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The following aspects of continuous flow electrophoresis were studied: flow and temperature fields, hydrodynamic stability, separation efficiency, and characteristics of wide-gap chambers (the SPAR apparatus). Simplified mathematical models were developed so as to furnish a basis for understanding the phenomena and comparison of different chambers and operating conditions. Studies of the hydrodynamic stability disclosed that a "wide-gap" chamber may be particularly sensitive to axial temperature variations which could be due to uneven heating or cooling. The mathematical model of the separation process includes effects due to the axial velocity, electroosmotic cross-flow and electrophoretic migration, all including the effects of temperature dependent properties.
INTRODUCTION

Hydrodynamics plays varied roles in the continuous flow electrophoresis of small particles, in some situations the suspending fluid does little more than carry particles through the apparatus, in others the flow is so convoluted that electrophoretic separation is impossible. One of the complicating factors is the role of buoyancy forces which can destabilize the flow or establish an unfavorable, but steady laminar flow. To circumvent such problems it has been suggested that the apparatus be operated in a microgravity environment where, due to the reduced size of buoyancy forces, the chamber could be made larger and field strength increased. Then populations of large biological particles could be fractionated into narrow subpopulations on the basis of unique surface characteristics which are reflected in the electrophoretic mobility. Such an undertaking obviously requires careful evaluations of many types. The purpose of this investigation is to furnish a basis for understanding the hydrodynamic characteristics of the chamber and their effects on the separation process. Particular emphasis is placed on the role buoyancy plays in establishing the basic flow and affecting its stability.

Work began on this project in February of 1977 with the objective of assembling and evaluating current knowledge of the hydrodynamics of continuous flow electrophoresis. Four tasks were specified for the one year contract period:

1. Develop models to describe the flow and temperature fields;
2. Investigate the hydrodynamic stability of the flow field;
3. Develop a model to predict electrophoretic separation efficiency;
4. Review the SPAR apparatus and experiment.
Work on these tasks is complete insofar as it is covered by this contract and results are described in this report. The studies begun here continue under a separate NASA contract with Princeton University. The main part of the report is divided into two parts: SUMMARIES AND CONCLUSIONS, and DESCRIPTION OF RESULTS, where more detailed information is set forth.
SUMMARIES AND CONCLUSIONS

More detailed information on the various subjects is contained in the DESCRIPTION OF RESULTS sections, here we simply summarize and discuss conclusions.

Flow and Temperature Fields

The temperature field enters the problem because it alters the electrophoretic mobility of the particles and causes density contrasts which lead to buoyancy driven flows. The non-uniform temperature field itself derives from heat effects associated with the electric field and current. Although the field is three-dimensional, it is possible to simplify matters using perturbation methods. For present purposes we sought to establish the edge effects due to cooling through the side walls containing the electrodes (cf. Figure 1), the effects of temperature dependent conductivities for heat and electricity, and estimate time scales for thermal equilibration of the chamber.

The edge effects were found to be substantial in that they extend into the chamber for distances of 1-2 chamber thicknesses from each side. This alters the mobility of particles in these regions and has a dramatic effect on the flow through buoyancy effects. It was also found that the effects of temperature dependent conductivities were substantial, the calculated temperature rise being 70% larger for a wide-gap chamber (0.5 cm thick) than that calculated assuming constant properties. Although temperature relaxation times for the fluid are only 10-15 seconds for a narrow-gap chamber (0.15 cm) 2-3 minutes are required to establish the steady field in a wide-gap chamber.

* Representative dimensions, fluid properties, operating conditions, etc., are summarized on Table I, p. 72. A schematic diagram is on p. 7.
FIGURE 1.

Schematic representation of an electrophoresis chamber.
SAMPLE INLET

BUFFER FLOW

2d

2h

FRONT COOLING SURFACE

2d

ELECTRODE

SIDE WALL

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For the flow field analytical solutions in two-dimensions were constructed to investigate ways buoyancy could alter the axial flow and to study edge effects. One-dimensional models were then developed to investigate the effects of temperature dependent transport properties on the axial flow and electro-osmotic cross flow.

Several conclusions can be drawn from this part of the study.

(a) It is necessary to include effects of temperature on transport properties. Models which ignore this, or treat matters inconsistently, can be qualitatively and quantitatively misleading, especially with wide-gap machines like the SPAR device. With narrow-gap machines operated with modest field strengths (cf. Table 1) the use of 'average' values is satisfactory since temperature variations are usually small.

(b) In wide-gap machines operating in a 1-g environment the steady-state axial velocity profile is unsatisfactory at modest field strengths insofar as electrophoretic separations are concerned. An earlier study by Ostrach (using a 'constant properties' one-dimensional model) identified a buoyancy-driven feature which made downflow operation unsatisfactory. Edge effects and alterations due to temperature depending properties accentuate the buoyancy feature making matters worse. Upflow, which was once suggested as a means of overcoming the difficulty, turns out to be only marginally better for the cases studied. In downflow the difficulty arises from a recirculating eddy in the center of the chamber; in upflow two eddys appear, attached to the front and rear cooling surfaces, and these restrict the area available for separation. A micro-gravity environment would suppress or eliminate secondary flows of this sort. Of course other means of eddy suppression ought not be ruled out.
(c) Experiments at General Electric using the wide-gap SPAR apparatus disclosed a meandering sample flow pattern thought to be evidence of the structure noted in (b). Subsequent calculations made with the models developed here showed that the actual power levels were far lower than those required according to the theory and thus the meandering flow must be due to another process.

Hydrodynamic Stability

In an attempt to ascertain the cause of the meandering observed in the General Electric experiments the stability of several chamber configurations was examined. Attention focussed on buoyancy driven instabilities for obvious reasons and investigations of other sorts of instability, e.g., those due to viscosity stratification or electrokinetic effects, etc., were deferred. Three sorts of instability were investigated: the inception of cellular motion due to heat generation in a quiescent layer, roll cells in a buoyancy driven shear flow and the effect of an axial temperature gradient on a fully developed flow. Critical temperature differences for the quiescent layer or the shear flow are much larger than those present in the experiments. For the vertical chamber with axial flow a new two-dimensional instability was identified with an expecially low critical Rayleigh number. For conditions characteristic of the SPAR machine the critical axial gradient is (ca.) 0.5°C/cm.
Further experimental work will be required to establish whether or not the flow 'meandering' is a manifestation of the instability predicted by the current theory. If it is, then a micro-gravity environment will provide a means of avoiding it. Other types of instability mechanisms should also be investigated, however, so as to provide a comprehensive picture.

Prediction of Electrophoretic Separator Performance

Using the flow and temperature fields described earlier, a mathematical model of continuous electrophoretic separation was developed. The model (in brief):

(a) Accept as input data the dimensions of the chamber, operating conditions and flowrates, transport properties of the buffer, location and size of the sample injection tube, mobility distribution of the sample, zeta-potential of the wall coating, number and size of the sample outlet streams, etc.

(b) Predicts the mobility distribution in each of the sample withdrawal streams.

Calculations were carried out using computer programs which have been tested on "model systems". Further refinements will be made under the current NASA contract with Princeton.

SPAR Electrophoresis Experiment

Throughout the course of this investigation attention focussed on understanding the behavior of wide-gap machines and predicting their performance. We now have models of the flow and temperature fields and can estimate the electrophoretic separation characteristics of a given device.
Although refinements will be necessary, the requisite 'first generation' models now exist for interpreting results from SPAR (or other) experiments. Furthermore, the effects of changes in process variables can be examined so as to optimize the separation.

Final Comments

Work begun during this program is being pursued on under a joint program coordinated by Dr. R.S. Snyder of MSFC. These tasks include:

(a) Experimental studies (at MSFC) to ascertain the reason for flow meandering in wide-gap machines at 1-g. This will serve to prove or disprove the proposal that observed unsatisfactory operation is due to a buoyancy driven instability and assist in developing ways to circumvent the problem.

(b) Case-studies with the flow and separation models developed here to ascertain the ultimate (theoretical) capabilities of continuous flow devices.

(c) Theoretical work to extend the capabilities of the model, to develop an understanding of three-dimensional effects and finite sample concentration, and to investigate other hydrodynamic instabilities which could limit resolution. Establishing the limitations due to gravity and those arising from other phenomena.

