal, Scheduled Time. One plate, shown in Fig. 5, was made this way. Immediately preceding each solvent advance, the dry plate (in its assembly) and the developer were heated at 20% of available power for the times indicated in Table 1 and then at 50% of available power for the additional times indicated. After each solvent advance, the plate was dried by conventional solvent removal.

Solvent Removals, Isolated

Each isolated solvent removal by heat followed a solvent advance made under strictly isothermal, ambient-temperature conditions.

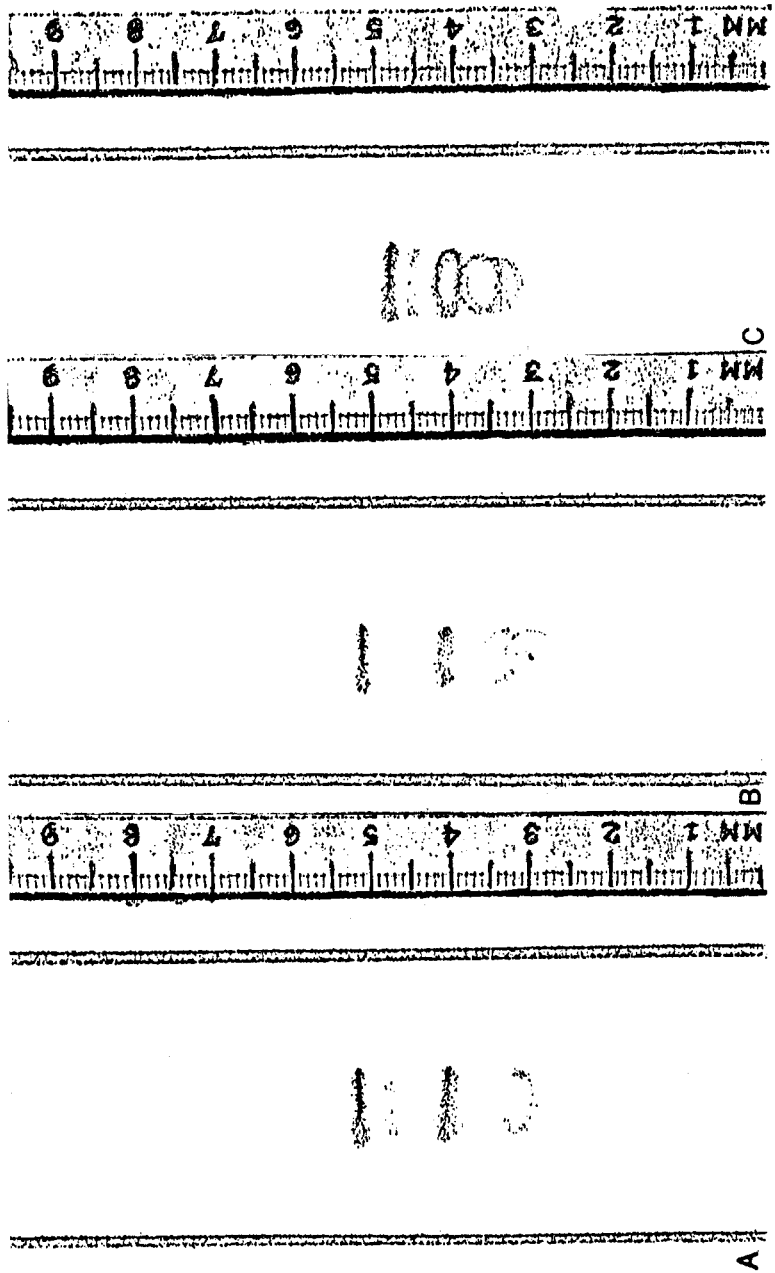


FIG. 4. Isolated PMD solvent advances corresponding to those following solvent removal by fixed-time heating. Origin from plate edge: 4a, 2.5 cm; 4b, 3.0 cm; 4c, 3.5 cm.

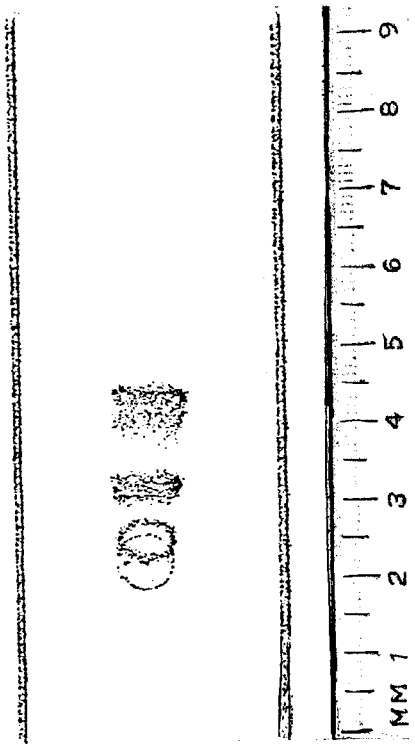


FIG. 5. Isolated PMD solvent advances corresponding to those following solvent removal by scheduled-time heating.

