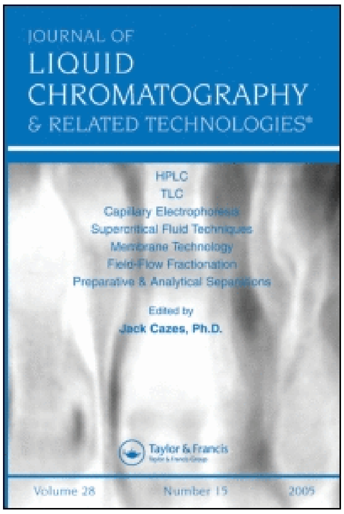

Journal of Liquid Chromatography & Related Technologies
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

MIXING, SETTLING, AND THE MOVEMENT OF THE INTERFACE BETWEEN THE MOBILE AND STATIONARY PHASES IN CCC

Philip L. Wood[a]; Ian A. Sutherland[a]
[a] Brunel Institute for Bioengineering, Brunel University, Uxbridge, UK

To cite this Article Wood, Philip L. and Sutherland, Ian A.(2001) 'MIXING, SETTLING, AND THE MOVEMENT OF THE INTERFACE BETWEEN THE MOBILE AND STATIONARY PHASES IN CCC', Journal of Liquid Chromatography & Related Technologies, 24: 11, 1699 — 1710
To link to this Article: DOI: 10.1081/JLC-100104372
URL: http://dx.doi.org/10.1081/JLC-100104372

J. LIQ. CHROM. & REL. TECHNOL., 24(11&12), 1699–1710 (2001)

STUDIES OF LIQUID-LIQUID CIRCULATION

MIXING, SETTLING, AND THE MOVEMENT OF THE INTERFACE BETWEEN THE MOBILE AND STATIONARY PHASES IN CCC

Philip L. Wood and Ian A. Sutherland*

Brunel Institute for Bioengineering, Brunel University,
Uxbridge, UB8 3PH, UK

ABSTRACT

In a J-type centrifuge, wave mixing is the primary method of mass transfer between mobile and stationary phases. It is observed centered about the proximal key node, while settling is observed centered about the Distal key node. A hypothesis using the Kelvin-Helmholtz wave stability criteria gives an explanation of why mixing and settling occur centered around these key nodes. The assumption of constant retention means that the stationary phase has zero linear velocity for all positions within the coil, and that the mobile linear velocity is constant. Closer examination of the Kelvin-Helmholtz wave stability criteria shows that the relative flow of two phases is important in creating waves. The interfacial movement can be viewed as a relative pumping action between the phases that increases the relative linear velocity of the mobile phase at the proximal key node to cause wave mixing. The interface can move within one loop of a coil. This can cause wave

---

*Corresponding author.

mixing at the proximal key node and settling at the distal key node when the lower phase is selected as the mobile phase.

The analysis presented, is based upon the following techniques: i) radial and tangential accelerations for helical wound coils on J-type centrifuges, ii) pressure gradient analysis, iii) Hagen-Poiseuille equation for laminar flow in circular bore tubing, iv) hydraulic mean depth to allow for two phase flow in circular bore tubing, v) computational numerical integration. A derived equation links the parameters: centrifuge radius (R), β value, rotational speed (ω), densities of both phases (ρ_L and ρ_U), viscosities of both phases (μ_L and μ_U), tubing bore diameter (d), and mobile phase volumetric flow rate (Q_m). It is hoped, that the derived equation can be used to qualitatively predict the effects of changes to the above parameters on CCC.

INTRODUCTION

The CCC process is made up of a series of mixing and settling zones, through which the mobile phase is pumped. Alternate mixing and settling are essential for chromatography to occur. During mixing, the interfacial area increases and each phase forms droplets in the other phase. Droplets have a high surface area to volume ratio that enhances the mass transfer of the sample-components between the two phases. The settling stage ensures that the mobile phase moves past the stationary phase, while no significant mass transfer occurs. This settling stage ensures that the mixing stages are discrete from each other.