Frequent observations of particle agglomeration and 'clumping' phenomena with cells underscore the need for an investigation of effects due to particle concentration. Recent theoretical studies (Batchelor, 1972) suggest substantial changes in sedimentation velocity at particle concentrations of a few percent but none of the extant studies deal with electrokinetic effects present in electrophoresis.

(d) The development of experimental techniques to test the model using mixtures of well-characterized particles. This will include micro-gravity experiments where appropriate.
DESCRIPTION OF RESULTS

I. Flow and Temperature Fields

Introduction

Inside a continuous flow electrophoresis chamber of the sort depicted on Figure 1 the temperature and velocity have a three-dimensional character. Cold buffer enters one end of the chamber and adjusts to the new geometry within a distance, $x_e$, which is given roughly by the formula (Schlichting, 1960)

$$x_e = 0.16 \text{Re} \, d.$$  

(1)

Here $d$ stands for the half-thickness and Re for the Reynolds number, $u_0 \frac{d}{v_0}$; $u_0$ is the mean axial velocity and $v_0$ the kinematic viscosity. Since the Reynolds number lies in the range 1-5, the entrance length is relatively short and here the velocity field can be modelled as being fully developed (viz. independent of $x$). As the buffer flow moves into the electrode region heat is added (volumetrically) through the action of the electric field and the associated current, so, to limit the temperature rise, the front and back walls are kept cold. For reasons that will be explained later (in the section on stability) the adjustment length for the temperature field can be substantial. Thus, in the electrode region the three-dimensional nature of the temperature field alters the structure of the velocity field through its effects on density and viscosity. Another contributory factor is the electro-osmotic cross-flow caused by the action of the field on the thin layer of charge in the fluid adjacent to the lateral boundaries. Although this velocity is typically much smaller than the axial velocity, it has a major role in altering the electrophoretic separation processes.
The models developed here to describe the temperature and velocity fields take advantage of three facts:

(i) The magnitude of the electro-osmotic velocity, \( w_o \), is small compared to \( u_o \).

(ii) The axial variation of the temperature is slow.

(iii) The effect of temperature on thermal conductivity and electrical conductivity is approximately linear over the temperature range of interest: \( 0^\circ - 35^\circ C \).

These facts justify the use of perturbation methods to develop a description of the temperature and velocity fields. First, because of (i), the velocity field can be split into two parts, an axial flow field due to forced and natural convection with a superimposed electro-osmotic flow. Next, due to the slow variation of the transport properties with axial position, (ii), the velocity fields can be split into a fully-developed part (independent of \( x \)) with corrections added later to allow for axial structure. Finally, the simple linear variation, (iii), makes the description of the temperature field particularly simple.

Separate parts of the sequel are devoted to:

A. Mathematical models for the structure of the temperature field
   a. A two-dimensional model in which the effects of temperature on thermal conductivity are suppressed. This provides a means of evaluating edge-effects due to heat transfer near the side wall electrodes.
   b. A one-dimensional model to evaluate effects of a temperature dependent thermal conductivity.
   c. A transient heat conduction model to estimate thermal relaxation times.
B. Models for the structure of the axial velocity field
   a. A two-dimensional, constant properties model provides a means of examining edge effects and buoyancy effects using the Boussinesq approximation.
   b. A one-dimensional, variable properties model is the basis for evaluating thermal effects and is used in the separation model described in Part III.

C. Models for the electro-osmotic cross-flow velocity
   a. A one-dimensional, variable properties model provides a basis for evaluating thermal effects and is used in the separation model (Part III).

Temperature Field

The equation for the conservation of thermal energy is

\[ C_p \ u \ \frac{\partial T}{\partial x} = \nabla \cdot (k \nabla T) + \sigma \varepsilon \]  \hspace{1cm} (2)

The symbols are: \( C_p \) - volumetric heat capacity, \( k \) - thermal conductivity, \( \sigma \) - electrical conductivity, and \( E_o \) - the electric field strength. \( E_o \) is assumed to be a constant throughout the analysis. Both \( k \) and \( \sigma \) vary with temperature in a linear fashion (see Figure 2) and so we write

\[ \sigma = \sigma_0 (1 + \sigma_1 \theta) \]  \hspace{1cm} (3)

\[ k = k_0 (1 + k_1 \theta) \]  \hspace{1cm} (4)

where \( k_0 \) and \( \sigma_0 \) are reference values evaluated at the wall temperature and \( \theta \) stands for a dimensionless temperature, i.e.,
FIGURE 2.

Thermal conductivity and electrical conductivity of the A-1 buffer.
$k$, watts cm\(^{-1}\) °C\(^{-1}\) $\times 10^3$

THERMAL CONDUCTIVITY

$\sigma$, (ohm-cm\(^{-1}\)) $\times 10^4$

ELECTRICAL CONDUCTIVITY

TEMPERATURE, °C
\[ T = T_w + \Delta T \theta \]  
\[ \text{(5)} \]

The wall temperature is \( T_w \) and \( \Delta T \) is a characteristic temperature difference. If we transform to dimensionless variables with \( u_0 \) as the characteristic velocity and \( d \) the characteristic length, then

\[ \text{Pe} \, u \frac{\partial \theta}{\partial x} = \nu \cdot [(1 + \kappa_1 \theta)\nu \theta] + 1 + \sigma_1 \theta \]  
\[ \text{(6)} \]

where \( \text{Pe} = \frac{C \mu d}{k} \) and \( \Delta T = \frac{\sigma_0 E_t d^2}{k} \).

Both \( \kappa_1 \) and \( \sigma_1 \) are less than unity: \( \kappa_1 \) is typically \( 0(10^{-2}) \) while \( \sigma_1 \) is \( 0(10^{-1}) \), and so it is convenient to represent the temperature by means of a perturbation series in \( \kappa_1 \), viz.

\[ \theta = \theta^{(0)} + \kappa_1 \theta^{(1)} + \kappa_1^2 \theta^{(2)} + \ldots \]  
\[ \text{(7)} \]

Substituting into (6) we generate a sequence of equations for \( \theta^{(0)} \), \( \theta^{(1)} \), ...

\[ \text{Pe} \, u \frac{\partial \theta^{(0)}}{\partial x} = \nu \theta^{(0)} + \sigma_1 \theta^{(0)} + 1 \]  
\[ \text{(8)} \]

\[ \text{Pe} \, u \frac{\partial \theta^{(1)}}{\partial x} = \nu \theta^{(1)} + \sigma_1 \theta^{(1)} + \nu \cdot (\theta^{(0)} \nu \theta^{(0)}) \]  
\[ \text{(9)} \]

e etc.

From equation (8) we deduce that in the thermal entrance region \( \theta^{(0)}/\Delta x \) is \( 0(\text{Pe}^{-1}) \) and varies exponentially. Viscosity and density also depend on temperature so that a description of the temperature field to the order implied by equations (8) and (9) would entail an expansion for the axial velocity of the form

\[ u = u^{(0)} + u^{(1)} + \ldots \]  
\[ \text{(10)} \]
where \( u^{(0)} \) is the constant properties solution, \( u^{(1)} \) accounts for temperature variations, etc. This expansion would be used to furnish complete velocity fields for (8) and (9). Because of the complexity of the problem it has not been practical here to attempt a solution which includes axial variations. Instead we have suppressed the axial structure and developed a 'zero-order' approximation with which we can assess the orders-of-magnitude of the thermal effects. An investigation of the details of the axial structure of the temperature and velocity fields is part of the work being done now under another NASA contract.

**Two-Dimensional, Constant Thermal Conductivity Model**

The major features of the fully developed temperature field can be found by solving equations (8) and (9), omitting the convective terms. The boundary conditions are:

(i) isothermal side walls, \( \theta = 0 \), at \( y = \pm 1 \).