TABLE 1

Cycle	Seconds duration	
	20%	50%
1	100	120
2	200	60
3	300	45
4	400	30
5	500	15

Thus no solvent advance was started until the developer had cooled essentially to room temperature from the heating of the previous solvent removal. Usually, 30 min or more was allowed for such cooling. (Two developers were employed, one cooling while the other was being used.) Also, the thin-layer plate and its assembly were cool at the start of each solvent advance.

After each isolated solvent removal by heat, the thin-layer plate was removed from the trough and from its assembly, and set aside to cool.

The solvent removals were carried out as described for the PMD Model.

The plates, Figs. 6 and 7, showing the solvent removal studies were developed with the same solvent advances as used for the plate shown in Fig. 3. Figures 6 and 7 must therefore be referred to Fig. 3.

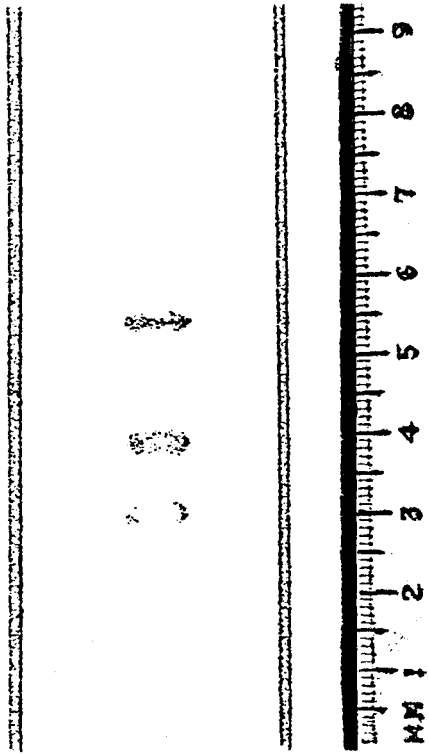


FIG. 6. Isolated PMD solvent removals following PMD-scheduled solvent advances at ambient temperature (see Fig. 3). Fixed-time heating.

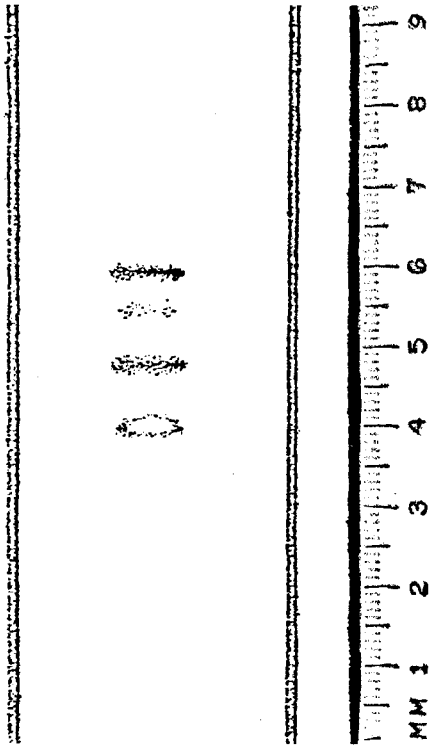


FIG. 7. Isolated PMD solvent removals following PMD-scheduled solvent advances at ambient temperatures (see Fig. 3). Scheduled-time heating.

Fixed Time. The fixed-time solvent removals lasted 100 sec, as indicated, at full power. These removals were sufficient to bring the solvent front to the origin, which had been placed 2.5 cm from the plate edge. The plate made this way is shown in Fig. 6.

Scheduled Time. The scheduled-time solvent removals were continued until the solvent front reached the origin, which had been placed 3.0 cm from the plate edge. The approximate durations of these scheduled solvent removals have been listed. The plate made this way is shown in Fig. 7.

PMD Stages, Isolated

Figures 8a–g show successive stages in a 3-cycle PMD Mode 1, 100-sec unit time program, with fixed time for solvent removal and with 100 sec

for preheat (preheat power is set by solvent removal power; here 10 or 100% of available power). The figures show the progress of the developments as listed in Table 2. Note that Fig. 8a isolates preheat as a spot reconcentration mechanism.