How alternating mixing and settling stages occur was not fully understood, but must be related to the mobile and stationary phases flowing passed each other. Sutherland et al[1] have suggested, that the mixing and settling zones are due to alternating zones of instability and stability of the relative flow between the mobile and stationary phases. The Kelvin-Helmholtz stability criterion was stated to predict when the flow would be unstable and stable.[1]

The conditions for this can be calculated with the Kelvin-Helmholtz stability criterion.[1-6]

$$\frac{(U_1 - U_2)^4}{g} < \frac{4T_i.(\rho_1 - \rho_2).(\rho_1 + \rho_2)}{(\rho_1\rho_2)^2}$$

where U_1 and U_2 are the velocities of the two liquids, T_i is the interfacial tension between the two liquids, and ρ_1 and ρ_2 are the densities of the two liquids as shown in figure 1. The dotted line represents the interface between the phases. The flow is unstable when the left hand side of the Kelvin-Helmholtz stability

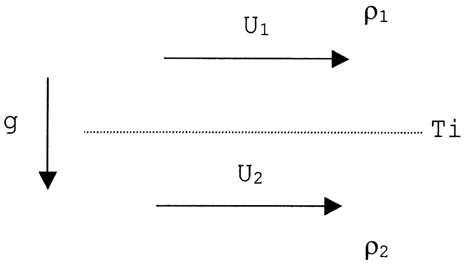

Figure 1. Flow relationship of two immiscible fluids used to derive the Kelvin-Helmholtz stability criterion.

criteria is larger than the right hand side. The flow is stable when the right hand side is larger than the left hand side. The g term represents the acceleration perpendicular to the direction of flow. This term (the acceleration due to gravity) appears in the inequality, since the inequality was originally derived for 1g environments. In a J-type centrifuge, the acceleration perpendicular to the flow is many times that of the acceleration due to gravity, and it is the radial acceleration predicted by equation 8 of the "Theory" section of Ref. 5, which replaces the g term. Figure 2 shows how the radial and tangential accelerations vary within one loop of a coil. The 0° and 360° degree angular positions represent the distal key nodes at each end of the loop under consideration. The 180° position represents the proximal key node. Examination of the curve for the radial acceleration shows that the minimum occurs at the proximal key node, which shows that the Kelvin-Helmholtz threshold velocity will also be minimum, hence, mixing is more likely to occur.

For the flow to be unstable, the relative velocity between the mobile and stationary phases must first exceed a value determined by the Kelvin-Helmholtz stability criteria. This value is called the Kelvin-Helmholtz threshold velocity. This threshold velocity (U_1-U_2) determines when waves, caused by a disturbance, will begin to increase in size. The waves continue to increase in size until mixing occurs. When the relative velocity drops below the Kelvin-Helmholtz threshold velocity, the size of the waves decreases, the flow is stable, and eventually settling occurs.[2]

EXPERIMENTAL

The values in Table 1 were calculated assuming a constant 80% retention through out the coil. The cross-sectional area occupied by the mobile phase can be calculated, 20% of the cross-sectional are of the tubing, and the velocity is