(ii) heat transfer through the side walls at \( z = \pm H \) modelled in terms of a heat transfer coefficient, \( h \), i.e.,

\[-k \frac{\partial T}{\partial z} = h_0(T - T_B)\]

\( T_B \) stands for the coolant temperature. For the zero-order field we have, in dimensionless form:

\[0 = -\gamma^2 \theta^{(0)} + \sigma_1 \theta^{(0)} + 1\]  

(11)

\[\theta^{(0)} = 0, y = \pm 1, \quad \frac{\partial \theta^{(0)}}{\partial z} = -B_i \theta^{(0)}, \quad z = \pm H\]
The Biot number, $Bi$, is $h_0/d/k_0$, $H = h/d$. For $Bi = 0$ the end walls are perfectly insulating and the temperature field is one-dimensional. For $Bi \to \infty$ the end walls are isothermal with $\theta^{(0)} = 0$. Fourier transforms were used to solve equation (11) and the solution is

$$\theta^{(0)}(y,z) = \sum_{n=1}^{\infty} \left[ A_n \cosh \lambda_n z + B_n \sin \frac{n\pi}{2} (1 + y) \right]$$

(12)

where

$$A_n = \frac{Bi (1+(-1)^n)}{n^2 \lambda_n^n} (\lambda_n \sinh \lambda_n H - Bi \cosh \lambda_n H)^{-1}$$

$$B_n = \frac{1-(-1)^n}{n \lambda_n^n}$$

$$\lambda_n^2 = \frac{n^2 \pi^2}{4} - \sigma_1$$

Figures 3-6 display representative features of the temperature fields for narrow-gap and wide-gap chambers. Figures 3 and 5 are perspective views, Figures 4 and 6 are sections. A noteworthy feature is that the effect of side walls, shown on Figures 4 and 6, persists for a distance of 2-3 half thicknesses into the chamber at each side. For the wide-gap chamber (0.5 cm wide) this distance is (roughly) 0.75 cm and for the narrow-gap chamber (0.15 cm wide), 0.45 cm. Thus, due to the effect of temperature, particles within these regions will have a different electrophoretic mobility from those in the interior. In addition, buoyancy effects will be accentuated.
FIGURE 3.

Perspective view of the temperature field for the narrow-gap chamber operating at conditions listed on Table I (uniform thermal conductivity). Note: $2d = 0.15$ cm, $2h = 5$ cm. Eq. (12).
Sections of the temperature field as shown in Figure 3.
FIGURE 5.

Perspective view of the temperature field for the wide-gap chamber operating at conditions listed on Table I (uniform thermal conductivity). Note: $2d = 0.5$ cm, $2h = 5$ cm. Eq. (12).
Sections of the temperature field as shown in Figure 5.
One-Dimensional, Variable Thermal Conductivity Model

A one-dimensional temperature field corresponds to a very wide chamber, \( H \to \infty \), or one with insulated end walls at \( z = \pm H \). The zero-order field is given by

\[
\Theta^{(0)}(y) = \frac{1}{\sigma_1} \left[ \frac{\cos N_1 y}{\cos N_1} - 1 \right]
\]

\[
N_1^2 = \sigma_1
\]

and perturbations are found from solutions to

\[
\frac{d^2 \Theta^{(1)}}{dy^2} + \sigma_1 \Theta^{(1)} = -\frac{1}{2} \frac{d^2}{dy^2} [\Theta^{(0)}]^2
\]

Using Fourier transforms the solution is found to be

\[
\Theta^{(1)}(y) = \frac{2}{\pi} \sum_{n=1}^{\infty} f_s(n) \sin \left[ \frac{\pi n}{2} (1+y) \right]
\]

where

\[
f_s(n) = -\frac{1}{2} \frac{n^2}{n^2 - 4\sigma_1 / \pi^2} S_n[(\Theta^{(0)})^2]
\]

\[
S_n[(\Theta^{(0)})^2] = \text{sine transform of } (\Theta^{(0)}(y))^2.
\]

Temperature fields for narrow-gap and wide-gap chambers are shown on Figure 7, along with that for constant thermal and electrical conductivities. Figure 7 depicts matters in dimensionless form and it is seen that with the wide gap the temperature rise is 1.7 times that expected with constant properties at the conditions shown, for the A-1 buffer this is nearly 30°C.
FIGURE 7.

One-dimensional temperature field calculated so as to account for temperature dependent thermal conductivity and electrical conductivity, A - wide-gap, B - narrow-gap, C - constant properties (cf. Table I). Eqs. (13) and (15).
With the narrow-gap, the temperature rise is so small, about 1.5°C, that variable-property effects are negligible. Here we also see (upon comparison with Figure 5) that the influence of variable thermal conductivity serves to alter the temperature rise. Accounting for the variable thermal conductivity lowers this maximum by about 2°C for the wide-gap chamber at the conditions shown.

One-Dimensional, Transient Response Model

An estimate of the minimum time required to reach a steady thermal state can be made using a transient thermal model which ignores convection, since it tends to increase the equilibration time by adding colder fluid and withdrawing warm fluid from the region of interest. A rough estimate can be found from the characteristic relaxation time scale, \( d^2/a_o \). For the A-1 buffer \( a_o = 1.39 \text{ cm}^2/\text{s} \) so that for a narrow-gap machine (\( d = 0.075 \text{ cm} \)) the time scale is about 4 seconds; for the wide-gap machine the time-scale is 45 seconds. To attain a condition near the steady-state generally requires 2-3 'relaxation times', as shown on Figures 8 and 9. Data for the graphs were calculated using the solution to a transient heat conduction problem with heat generation, viz.

\[
\frac{\partial \theta}{\partial t} = \frac{\partial^2 \theta}{\partial y^2} + 1 + \sigma \frac{\partial \theta}{\partial y}
\]

with \( \theta(t,-1) = 0, \theta(0,y) = 0 \). The solution given by Carslaw and Jaeger (1959) is
Transient temperature response for a narrow-gap chamber (one-dimensional, constant thermal conductivity) (cf. Table I). Eq. (17).
Transient temperature response for a wide-gap chamber (one-dimensional, constant thermal conductivity) (cf. Table I). Eq. (17).
Obviously a more refined estimate could be made by solving a two-dimensional with convection included, however, the characteristic time for equilibration would probably not change too much. Thus, in any experiment with wide-gap machine at least 2-3 minutes should be allowed for thermal equilibration.

Axial Velocity Field

Equations to describe the axial velocity are derived from the Navier-Stokes equations using an expansion described earlier

\[ u(y,z) = u\textsuperscript{(0)}(y,z) + u\textsuperscript{(1)}(y,z) + ... \]  

The effects of buoyancy on the two-dimensional flow field are described in the first part of this section. In the second, where the effects of the lateral boundaries at \( z = \pm H \) are ignored, exact solutions are possible which account fully for the effects of temperature on viscosity and buoyancy.
Two-Dimensional, Constant Transport Properties Model

The 0(1)-velocity is described by solutions to

\[ 0 = -K + N_2 - N_3 \theta^{(0)} + v^2 u^{(0)} \tag{18} \]

where \( K \) is a constant (dimensionless) axial pressure gradient;
\[ N_2 = \frac{gd^3}{u_0^2} Re \] and \( N_3 = \frac{gd^3 \Delta T}{v_0^2} Re, \] \( Re = \frac{du_0}{v_0} \). The parameter \( N_3 \) describes the magnitude of the buoyancy effect while \((-K + N_2)\) is a constant to be determined from the fact that the volumetric flowrate is independent of the temperature rise, since the velocity is scaled using the mean velocity, \( u_0 \). Using Fourier Sine transforms we find

\[ u^{(0)}(y, z) = \frac{2}{\pi} \sum_{n=0}^{\infty} g_s(n) \sin \frac{\pi}{L} (1+y) \tag{19} \]

with

\[ g_s(n) = B_n \cosh \frac{n\pi}{L} z + C_n \cosh \lambda_n z + D_n \]

The coefficients \( B_n, C_n, \) and \( D_n \) are found from the relations

\[ B_n \cosh \frac{n\pi H}{2} + C_n \cosh \lambda_n H + D_n = 0 \]

\[ C_n = -\frac{N_3 A_n}{\sigma_1} \]

\[ D_n = -\frac{4}{n^2 \pi^2} \left[ \frac{K-N_2}{n} \left(1-(-1)^n\right) + \frac{N_3}{n} \left(1-(-1)^n\right) \frac{\lambda_n^2}{\sigma_1} \right] \tag{20} \]

Finally \((-K + N_2)\) is found from the requirement that

\[ \int_{-H}^{1} \int_{H}^{1} u^{(0)}(y, z) \, dy \, dz = 4H \tag{21} \]
Axial velocity field for the narrow-gap chamber (cf. Table 7). The velocity is almost indistinguishable from the fully developed parabola, $3(1 - y^2)/2$ except near the side walls at $z = \pm 33.5$. Eq. (19).
FIGURE 11.