All spots for the eight separate plates comprising Fig. 8 were placed 2.0 cm from the plate edge.

TABLE 2

Fig. 8	Preheat (one time per run)	Previous cycles	Current cycle		Completed cycles to date
			Solvent advance	Solvent removal	
a	×				
b	×		×		
c	×		×	×	1
d	×	1	×		
e	×	1	×	×	2
f	×	2	×		
g	×	2	×	×	3

RESULTS AND DISCUSSION

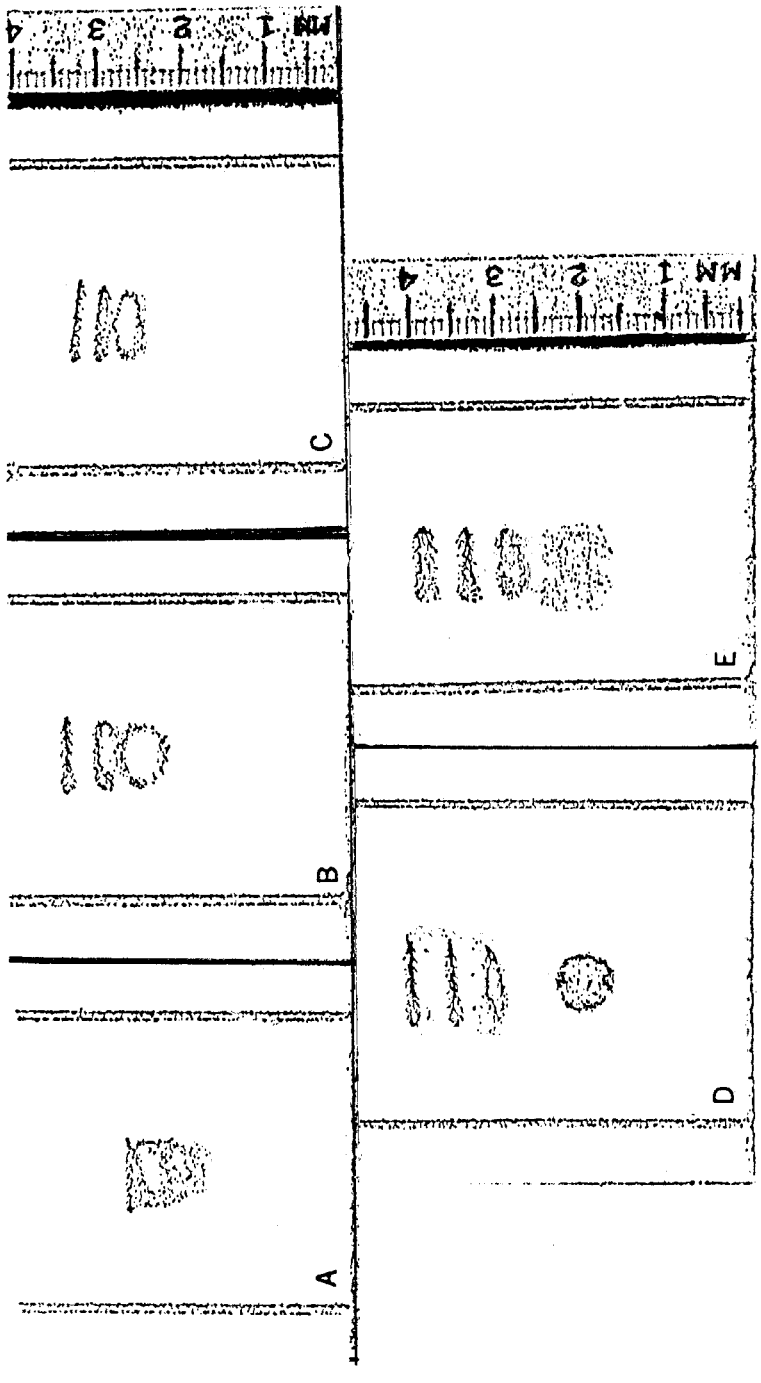
Spot Shape

The sole reason for forming the deposited spots into rings was that this technique reveals and emphasizes subsequent changes in spot shape.