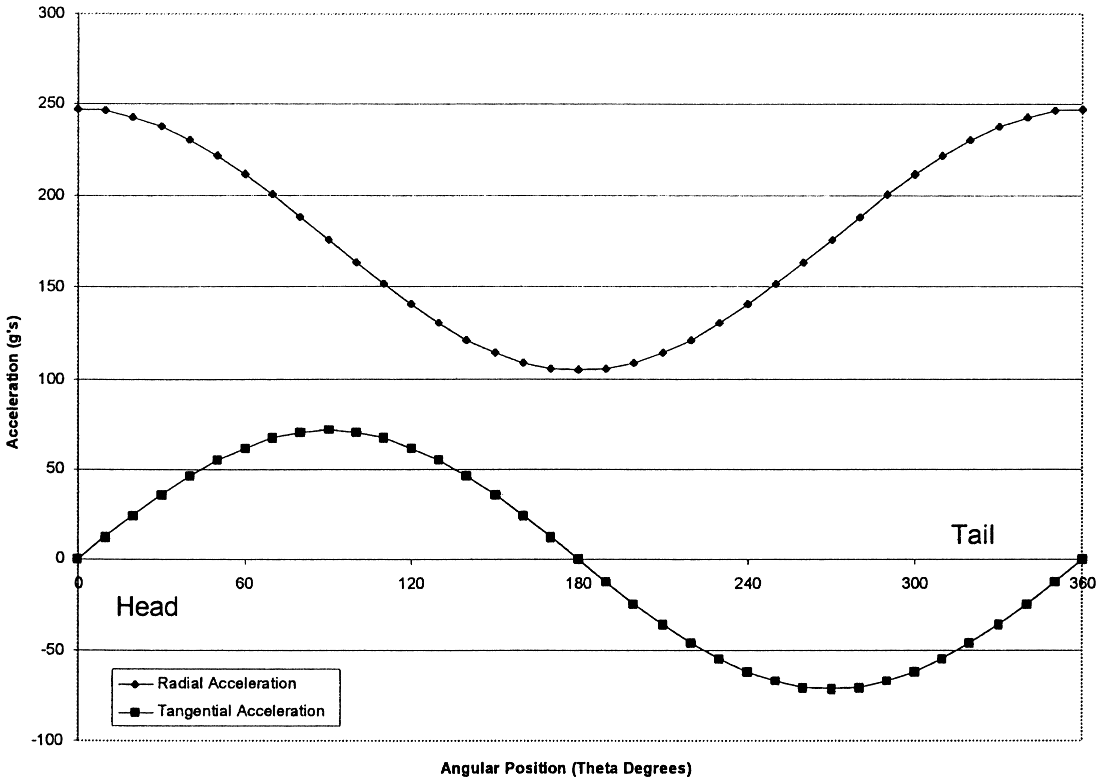

Figure 2. Variation of the radial and tangential accelerations that the upper and lower phases exert upon the coiled tubing plotted against the angular position in a single loop of coiled tubing. Equations 7 and 8 of Ref. 5 were used with a rotor radius of 100 mm, a β value of 0.615, and a rotational speed of 800 rpm.

Table 1. Mean Flow Speed (m/s) of the Mobile Phase for Various Open Tube Bores with Stationary Phase Retention of 80%

Tube Bore (mm)	Mobile Phase Flow Rate (mL/min) 1	2	4
1.6	0.041	0.083	0.166
3.35	0.009	0.019	0.038
4.75	0.005	0.009	0.019

determined by dividing the flow rate by the cross-sectional area of the mobile phase.

Figure 3 was generated using a rotor radius of 100 mm, a rotational speed of 800 rpm, the Kelvin-Helmholtz threshold velocity formula for two immiscible liquids, and the values shown are for a β-value of 0.615. The interfacial tension of Hexane/Water was assumed to be the same as that for Heptane/Water. The densities of the upper and lower phase for both phase systems were taken from Table 3.

The relative flow velocities in figures 5 and 6 were calculated using a mobile phase flow rate of 2 mL/min and assuming the same profile of the interface as shown in Figure 4. The relative flow velocity is the addition of the lower (mobile) phase and the upper (stationary) phase velocities, since the phases are flowing in the opposite directions.

RESULTS AND DISCUSSION

The flow velocities generated in the coil due to pumping the mobile phase shown in Table 1, are all lower than threshold velocities required for mixing shown in Figure 3. For mixing to occur, there must be another factor influencing the relative velocity between the mobile and stationary phases, other than that caused by externally pumping the mobile phase. The hypothesis examined in this

Table 2. Interfacial Tension Values Used to Calculate the Kelvin-Helmholtz Threshold Velocity, Values Taken from Ref [7]

Phase System	Interfacial Tension $\times 10^{-3}$ N/m 20°C
n-Hexane/Water, 2A, (Hydrophobic)	51.1
n-Butanol/Water, 2C, (Hydrophilic)	1.58