Perspective view of the axial velocity field for the wide-gap chamber (cf. Table I). Downflow at $g = 980 \text{ cm/s}^2$. Eq. (19).
Central sections of the field shown on Figure 11. Note weak upflow in center and strong downflow along front and back cooling walls and near side walls at $z = \pm 10$. 
DIRECTION OF MAIN FLOW AND GRAVITY
FIGURE 13.

Perspective view of the axial velocity field for the wide-gap chamber (cf. Table I). Upflow at g = 980 cm/s². Eq. (19).
Central sections of the field shown on Figure 13. Note downflow along front and rear cooling walls and near the side walls at $z = \pm 10$. 
Results of representative calculations are shown on Figures 10-14. Figure 10 shows that buoyancy has relatively little effect on a narrow-gap chamber due to the excellent heat transfer. The maximum temperature rise for this case is about 1°C. End effects due to the no-slip condition at \( z = \pm \frac{1}{4} \) are similar to those encountered with the temperature field.

With the wide-gap chamber dramatic effects are present at unit gravity. Figure 11 is a perspective view, Figure 12 shows the velocity across two sections of the chamber with downflow. Here buoyancy causes regions of reversed flow throughout a large part of the central section. With a finite length chamber this implies that a large recirculating eddy would be present in the central section and the chamber would be difficult to use as an electrophoretic separation device. If the flow is reversed, the recirculating eddy splits apart and is attached to the side walls, as is shown on Figures 13 and 14. Since the recirculating regions are adjacent to the side walls, the central section allows more-or-less free passage of fluid and any sample. Of course, any sample that migrated into the eddy structures would be difficult to recover.

**One-Dimensional Velocity Fields**

Effects due to a temperature dependent viscosity and buoyancy can be investigated easily using a one-dimensional model and, in fact, exact solutions can be obtained. Since variations are confined to the \( y \)-direction we have

\[
0 = K + N_2 \rho + \frac{d}{dy} \nu \frac{du}{cy} \tag{22}
\]

with

\[
u(\pm 1) = 0
\]
The solution is
\[ u(y) = - K \left( \frac{1}{\mu} \int_{\lambda}^{Y} \int_{0}^{y} \frac{1}{\mu} dy \right) + N_{2} \left( \frac{1}{\mu} \int_{0}^{y} \frac{1}{\mu} dy \right) \] (23)
with \( K \) determined from the fact that the velocity scale is the mean velocity. Thus
\[ u(y) = - K \left( \frac{1}{\mu} \int_{\lambda}^{Y} \int_{0}^{y} \frac{1}{\mu} dy \right) + N_{2} \left( \frac{1}{\mu} \int_{0}^{y} \frac{1}{\mu} dy \right) \] (24)

Given expressions for the dimensionless viscosity, and density, both scaled on values evaluated at the wall, it is a straightforward task to evaluate the integrals. In the calculation a two-term expression was used for the temperature, viz.
\[ \theta = \theta^{(0)} + k \theta^{(1)} \] (7)

For purposes of comparison we can evaluate the velocity field for uniform viscosity and buoyancy. The temperature field is
\[ \theta^{(0)}(y) = \frac{1}{\sigma_{1}} \left[ \cos \frac{N_{3} y}{\cos N_{1}} - 1 \right] , \quad N_{1}^{2} = \sigma_{1} \] (13)

and the velocity works out to be
\[ u^{(0)}(y) = \frac{3 N_{3}}{2} (1-y^{2}) - \frac{3 N_{3}}{\sigma_{1}} \left[ \frac{1}{\sigma_{1}} + \frac{1}{3} - \frac{1}{\sigma_{1} N_{1}} \tan N_{1} \right] (1-y^{2}) + \frac{N_{3}}{\sigma_{1}} \left[ \frac{1}{\sigma_{1}} + \frac{1}{2} (1-y^{2}) - \frac{1}{\sigma_{1}} \frac{\cos N_{1} y}{\cos N_{1}} \right] \] (25)

For \( \sigma_{1} \rightarrow 0 \) we recover an earlier result due to Ostrach (1976),
It is important to note that the magnitude of the buoyancy effect as compared to forced convection is contained in the dimensionless group \( N_3 \), viz.

\[
\frac{g \beta}{v \rho_0} \frac{\sigma_0 E^2 d^4}{k_0}
\]

Here \( \beta \) represents an average coefficient of thermal expansion, \( \rho^{-1}(\partial \rho/\partial T) \). Thus, all other things being equal, going from a narrow-gap machine (2d = 0.15 cm) to a wide-gap machine (2d = 0.5 cm) alters the effect of gravity by a factor of \( (10/3)^4 \), roughly two orders-of-magnitude. It follows that whereas gravity forces play minor roles with narrow-gap machines the situation with wide-gap machines is quite the opposite.

For \(|N_3| \ll 1\) the velocity differs very little from the familiar parabolic profile characteristic of forced convection. For \(|N_3| \gg 1\) regions of reverse flow are present as illustrated on Figures 15 and 16. Qualitatively these profiles are similar to those derived from the two-dimensional model; quantitatively, however, they differ in the magnitude of the maximum velocity, which is higher here due to the effects of variable viscosity and buoyancy. Note regions of reversed flow in the center for downflow and adjacent to side walls for upflow (see insets).

The recirculating eddy present in the downflow configuration at these operating conditions renders this configuration almost useless for electrophoretic separation. Even if the flow was steady, the sample stream would be deflected towards the wall where electro-osmotic effects would be strong.
One-dimensional velocity field calculated with allowance for variable viscosity and density (cf. Table I). Downflow, \( g = 980 \, \text{cm/s}^2 \). Eq. (23).
One-dimensional velocity field calculated with allowance for variable viscosity and density (cf. Table I). Upflow, \( g = 980 \, \text{cm/s}^2 \). Eq. (23).
In the upflow configuration the eddys attached to the walls take up nearly 80% of the cross section and severely reduce the region available for sample separation.

At the same time, however, it must be recognized that the calculations presented here do not exhaust the set of configurations and operating conditions. The recirculations, for example, can be changed by independent control of the pressure gradient. Lowering the field strength has a dramatic effect on the buoyancy, since the temperature rise is proportional to $E_0^2$, but would necessitate an increase in the length of the chamber, etc. There are, therefore, a number of options which remain to be investigated, the calculations given here simply illustrate the hydrodynamic phenomena.

**Electro-osmotic Cross-flow Velocity Field**

The presence of a thin layer of space charge adjacent to the boundaries in the y-z plane (cf. Figure 1) along with the transverse electric field causes a well-known electro-osmotic flow (Shaw, 1969). In a parallel plate system open to reservoirs at $z = \pm H$ where $H \gg 1$ the velocity profile would appear to be flat up to a very small distance from the wall (a few multiples) of the Debye thickness, $\kappa^{-1}$) where a rapid transition occurs to accommodate the no-slip condition. For most purposes the double-layer thickness, $\kappa^{-1}$ is so small that we can approximate the velocity just outside this layer using one of the Smoluchowski equations,

$$w_0 = - \varepsilon E_0 \zeta/u$$

where $w_0$ is the velocity in the z-direction, $\varepsilon$, the dielectric constant, $E_0$, the field strength and, $\zeta$, the zeta-potential of the wall material in
contact with the solution in question. This apparent slip-velocity is of the order of a few microns per second for a potential gradient of one volt per centimeter. When the flow is constrained by walls at \( z = \pm H \) the profile is forced to be (roughly) parabolic so as to accommodate the condition of no net flow across the \( y-z \) plane. Although this velocity is too slow to affect the axial flow substantially the cross-flow interferes with any electro-phoretic separation by stretching the sample cross section. To provide an estimate of thermal effects and furnish a consistent representation of the velocity field for use in the separations model a simplified model is used. In this model end effects are omitted except insofar as the \( \psi^' \) is force the flow to turn round as sketched in Figure 17. The electro-osmotic velocity can be calculated from solutions to

\[
0 = -\frac{\partial p}{\partial z} + \frac{\partial}{\partial y} u \frac{3w}{\partial y}
\]

(28)

with \( w = w_0 \) at \( y = \pm d \).