A Multiple Development Quality Index

That UMC improves resolution, compared to a single development, has long been recognized (*10*). However, PMD (Fig. 1) has not previously been compared with UMC (Fig. 2) under such strictly comparable conditions, nor have the PMD solvent advance and removal spot-reconcentrating mechanisms been compared with each other. We wish to develop a numerical means of comparing the various types of multiply developed, ring-spotted chromatograms presented here. They differ in minimum or average spot width, range of spot width over the whole chromatogram, and center-to-center separation.

Any given potential separation of one spot from another like it requires a distance along the plate that is directly proportional to the spot top-to-bottom width. From the point of view of plate utilization, a quality



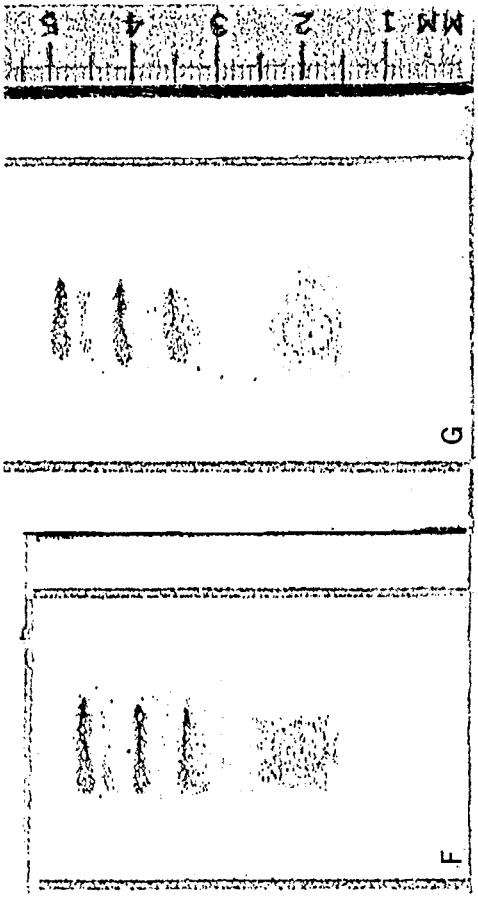


FIG. 8. Successive stages in a 3-cycle PMD, Mode 1, fixed time, 100-sec unit time. See Table 2.

index of such a separation is inversely proportional to the spot top-to-bottom width w.

Also, as comparison of Figs. 1 and 2 emphasizes, the thin-layer plate is more efficiently utilized overall if the spot top-to-bottom width is uniform throughout the chromatogram. Conversely, as the maximum-to-minimum spot width ratio w_{max}/w_{min} increases, part of the plate becomes less well used. Thus, for a whole chromatogram, its plate utilization quality is inversely proportional to the top-to-bottom width ratio of the widest to the narrowest spots, w_{max}/w_{min}.

Finally, there is no foregoing the necessity of putting distance Δd between the spots, regardless of how wide or narrow they may be. The quality of a separation is directly proportional to Δd.

In summary, the overall quality index should be inversely proportional to spot width w and spot width ratio w_{max}/w_{min}, and directly proportional to the spot separation distance Δd:

$$\frac{1}{w} \times \frac{1}{w_{max}/w_{min}} \times \Delta d$$

But this combination is equivalent to the classic resolution $R = \Delta d/\bar{w}$ multiplied by (w_{min}/w_{max}), which ratio we call R_w.

Therefore we shall use a multiple development quality index $Q_{MD} = RR_w$ to express both the actual resolution and the concurrent degree of plate utilization.

For instance, for a multiply developed chromatogram having a uniform spot width of 1.0 mm and a Butter Yellow–Indophenol Blue separation of 10.0 mm, the Q_{MD} would be 10.

Tabulated data are shown in Table 3.

(Note: Each spot of each chromatogram in this study was deliberately given an original, if fortuitous, 10 mm diameter. To compare the Q_{MD} of an otherwise comparable single-pass development with the Q_{MD} values shown in Table *3*, we must either produce or assign a $\bar{w}$ of about 10.)

Solvent Advances, Isolated

Solvent Advances at Ambient Temperature

Straight UMC, Fig. 2, differs little from PMD-scheduled solvent advances, Fig. 3. Each shows that R_F-dependent spot reconcentration that relies solely on ambient-temperature solvent advance. The lowest R_F spot in each remains nearly circular—but the one in Fig. 2 seems to have shrunk.