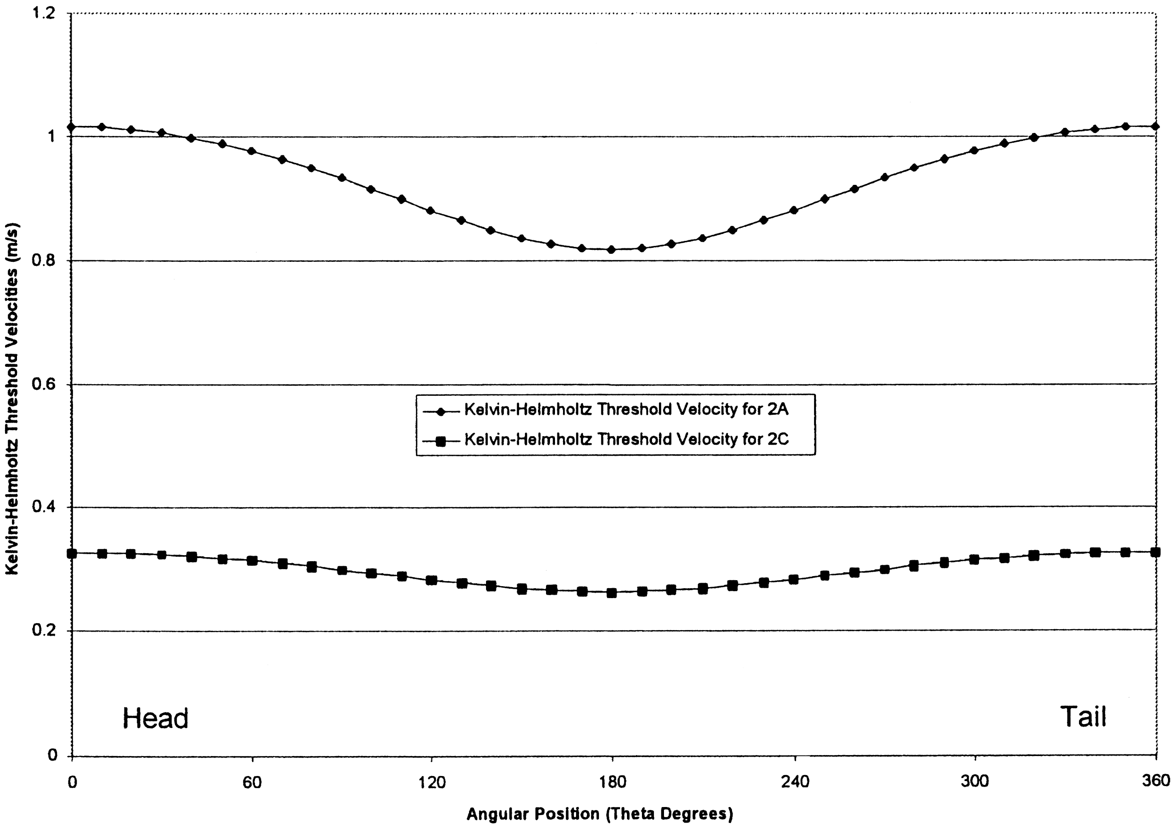

Figure 3. Kelvin-Helmholtz threshold velocity for the n-heptane/water phase system (2A) and the n-butanol/water phase system (2C) for one helical loop ($\beta = 0.615$, R=100 mm, 800 rpm).

Table 3. Physical Properties of the Phase Systems, at 30°C, Values Taken from Ref. [8]

Phase System	Lower Phase Density (kg/m^3)	Upper Phase Density (kg/m^3)
2A	994	658
2C	981	831

paper, is that there is a combination of pumping the mobile phase and the internal pumping of the phases within a loop, causing the relative velocity between the phases to exceed the Kelvin-Helmholtz threshold velocity.

Figure 4 shows that the interface moves inwards and outwards, depending upon the angular position within a single loop of a coil. This interfacial movement is the cause of the pumping effects reported.[6] An explanation of this interfacial movement is given in Ref. 5. The interfacial movement causes the retention of each phase to change with angular position. The upper phase attains a maximum retention at the proximal key node and the lower phase obtains its maximum retention at the distal key node. If the lower phase is made the mobile phase at a low flow rate, such that the profile of the interfacial is not substantially changed, then the lower (mobile) phase will have its minimum velocity at the distal key node and its maximum velocity at the proximal key node. The velocity of the lower (mobile) phase will change between the key nodes, depending upon how the retention of the mobile phase changes. Figures 5 and 6 compare the Kelvin-Helmholtz threshold velocities of Figure 3 with the pumped velocity of the lower (mobile) phase for the n-Heptane/Water and n-Butanol/Water phase systems, respectively.