The pressure gradient arises from the need to balance viscous forces outside the double-layer so as to produce a field with no net flow. If we scale the velocity using \( w_0 \), and lengths using \( d \), and write the viscosity as \( \nu_0 \nu(\theta) \) to account for the thermal effects we obtain

\[
0 = k_2 + \frac{d}{dy} \nu \frac{dw}{dy}
\]

(29)

which can be integrated to

\[
w = k_2 \int_{y}^{y_1} \frac{y_1}{\nu} dy_1 - 1
\]

(30)
The constant $K_2$ is found from the requirement that the net flow across any cross section in the $x$-$y$ plane be zero.

Although effects due to temperature dependent viscosity are relatively small and change the velocity only about 10\% in the wide-gap chamber, these could be significant if one were modelling the separation of particles with small mobility differences.
FIGURE 17.

Electro-osmotic crossflow velocity: A - plan view showing recirculation caused by end walls, B - velocity profile with constant viscosity, \((1 - 3y^2)/2\); C - velocity profile with wide-gap (cf. Table 1).
VELOCITY, w
II. HYDRODYNAMIC STABILITY

Introduction

During the early stages of development of the wide-gap machines large scale, irregular convective mixing was observed during experiments with dye tracers. One of the possible causes is the flow reversal caused by buoyancy (see Part I and Ostrach, 1976). At that time, the absence of quantitative data prevented a rest of this hypothesis. Later experiments at General Electric by H. Semon (1977) provided additional data and disclosed a persistent "wavering" of the sample stream at low power inputs; higher power levels caused irregular mixing. However, the power levels corresponded to maximum (centerline) temperatures only a few degrees higher than the wall (buffer) temperature, far below those which could produce the w-shaped profiles described in Part I. According to the analysis in Section I the dimensionless group \( g \sigma_0 \beta d^4 / \nu_k u_0 \) must be \( O(10^2) \) or more for buoyancy to alter the velocity profile significantly; in the General Electric experiments it was \( O(1) \). Changes in the orientation of the flow relative to gravity changed the allowable power levels somewhat but irregular flow persisted at voltage gradients necessary to give a significant electrophoretic separation. It was decided, therefore, to investigate the hydrodynamic stability of the flow.

Several sorts of phenomena are included under the general topic of hydrodynamic stability: the inception of motion in an otherwise quiescent system, the transition from one steady laminar flow to another and transition from laminar to turbulent flow. To establish orders-of-magnitude, theories for the stability of a horizontal layer and of a shear flow were reviewed. As a result it appears that neither of the buoyancy mechanisms involved in these...
two situations would be able to destabilize the flow.

Previous work on the stability of stratified fluid layers has centered on quiescent layers (cf. Chandrasekhar, 1961; Ostrach, 1964). This body of literature was combed to locate reports of special significance to the problem at hand which is distinguished by the combined processes of volumetric heat generation and fluid motion. Sparrow, Goldstein and Jonsson (1964) studied the buoyancy driven instability of a quiescent, horizontal layer bounded above and below by rigid walls and heated internally. Although a nonlinear profile does lower the critical temperature difference considerably, calculations based on parameter values for the wide-gap chamber showed that the critical difference is about 10°C which is well above the values reported by General Electric for the horizontal (or vertical) configuration with flow. Allowance for the effect of temperature on the rate of heat generation (which is not part of the Sparrow-Goldstein-Jonsson theory) may lower the critical temperature difference somewhat but an extension of this sort was not attempted, since it seemed best to understand the behavior of the vertical configuration which would be used in electrophoretic separation.

Vest and Arpaci (1969) studied the stability of natural convection in a vertical slot, where however, since the base flow is driven by an antisymmetric temperature, it is different from that in a continuous flow electrophoresis chamber. The stability with respect to roll waves, oriented perpendicular to the main flow was examined and a critical Grashof number of 7880 was found for this sort of instability. In the General Electric experiment the Grashof number based on the maximum centerline temperature is roughly 10 and, although the circumstances are quite dissimilar, it seems unlikely
that the observed meandering derives from a shear instability of the sort studied by Vest and Arpaci.

If the meandering and subsequent large scale convection are the result of an instability, then it must be one where the critical Rayleigh number is small. One possibility is that the instability derives from small axial temperature gradients which result from uneven heating or cooling. In addition the thermal region near the entrance to the electrode section extends over a region of several chamber half-thicknesses. This gradient can be estimated from an earlier equation describing the balance between convection, conduction and generation,

\[ \text{Pe } u(y) \frac{\partial \theta}{\partial x} = \frac{\partial^2 \theta}{\partial y^2} + 1 + \sigma \theta \]

for temperature-independent properties. This equation has solutions which decay exponentially with \( \lambda \) and have the form

\[ e^{-\lambda_n x} x \text{Pe}^{-1} f_n(y) \]

The mode which has the smallest eigenvalue, \( \lambda_n \), fixes the relaxation distance, \( x_e \). An estimate of the smallest eigenvalue can be found from the problem where the variable velocity \( u(y) \) is approximated by the (constant) average velocity. A more exact calculation would refine this estimate but would not change the order-of-magnitude. It is found that the distance over which the temperature adjusts to the ohmic heating is approximately \( \text{Pe}(d) \). For the wide-gap chamber operation at the conditions listed on Table I the Peclet number is about 50. Thus, since the temperature rise here is over 30 degrees, axial gradients at the inlet (and outlet) are of the order of 1°C/cm.
Stability of a Fully Developed Flow with an Axial Temperature Gradient

The presence of cold fluid above warmer fluid (in the region above the electrodes with the downflow configuration and in the electrode "outlet" region with upflow) is an unstable configuration. To study the stability of the velocity and temperature fields they were modelled with a fully developed axial flow in a rectangular channel. The basic temperature field consists of an axial gradient of magnitude denoted by \( \Delta T \), with lateral variations due to the balance between heat generation and conduction through the walls, i.e.,

\[
T = T_w + Ax + \Delta T \Theta(y,z)
\]  (31)

If we scale lengths, velocities, etc., as before we obtain

\[
\frac{3u}{3t} = -\frac{3p}{3x} + N_s(1-\beta \Delta T) - \frac{Ra}{Pr \cdot Re} x + \frac{Gr}{Re} \Theta + v^2 u
\]  (32)

\[
Pr \frac{\partial \Theta}{\partial t} + \frac{Ra \cdot Re}{Gr} u = v^2 \Theta + 1
\]  (33)

where

\[
Gr = g \beta \Delta T d^3/\nu^2_0 \quad \text{Grashof number}
\]

\[
Ra = g \beta Ad^4/\nu_0^2 \Theta_0 \quad \text{Rayleigh number}
\]

\[
Re = u_0 d/\nu_0 \quad \text{Reynolds number}
\]

\[
Pr = \nu_0/\Theta_0 \quad \text{Prandtl number}
\]

with \( u = \Theta = 0 \) on boundaries at \( y = \pm 1, z = \pm H \).
The mathematical problem is to identify conditions under which temperature and velocity fields other than the steady-state, symmetric forms exist. In particular, we are interested in forms with an exponential time dependence, \( e^{\omega t} \). The demarcation between stable and unstable flows is \( \omega = 0 \). Because of the linear structure of the equations, we can show that the imaginary part of \( \omega, I_m(\omega) \), is zero so that the so-called "exchange of stabilities" principle is satisfied and any disturbance will grow exponentially. Thus, we simply look for conditions where \( \omega = 0 \). The problem is decomposed into the sum of the steady parts \( u, \theta \) and perturbations \( \hat{u} \) and \( \hat{\theta} \).

This problem is very similar to one solved earlier by Ostrach (1955). Here, those results are extended to include two-dimensional effects and other disturbance planforms. In the early work, the instability was identified through a degeneracy in a base flow, now we see that the disturbances are superimposed on a symmetric base flow.