TABLE 3

Fig.	w (mm) R_F 0.4	R_F 0.15	R_F 0.08	Origin (mm)	Movement (mm) R_F 0.4	R_F 0.15	R_F 0.08	Separation d (mm) 0.4–0.08	$R=\frac{d}{\bar{w}}$	$R_w=\frac{w_{0.4}}{w_{0.08}}$	$Q_{MD}=RR_w$
1	2.0	2.0	2.0	25.	32.5	21.0	11.5	20.0	10.0	1.0	10.0
2	2.0	3.0	7.0	25.	53.5	40.0	21.0	32.5	7.2	0.29	2.1
3	2.5	5.0	9.5	25.	42.5	25.5	9.0	33.5	5.7	0.26	1.5
4a	2.0	2.5	5.0	25.	26.0	15.0	6.0	20.0	5.7	0.40	2.3
4b	2.0	2.5	5.0	30.	21.0	10.5	4.0	17.0	4.9	0.40	2.0
4c	2.0	4.0	6.0	35.	13.5	6.0	2.0	11.5	2.9	0.33	1.0
5	2.5	3.5	5.0	25.	26.0	10.0	3.0	23.0	6.1	0.50	3.0
6	2.0	3.0	3.5	25.	30.0	14.5	6.0	24.0	8.7	0.57	5.0 (3.4[a])
7	2.0	2.5	3.0	30.	29.0	17.5	9.5	19.5	7.8	0.67	5.2 (4.3[b])
8f	2.0	2.0	2.0	20.	27.0	20.0	9.5	17.5	8.8	1.0	8.8
8g	2.0	2.0	2.0	20.	31.0	23.5	17.0	14.0	7.0	1.0	7.0

[a] d of Fig. 2/$\bar{w}$ of Fig. 6.
[b] d of Fig. 3/$\bar{w}$ of Fig. 7.

Apparent Ring Shrinkage

The lowest R_F (Indophenol Blue) ring of Fig. 2 seems smaller than either the Fig. 2 ring origin or the comparable ring of Fig. 3. This apparent shrinkage is caused by diffusion. Diffusing randomly, the molecules cause the locus of highest molecular density to move inward. Based on diffusion within the solvent, such shrinkage is different from spot reconcentration, which is based on some molecules moving toward the solvent front while others beyond it cannot.

Solvent Advances at Temperatures above Ambient

Two sets of plates exist here. The origin placement was held constant in one, varied in the other.

In the constant-origin study, the heating preceding the solvent advances varied. We compare the resultant ring shapes and chromatographic quality in Table 4.

The stronger the preceding heating, the greater the spot reconcentration but the closer the spots to each other. Thus chromatogram quality is better with heating than without, but best with the more moderate, scheduled-time heating.

TABLE 4

Fig.	Heating precondition[a]	Ring shapes: Sudan Red (middle spot)	Ring shapes: Indophenol Blue ellipse ratio[b]	Q_{MD}
3	1	Open	0.95	1.5
4a	2	Closed	0.5	2.3
5	3	Closed	0.7	3.0

[a]Heating intensity: $1 < 3 < 2$.
[b]Ellipse ratio: longitudinal diameter/lateral diameter.

In the varied-origin, constant (fixed-time) heating study, Q_{MD} decreased with distance of the origin from the plate edge (Table 5). Both spot reconcentration and center-to-center separation decrease with increasing distance of the origin from the plate edge, thus the correspondingly decreasing quality index.

TABLE 5

Fig.	Distance, origin to plate edge (mm)	Q_{MD}
4a	2.5	2.3
4b	3.0	2.0
4c	3.5	1.0

Solvent Removal, Isolated

Figures 6 and 7 show the chromatograms made with the isolated solvent removals. We can now compare fixed vs scheduled times with respect to the resultant ring shapes and chromatographic quality (Table 6).

Compared to the stronger, fixed-time heating, the gentler but slower scheduled-time heating produces slightly better spot reconcentration and chromatographic quality.

Isolated Solvent Advances and Removals Compared

The isolated solvent advances and removals can now be directly compared with respect to spot reconcentration efficiency. However, to obtain comparable quality indices, we must use the center-to-center

TABLE 6

Fig.	Heating[a]	Ring shapes: Sudan Red	Ring shapes: Indophenol Blue[b]	Q_{MD}
6	Fixed	Open	0.4	5.0
7	Scheduled	Closed	0.4	5.2

[a]Heating intensity: fixed > scheduled. Heating duration: scheduled > fixed.
[b]Longitudinal diameter corrected for 0.5 mm contribution from ambient-temperature solvent advances, see Fig. 3. Ellipse ratio: corrected longitudinal diameter/lateral diameter.