Figure 5 shows that the relative flow velocity exceeds the Kelvin-Helmholtz Threshold velocity between 80° and 280° for the n-Heptane/Water phase system. Figure 6 shows that the relative flow velocity exceeds the Kelvin-Helmholtz Threshold velocity between 70° and 290° for the n-Butanol/Water phase system. Conway has observed mixing occurring between angles of 120° and 240°.[4] Mixing does not occur as soon as the Kelvin-Helmholtz threshold velocity is exceeded, but the flow is unstable and waves will form and then begin to grow.[2] The waves may need 50° to grow large enough to be observed as mixing waves. However, mixing waves will not begin to reduce in size until the flow velocity is below the Kelvin-Helmholtz threshold velocity, which does not occur until 280°, Figure 5, and 290°, Figure 6. This means, that mixing continues until these angles are reached and that settling does not occur until after these angles. Although, Figures 5 and 6 show how mixing can occur when the lower phase is mobile, the results do not fully match those observed.

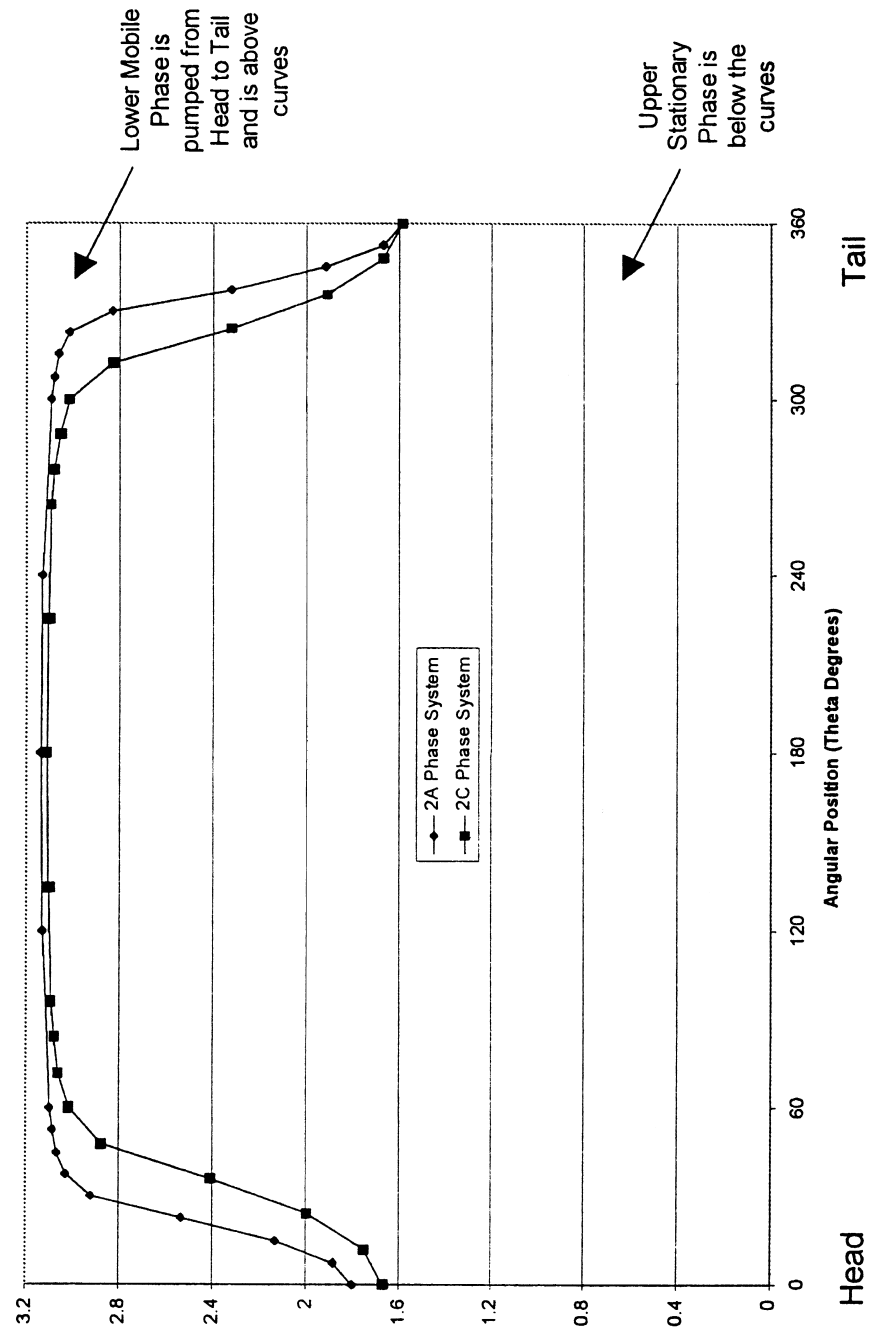

Figure 4. Positions of the Interfaces for the 2A and 2C phase systems taken from the calculations developed in Ref. 5.

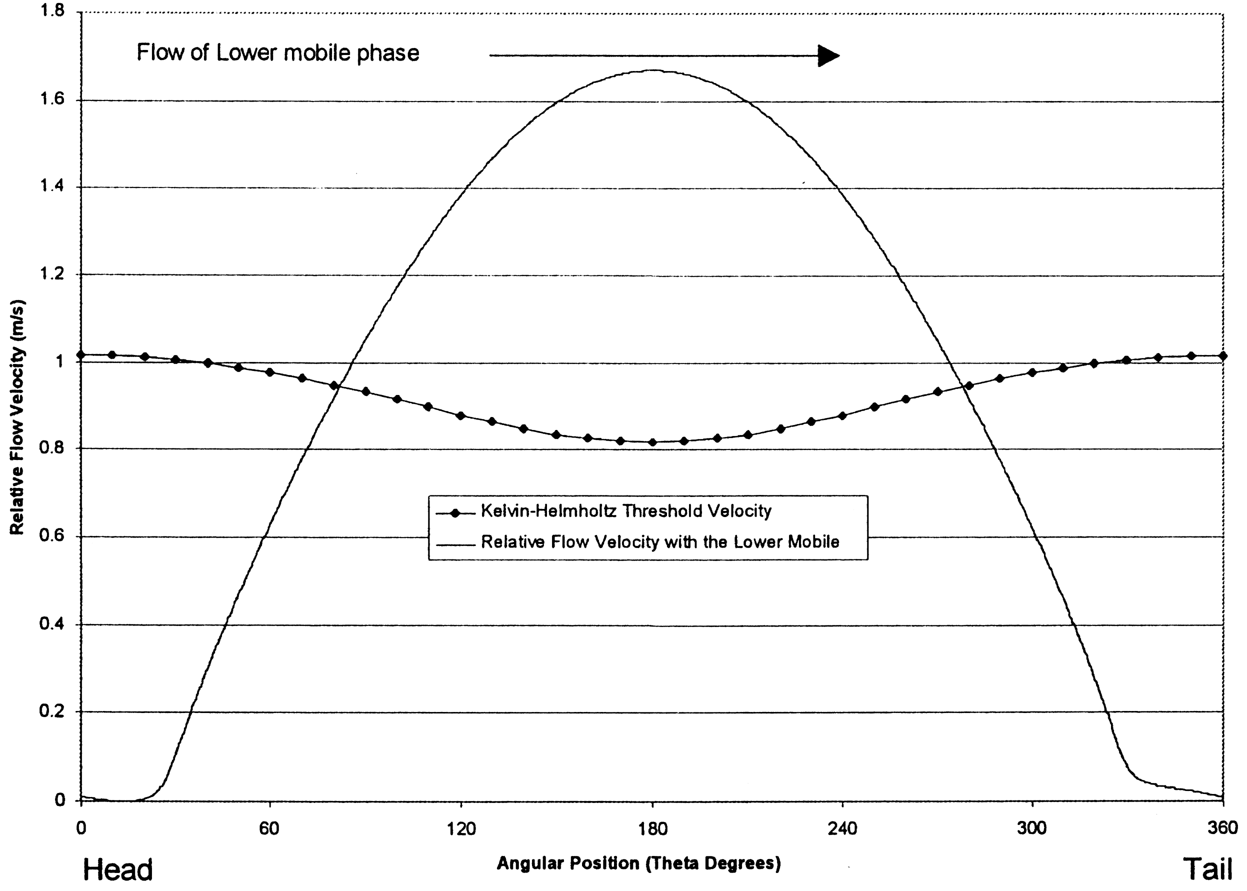

Figure 5. Kelvin-Helmholtz threshold velocity and relative flow velocity for the n-heptane/water phase system (2A) for one helical loop ($\beta = 0.615$, R=100 mm) with the lower phase mobile.