For the disturbance flow, \( \hat{u} \), and temperature, \( \hat{\theta} \), we have

\[
\nabla_{II}^2 \hat{u} = Ra \hat{u} \\
\hat{\theta} = Ra^{-1} \nabla_{II}^2 u
\]

(34)

where \( \nabla_{II}^2 \) stands for the two-dimensional Laplace operator. \( \hat{u} = \hat{\theta} = 0 \) on the boundaries. One set of solutions will, of course, simply be multiples of the symmetric (with respect to the x-z and x-y planes) steady-state solutions. We are interested in anti-symmetric solutions, which represent no change in the volumetric flow rate through the y-z plane.

In general, the solutions to these equations can be written...
\[ \dot{u} = u_1 + u_2 \]

where

\[ \nabla_{11}^2 u_1 = \lambda^2 u_1 \]

\[ \nabla_{11}^2 u_2 = -\lambda^2 u_2 \]

\[ \lambda^4 = Ra \]

The temperature is

\[ \Theta = Ra^{-1/2}(u_1 - u_2), \]

Solutions are

\[ u_1 = \sin qz(A_1 \sinh \gamma_1 y + B_1 \cosh \gamma_1 y) \]

\[ + \cos qz(A_2 \sinh \gamma_1 y + B_2 \cosh \gamma_2 y) \]

\[ \gamma_1^2 = q^2 + \lambda^2 \]

\[ u_2 = \sin qz(A_3 \sinh \gamma_2 y + B_3 \cosh \gamma_2 y) \]

\[ + \cos qz(A_4 \sinh \gamma_2 y + B_4 \cosh \gamma_2 y) \]

\[ \gamma_2^2 = q^2 - \lambda^2 \]

To satisfy the boundary conditions on the walls at \( z = \pm H \) either

(i) \( \sin qH = 0, A_2 = B_2 = A_4 = B_4 = 0, \ q = n\pi/H \); \quad (39)

or

(ii) \( \cos qH = 0, A_1 = B_1 = A_3 = B_3 = 0, \ q = \frac{2n-1}{2} \frac{\pi}{H} \). \quad (40)

The first condition, (i), corresponds to a disturbance that is anti-symmetric with respect to the \( x-y \) plane with upflow on one side and downflow on the
other. This form preserves the volumetric flow rate. The second condition, (ii), describes a flow with the requisite asymmetry if $B_2 = B_4 = 0$.

Next, with (i), either $B_1 = B_3 = 0$ so that $A_3 = 0$ and $\gamma_1 = \pm \sin \pi$ which gives

$$\lambda^4 = \eta^4 (1 + H^{-2})^2$$

(41)

or $A_1 = A_3 = 0$ so that $B_3 = 0$ and $\gamma_1 = i(2n - 1)\pi/2$ which gives

$$\lambda^4 = \eta^4 [(2n - 1)^2/4 + \eta^2/H^2]^2$$

(42)

With (ii), $A_4 = 0$ and $\gamma_2 = \pm \sin \pi$ so that

$$\lambda^4 = \eta^4 [\eta^2 + (2n-1)^2/4H^2]^2$$

(45)

The mode with the lowest critical Rayleigh number corresponds to the velocity field

$$u_1 = \sin \frac{\pi x}{H} \cos \frac{\pi y}{2}$$

(44)

with the critical Rayleigh number

$$Ra_c = \frac{\eta^4}{16} (1 + H^{-2})^2$$

(45)

For the wide-gap chamber with dimensions noted on Table 1, $Ra_c = 6.58$ and with a narrow-gap, $Ra_c = 6.11$. Using data for water at $10^\circ C$ the critical temperature gradient is $0.5^\circ C/cm$ for the wide-gap and $53^\circ C/cm$ for the narrow-gap chambers. These results agree qualitatively with experimental observation in that they disclose a great deal of sensitivity to axial
temperature gradients for the wide-gap machine. With a narrow-gap it would be rather difficult to achieve the axial gradients large enough to excite the instability.

Although this explanation is consistent with experimental findings for a vertical configuration, questions still remain with regard to tilted or horizontal configurations. These cases have not been analyzed in detail but it is easy to show (mathematically) that buoyancy effects coupled with an axial gradient will destroy the unidirectional character of the flow. At present, however, neither experimental data or quantitative theoretical results are sufficient to assess this matter fully.
III. MODELING THE ELECTROPHORETIC SEPARATION IN CONTINUOUS FLOW ELECTROPHORESIS

Introduction

We turn now to the task of describing how a sample with a particular mobility distribution will be altered in its passage through a continuous flow electrophoresis chamber. To provide a first approximation we have chosen to base the model on one-dimensional approximations to the various flow and temperature fields and ignore, for the present, two-dimensional effects due to side walls at \( z = \pm h \) (except insofar as they cause the electro-osmotic flow to recirculate). Consequently, the temperature and velocity fields and the particle mobility are functions of \( y \) alone. This procedure is accurate as long as \( d/h \ll 1 \) and regions near the side walls are ignored. The effect of the side walls requires much more extensive analysis and remains to be done.

The model for separation is based on the fact that the velocity of a particle of a given mobility can be written as a superposition of an axial velocity, \( u(y) \), and a transverse component made up of the electro osmotic flow velocity, \( w(y) \) and the particle velocity due to electrophoresis, \( v_m(y) \). Thus for a particle of mobility, \( m \), say, the velocity is

\[
\hat{u}(y) + \hat{k} [w(y) + v_m(y)]
\]

and particles which enter with the sample at \( y_o, z_o \) will exit at \( y = y_o, z = z_o + L[w(y) + v_m(y)]/u(y) \). If we denote the mobility distribution as \( N_m(x, y, z) \), to represent the number density of particles with mobility, \( m \), then

\[
u(y) \frac{\partial N_m}{\partial x} + [w(y) + v_m(y)] \frac{\partial N_m}{\partial z} = 0
\]
describes the fact that the particles are conserved. The number density at a point \(x, y, z\), is, therefore,

\[
N_m(0, y, z - x[w(y) + v_m(y)]/u(y))
\]

and the problem is simply one of "tracking".

To predict the mobility distributions at the exit from the separator we suppose that there are a number of collector tubes of area \((2d)(A)\) at the outlet plane. The mobility distribution in a given collector is simply

\[
\langle N_m \rangle_i = \frac{1}{\Delta} \int_{z_i}^{z_{i+\Delta}} N_m(z)dz
\]

\[
N_m(z) = \frac{\int_{-d}^{d} N_m(L, y, z)u(y)dy}{-d \int_{-d}^{d} u(y)dy}
\]

A computer program was written to implement this scheme; representative results are tabulated in Table II.

Outline of the Computation Method

The computation of the mobility distribution in the collection streams proceeds as follows:

Main Input Data are:

Camera dimensions \((2d, z_h)\)

Electrode length \((l)\)

Number of collection streams
Electric field strength
Buffer flowrate
Buffer temperature
Constants in the linear equation for buffer electrical conductivity
Constants in the linear equation for buffer thermal conductivity
Buffer conductivities for heat and electricity at the wall temperature
Coating mobility at 20°C
Mobility distribution of sample at 20°C
Location and size of sample stream.

The program then evaluates the temperature field using Eqs. (7), (13) and (15) and calculates the local values of density and viscosity using analytical formulas supplied. (Other relations can be used if necessary.) Then the local axial and electro-osmotic velocities are calculated using the appropriate equations for temperature dependent properties cited in Part I, Eqs. (23) and (30). Finally, particles on the edge of the sample are tracked to the outlet, using mobilities which reflect the local temperature, and the mobility distribution for each collector calculated.

Numerical output includes
Temperature field
Axial velocity field
Electro-osmotic velocity field
Mobility distribution for each collector.
Results of two representative calculations, one with a narrow-gap chamber, the other with a wide-gap, are shown on Table II. General conditions are as shown on Table I. The sample contained two types of particles in equal amounts. Although both configurations show complete separation the wide-gap configuration separates the sample into two widely spaced collectors; the narrow-gap chamber barely separates the two fractions and if either fraction contained a distribution of mobilities there would be overlap. It was not possible to operate the wide-gap chamber model at 1-g without the recirculating flows described in Part II - so gravity had to be suppressed in the calculation.