TABLE 7

	Ambient	Fixed: Advances	Fixed: Removals	Scheduled: Advances	Scheduled: Removals
Sudan Red ellipse ratio	0.42	Closed	Open	Closed	Closed
Indophenol Blue ellipse ratio	0.94	0.5	0.4	0.7	0.4
Q_{MD}	1.5	2.3	3.4	3.0	4.3

separations from the isolated solvent advance chromatograms shown on Figs. 2 and 3 for the isolated solvent removal calculations, rather than the greater but abnormal separations from the ambient-temperature solvent advances used for Figs. 6 and 7 (Table 7).

The numbers show that solvent removal is more effective than solvent advance in both spot reconcentration and chromatographic quality. However, the ring for Sudan Red was not closed by fixed-time solvent removal, although it was for the corresponding advance. This suggests qualitatively that solvent advance may be the more effective with higher R_F spots in spot reconcentration.

Preheat Isolated

The plate shown in Fig. 8a was made with good conditions for preheat efficiency. The origin was 2.0 cm from the plate edge rather than 2.5 cm, the preheat time was 100 sec, and the heating was done with 60% of maximum power. Nevertheless, the spot for Indophenol Blue was not fully washed into the front.

Thus preheat by itself may not cause all spots to begin development as fully concentrated lines. If this were very important, the plate could be held under INTERIM power for a minute or so before the START button was pushed.

Successive Early PMD Stages

Figures 8a through 8g show the successive stages of a PMD 3-cycle development, stopped after each stage. (When stopped after a solvent advance, the plate was dried conventionally.)

The spot concentration of Butter Yellow was already completed with preheat. By the end of the second solvent advance, the ellipse from Sudan Red had closed. The Indophenol Blue ellipse became closed after the third solvent advance.

The quality index Q_{MD} falls from 8.8 for Fig. 8f, taken after the third solvent advance, to 7.0 for Fig. 8g, taken after the final solvent removal. The decrease was caused by the compression of the chromatogram during the solvent removal. Further PMD cycles gradually extend the chromatogram, so that after 5 cycles it shows a quality index of 10.0.

UMC and PMD Compared

This study also allows a comparison of UMC with that PMD in which the solvent is removed by heat under fixed-time conditions (Table 8).

Some other differences between UMC and PMD, not shown by this comparison, have been described (*1–8*).

TABLE 8

	UMC	PMD
Fig.	2	1
Butter Yellow	Closed	Closed
Sudan Red ellipse ratio	0.33	Closed
Indophenol Blue ellipse ratio	0.87	Closed
Resolution	7.2	10.0
Q_{MD}	2.1	10.0
Overall time (sec)	3500[a]	2000

[a]For plate removal from solvent, drying in air, and replacement in solvent; 200 sec allowed per development.

CONCLUSIONS

The mechanisms involved in PMD solvent removal by heating and solvent advance following solvent removal by heating have been experimentally isolated and measured for a 5-cycle program.

Solvent removal is more effective than solvent advance in reconcentrating spots and yielding chromatographic quality generally, although solvent advance may be the more effective with higher R_F spots.

The stronger the heating for solvent removal, the more concentrated the spots but the more compressed the chromatogram. The conclusions concerning heating strength and duration are mixed.

REFERENCES

1. J. A. Perry, K. W. Haag, and L. J. Glunz, *J. Chromatogr. Sci.*, *11*, 447 (1973).
2. J. A. Perry, T. H. Jupille, and L. J. Glunz, *Ind. Res.*, *16*, 55 (1974).
3. J. A. Perry and L. J. Glunz, *J. Assoc. Off. Anal. Chem.*, *57*, 832 (1974).
4. T. H. Jupille and J. A. Perry, *J. Chromatogr.*, *99*, 231 (1974).
5. T. H. Jupille and H. M. McNair, *Amer. Lab.*, p. 54 (September 1974).
6. T. H. Jupille and J. A. Perry, *J. Chromatogr. Sci.*, *13*, 163 (1975).
7. T. H. Jupille and A. Curtice, *Chromatographia*, *8*, 193 (1975).
8. J. A. Perry, T. H. Jupille, and L. J. Glunz, *Anal. Chem.*, *47*, 65A (January 1975).
9. U.S. Patent 3,864,250 (February 4, 1975).
10. E. Stahl (ed.), *Thin-Layer Chromatography: A Laboratory Handbook*, 2nd ed., Academic, New York, 1972.
11. E. V. Truter, *Adv. Chromatogr.*, *1*, 113–152 (1965).
12. J. A. Thoma, *Anal. Chem.*, *35*, 214 (1963).

Received by editor November 18, 1974