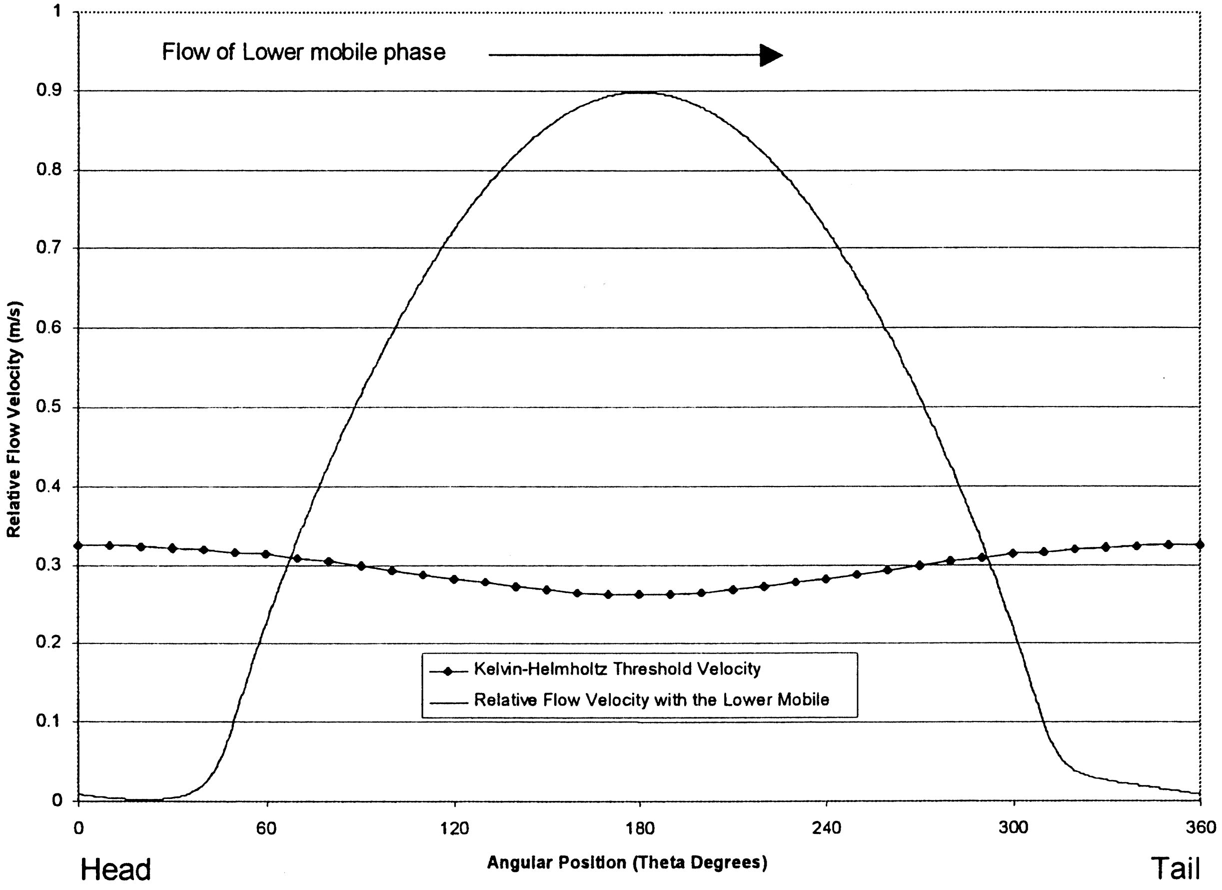

Figure 6. Kelvin-Helmholtz threshold velocity and the relative flow velocity for the n-butanol/water phase system (2C) for one helical loop ($\beta = 0.615$, R = 100 mm) with the lower phase mobile.