These calculations are intended to be illustrative of the sorts of results that can be obtained using the models derived here. Much more extensive calculations are required to establish the differences in the separatory capabilities of various continuous flow devices.
TABLE I.
Parameters used in numerical calculations

Fluid Properties (A-1 Buffer)

<table>
<thead>
<tr>
<th>Property</th>
<th>Narrow-Gap</th>
<th>Wide-Gap</th>
</tr>
</thead>
<tbody>
<tr>
<td>Buffer Temperature</td>
<td>10°C</td>
<td></td>
</tr>
<tr>
<td>Density</td>
<td>1.0 g/cm³</td>
<td></td>
</tr>
<tr>
<td>Viscosity</td>
<td>$1.33 \times 10^{-2}$ g/cm-s</td>
<td></td>
</tr>
<tr>
<td>Thermal Conductivity</td>
<td>$5.82 \times 10^{-3}$ watts/cm-°C</td>
<td></td>
</tr>
<tr>
<td>Electrical Conductivity</td>
<td>$6.9 \times 10^{-4}$ (ohm-cm)$^{-1}$</td>
<td></td>
</tr>
<tr>
<td>Coefficient of Expansion</td>
<td>$8.62 \times 10^{-5}$ (°C)$^{-1}$</td>
<td></td>
</tr>
<tr>
<td>Thermal Conductivity Coefficient</td>
<td>$2.58 \times 10^{-3}$ (°C)$^{-1}$</td>
<td></td>
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<tr>
<td>Electrical Conductivity Coefficient</td>
<td>$3.12 \times 10^{-2}$ (°C)$^{-1}$</td>
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Chamber Parameters

<table>
<thead>
<tr>
<th>Property</th>
<th>Narrow-Gap</th>
<th>Wide-Gap</th>
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</thead>
<tbody>
<tr>
<td>Electric Field Strength, v/cm</td>
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<td>70</td>
</tr>
<tr>
<td>Gravitational Constant *, cm/s²</td>
<td>980</td>
<td>980</td>
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<td>Gap Distance (2d), cm</td>
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<td>0.5</td>
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<tr>
<td>Width (2h), cm</td>
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<td>5</td>
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<tr>
<td>Length, cm</td>
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<td>10</td>
</tr>
<tr>
<td>Volumetric Flow, cm³/s</td>
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<td>0.7</td>
</tr>
<tr>
<td>Average Velocity, cm/s</td>
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<td>0.28</td>
</tr>
<tr>
<td>$\Delta T_T$, °C</td>
<td>3.17</td>
<td>35.2</td>
</tr>
<tr>
<td>Re</td>
<td>2.63</td>
<td>5.26</td>
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</table>

N₂

<table>
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<tr>
<th></th>
<th>Narrow-Gap</th>
<th>Wide-Gap</th>
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<tr>
<td>N₂</td>
<td>$8.88 \times 10^{2}$</td>
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<td>N₃</td>
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<td>k₁</td>
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<tr>
<td>a₁</td>
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* except where noted
TABLE II

Red-Blood Cell Separation in Narrow-Gap and Wide-Gap Machines

To demonstrate the use of the separation model two runs were made at conditions shown on Table I with samples made up of equal amounts of particles ('red-blood cells') with mobilities of $2.15 \, \mu\text{m-cm/v-s} \,(m=1)$ and $4.15 \, \mu\text{m-cm/v-s} \,(m=2) (at \, 20^\circ \text{C})$.

Other conditions were:
- Coating mobility $2.15 \, \mu\text{m-cm/v-s}$
- Sample inlet size $0.05 \, \text{cm}$
- Number of collectors 100

Locations of Separated Particles

<table>
<thead>
<tr>
<th>Narrow-Gap Chamber</th>
<th>1-13</th>
<th>14</th>
<th>15</th>
<th>16</th>
<th>17</th>
<th>18</th>
<th>19</th>
<th>20</th>
<th>21-100</th>
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</thead>
<tbody>
<tr>
<td>$m = 1$</td>
<td>0</td>
<td>.177</td>
<td>.590</td>
<td>.233</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>$m = 2$</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>.159</td>
<td>.477</td>
<td>.353</td>
<td>.021</td>
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<table>
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<tr>
<th>Wide-Gap Chamber*</th>
<th>1-20</th>
<th>21</th>
<th>22</th>
<th>23</th>
<th>24-29</th>
<th>30</th>
<th>31</th>
<th>32</th>
<th>33-100</th>
</tr>
</thead>
<tbody>
<tr>
<td>$m = 1$</td>
<td>0</td>
<td>.043</td>
<td>.546</td>
<td>.411</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>J</td>
<td>0</td>
</tr>
<tr>
<td>$m = 2$</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>.282</td>
<td>.610</td>
<td>.108</td>
<td>0</td>
</tr>
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</table>

* $g = 0$
ACKNOWLEDGMENTS

Dr. R.S. Snyder of MSFC, Dr. G.V.F. Seaman of the University of Oregon and Dr. R.N. Griffin of General Electric have each helped us to understand the many problems involved in the electrophoresis of small particles. Kurt Hebert of Princeton University did much of the computer programming.
BIBLIOGRAPHY


Four programs were written in FORTRAN IV:

ZTEMP - the two-dimensional temperature field, Eq. (12)
VELO - the two-dimensional velocity field, Eq. (19)
TRANS - the transient temperature field, Eq. (17)
TEMP - the separation model.

Listings of these are given on the following pages.
PORTSAN LEVEL 21

DATE = 7a110

09/14/89

J570

0322 m=2*k-1

0023 LABED2=K*W2*FL/4. - SIGMA1

0324 T=(1,-(-1.)*W)/W/LANBD2

0025 LABEDA=LANBD2*O.5

0326 A1=LANBD2*(Z-W)

0027 A12=LABEDA*(2+W)

0028 A13=-2. *LABEDA

0029 IF (A11+5U.) 1,1,2

0030 1

0131 GC TO 3

0032 3

0033 IF (A12+5C.) 4,4,5

0034 4

0135 GC TO 6

0036 6

0037 IF (A13+5U.) 7,7,8

0038 7

0039 GC TO 9

0040 9

0141 CONTINUE

0142 I=(I-5+2)/((LABEDA*(1,-I3)-Z*1.144+1))/)

0143 CONTINUE

0044 1

0045 IF (A11+5U.) 2,2,3

0046 2

0047 3G G(X1)=Y

0048 IF (Y1)=1*COL (1,1)

0349 K=2*NYPTS

0350 EC 64 I=1, NYPTS

0351 K=K-1

0352 I=I+1*ZZIS

0353 EC 50 J=1, NZPTS

0354 L=L-1

0355 5J Z=ZP (L,K)+1*EP (NYPTS+1-J,NYPTS+1-J)

0356 6J CONTINUE

0357 NYPTS+1

0358 AZ=AZTIS-1

0059 K=2*NYPTS

0060 EC 62 I=1, NYPTS

0061 K=K-1

0062 EC 92 J=1, NZ

0363 62 (NZTIS-J,K) = TEMP (NYPTS+J,K)

0064 NYPTS+1

0065 NZ=NP2S-1

0066 EC 85 J=1, NZ

0067 EC 65 I=1, NZ

0068 55 TEMP (J,NYPTS-1) = TEMP (J,NYPTS+1)

0069 2=C2-W

0070 EC 7C J=1, NZ

0071 W=4*LZ

0072 7C 54 (J)=Z

0074 EC 60 I=1, NZ
FORTRAN IV G LEVEL 21

DATE = 78119

09/10/4S

C

0075 Y=Y+LY
0076 X=X*(Y(J))
0077 JT=IF*10.+1.
0078 NCONT=JT*2
0079 TMAX=JT/10.

C

C PRINT THE FIELD FOR Y=0.
C
0080 WHITE (6,103)
0081 WHITE (6,110) SIGMA1,KERNS,8,31,MYPTS,NYPTS
0082 WHITE (6,101)
0083 WHITE (6,120)
0084 DC 40 J=1,80
0085 40 WHITE (6,103) ZZ(J),TEMP(J,MYPTS)

C

C PRINT THE FIELD FOR Z=0.