The Kelvin-Helmholtz stability criterion was derived for non-viscous fluids flowing in the same direction (no viscous effects), with the boundaries surrounding the two-phase flow assumed to be at an infinite distance from the interface (no wall effects), and occurring in a 1 g environment.[2] This does not match the conditions found in the coils of a J-type centrifuge and is probably inaccurate. To make this criterion match the conditions in the coils of a J-type centrifuge, it requires the following modifications to its derivation:

a) Circular bore tubing with a finite radius (wall effects).

b) The viscosities of each phase (viscous effects) need to be taken into account.

c) A radial acceleration term, similar to equation 8 in Ref. 5, needs to replace the g term.

d) Allowance for the phases to flow in opposite directions.

The prediction of the relative flow was based upon the movement of the interface without an externally pumped mobile phase. An accurate method of predicting the interfacial movement needs to be found that includes an externally pumped mobile phase. The method will need to be able to predict the interfacial movement for both upper phase mobile and lower phase mobile. The basis for such a method of prediction is the recommendations made in Ref. 6, that were to pump the lower mobile phase from head to tail and the upper mobile phase from tail to head. Essentially, the mobile phase is pumped in the direction that it would flow without external pumping, i.e., in the direction that the J-type centrifuge would pump the selected mobile phase. Hence, the external pump is not providing all of the effort required to pump the mobile phase through the coil.

The total amount of effort to pump the mobile phase is the sum of the effort of the external pump plus the effort provided by the J-type centrifuge. The addition of these two pumping effects has yet to be modeled to help predict the movement of interface. Figure 4 shows that the radial position of successive distal key nodes changes when there is no externally pumped mobile flow; successive distal key nodes are represented by the 0° and 360° positions on Figure 4. When hydrodynamic equilibrium is obtained by externally pumping a mobile phase, the radial position of the interface at each distal key node will be the same, otherwise, stationary phase will still be eluted from the coil. The mathematical model, when fully developed, should also show that the stationary phase retention decreases as the mobile phase flow rate increases.

CONCLUSIONS

A basic model for predicting the range of flow instability (mixing) within a coil planet centrifuge has been developed. This basic model should be developed further to encompass the following improvements: i) external pumping the mobile phase needs to be added to the model; ii) this is the condition for hydrodynamic equilibrium when the radial position of the interface at each distal key node is the same; iii) future modeling also needs to include the situation when the upper phase has been selected as the mobile phase. The Kelvin-Helmholtz stability criterion needs to be developed further to include the viscosities of the phases and the boundary influences of the tubing wall.

REFERENCES

1. Sutherland, I.A.; Jones, S.; Heywood-Waddington, D. A Preliminary Study of the Hydrodynamics of a Range of Solvent Systems in a Single Layer Coil Planet Centrifuge, Preparative Countercurrent Chromatography, Proc. Pittsburgh Conference and Exposition on Analytical Chemistry and Applied Spectroscopy, Atlantic City, 1986.
2. Craik, A.D.D. *Wave Interactions and Fluid Flows*, Cambridge University Press: 1992.
3. Ito, Y. Speculation on the Mechanism of Unilateral Hydrodynamic Distribution of Two Immiscible Solvent Phases in the Rotating Coil. J. Liq. Chromatogr. **1002**, *15* (15/16), 2639-75.
4. Conway, W.D, *Countercurrent Chromatography Apparatus, Theory and Applications*, VCH Publishers Ltd.: UK, 1990; 192, Fig 5-20.
5. Wood, P.; Sutherland, I.A.A. New Hypothesis on the Hydrodynamic Distribution of the Upper and Lower phase in CCC. J. Liq. Chromatogr. & Rel. Technol. **2001**, *24*.
6. Sutherland, I.A.; Muytjens, J.; Prins, M.; Wood, P.A. New Hypothesis On Phase Distribution In Countercurrent Chromatography. J. Liq. Chrom. & Rel. Technol. **2000**, *23* (15), 2259-2276.
7. *CRC Handbook of Chemistry and Physics*, 69th Ed.; CRC Press: 1988-1989.
8. Prins, M. *Countercurrent Chromatography, Research on the Physical Properties*, Brunel University publication: 1998.

Received October 28, 2000 Author's Revision November 23, 2000
Accepted December 3, 2000 Manuscript 5481