C

0086 WHITE (6,132)
0087 102 FORMAT ("Y",9X,"Y",10X,"TEMPERATURE")
0088 DC 41 I=1,MY
0089 41 WHITE (6,103) XX(I),TEMP(NYPTS,1)
0090 CALL PLVUZ (11,32,XX,IEE, coc,5,5,4.,TMAX+1.5,201,101,MY,NY,C,
0091 X,NCONT,J=1,TMAX,,O,0,0,0,-1.17,"TEMPERATURE FIELD")
0092 SIGF
0093 END

ORIGINAL PAGE IS OF POOR QUALITY
**VELOCITY DETERMINES THE DOWNWARD VELOCITY IN THE ELECTROPHORESIS CELL AS**

**FUNCTION OF BOTH Y AND Z. THROUGH A CALL TO THE PROCEDURE PLEVEL, 11**

**THE 3D PLOT A PERSPECTIVE VIEW OF THE VELOCITY SURFACE.**

**SIGMA - PARAMETER IN THE EXPRESSION FOR THE ELECTRICAL**

**CONDUCTIVITY**

**LI - THE L I T E R A R Y**

**W - THE ASPECT RATIO OF THE CHANNEL**

**NPTS - THE NUMBER OF TERMS TO BE TAKEN IN THE SERIES FOR THE**

**VELOCITY**

**NWIT - THE NUMBER OF EQUALLY SPAINED CONSTANT-Y LINES FROM (AND**

**INCLUDING) Y=0 TO Y=1 ALONG WHICH THE VELOCITY IS**

**EVALUATED**

**NWIT - THE NUMBER OF EQUALLY SPAINED CONSTANT-Z LINES FROM (AND**

**INCLUDING) 2=N TO 2=W ALONG WHICH VELOCITY IS EVALUATED.**

**DIMENSION (2N+1,2N+1,2W+1,2Z+1)**

**REAL LAMBDA, LAMBDAB, LAMDA3, Z**

**100 FORMAT (1H1)**

**110 FORMAT (' ', 2X, 'SIGMA = ', 2E10.3, 2X, 'EIO = ', 2E10.3, 2X,**


**120 FORMAT (' ', 2X, 'NITTS = ', 2E10.3, 2X, 'VELOCITY = ', 2E10.3)**

**130 FORMAT (1*)**

**140 FORMAT (2(5X, 1PEW.4))**

**150 FORMAT (5F10.3, 3J15)**

**3010**

**3110**

**3111**

**3112**

**3113**

**3114**

**3000**

**3010**

**3015**

**3016**

**3117**

**3118**

**3119**

**3200**

**3209**

**3221**

**3222**

**3223**

**3224**

**3325**

**3326**

**3027**

**Determine the constant, K, from the integrated velocity.**

**IC 5 W=1, NITTS**
POETBAN IV G LEVEL 21

MAIN

DATE = 7/110  5/14/24

0029  K=K+2
0030  LAMBDA=LAMBDA**0.5
0031  PHI2=K*K
0032  PHI4=PHI2*PHI2
0033  F1=F1/F2
0034  PI=PHI1*PHI2
0035  PIANH=PIANH(Y+PHI2)
0036  PIANH2=PIANH(LAMBDAM)
0037  S1=(S1+1)/F1
0038  S2=S1/LAMBD2
0039  S3=(S1+1)/F1
0040  SUM1=SUM1+S1
0041  SUM2=SUM2+S2
0042  SUM3=SUM3+S3
0043
0044  A1=SUM1*32./W/(PI**4)
0045  A2=SUM2*32./W/(PI**4)
0046  A3=GMN*2/PHI1/N1/SIGMA1/LE/PI/W
0047  F2=(N1+1)*A1-A2+B3*PHI1/A1
0048  W=111 (6,142) /FK
0049  112 FCERAT (1,2X,E10,3)
C

C EVOLUTION VELOCITY AT EACH (Y,2) POINT TAKEN.

C

C

20 1) K=1, KILKNS
0006  LAMBDA=LAMBDA**0.5
0007  F3=F3*F0
0008  F2=F2*F0
0009  C1=F1*(F1+F3)/FK
0010  C2=PHI2/LAMBD2
0011  AEG1=K*(Z+h)/TP1
0012  AEG2=K*(Z+h)/TP1
0013  AEG3=K*FIN
0014  A3=PHI1*(Z+h)/TP1
0015  A3=PHI1*(Z+h)
0016  A3=2.*LAMEDA*(LAM)
0017  A3=2.*LAMEDA*(LAM)
0018  IF (AEG1.LE.-100.) E1.E1.
0019  IF (AEG2.LE.-100.) E2.E2.
0021

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OF POOR QUALITY
```
0082 IF (ARG2 .GT. -100.) E2 = EXP (AN2)
0083 IF (ARG3 .LE. -1) ) ) ) E3 = .
0084 IF (ARG3 .GT. -100.) E3 = EXP (AN3)
0085 IF (ARG4 .LE. -100.) E4 = .
0086 IF (ARG4 .GT. -100.) E4 = EXP (AN4)
0087 IF (ARG5 .LE. -100.) E5 = .
0088 IF (ARG5 .GT. -100.) E5 = EXP (AN5)
0090 IF (ARG6 .GT. -100.) E6 = EXP (AN6)
0091 R = ( (1 + E2) ) / (1 + E3)
0092 A1 = C1 * E
0093 A2 = C2 * E
0094 A3 = C3 * ( (E4 + E5) ) / ( (1 + 1) )
0095 A4 = C4 * E
0096 A5 = C5 * E
0097 A6 = C6 * E
0098 CONTINUE
0099 U (J, I) = -1. * SUB / ID
0100 IF (J (J, I) .LE. UNIM) UNIM = U (J, I)
0101 IF (I (I, J) .LE. UNAM) UNAM = U (J, I)
0102 C1 = 0.
0103 C2 = 0.
0104 C3 = 0.
0105 C4 = 0.
0106 C5 = 0.
0107 C6 = 0.
0108 C7 = 0.
0109 C8 = 0.
0110 C9 = 0.
0111 U (J, I) = U (J, I)
0112 K = NYPTS - 1
0113 L = J (J, I) - 1
0114 M = I (I, J) - 1
0115 CONTINUE
0116 U (J, I) = U (J, I)
0117 U (J, I) = U (J, I)
0118 U (J, I) = U (J, I)
0119 U (J, I) = U (J, I)
0120 U (J, I) = U (J, I)
0121 U (J, I) = U (J, I)
0122 U (J, I) = U (J, I)
0123 U (J, I) = U (J, I)
0124 U (J, I) = U (J, I)
0125 U (J, I) = U (J, I)
0126 U (J, I) = U (J, I)
0127 U (J, I) = U (J, I)
0128 U (J, I) = U (J, I)
0129 U (J, I) = U (J, I)
0130 U (J, I) = U (J, I)
0131 U (J, I) = U (J, I)
0132 U (J, I) = U (J, I)
0133 U (J, I) = U (J, I)
0134 U (J, I) = U (J, I)
```
```

<table>
<thead>
<tr>
<th>Line</th>
<th>Code</th>
<th>Description</th>
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<tbody>
<tr>
<td>0135</td>
<td>WRITE (6, 12)</td>
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<tr>
<td>0136</td>
<td>WRITE (6, 130)</td>
<td></td>
</tr>
<tr>
<td>0137</td>
<td>CC 4 I = 1, NZ</td>
<td></td>
</tr>
<tr>
<td>0138</td>
<td>WRITE (6, 140) ZZ(I), U(I, NK)</td>
<td></td>
</tr>
<tr>
<td>0139</td>
<td>WRITE (6, 160)</td>
<td></td>
</tr>
<tr>
<td>0140</td>
<td>WRITE (6, 130)</td>
<td></td>
</tr>
<tr>
<td>0141</td>
<td>CC 93 I = 1, NY</td>
<td></td>
</tr>
<tr>
<td>0142</td>
<td>WRITE (6, 140) YY(I), U(NKPTS, I)</td>
<td></td>
</tr>
<tr>
<td>0143</td>
<td>JMIN = 1.1 JMAX</td>
<td></td>
</tr>
<tr>
<td>0144</td>
<td>UMAX = 1.1 UMAX</td>
<td></td>
</tr>
<tr>
<td>0145</td>
<td>CALL FERVO (11, ZZ, YY, U, W, 5, 3, 3, 2) I, 101, NZ, NY, J, JMIN, JMAX,</td>
<td></td>
</tr>
<tr>
<td></td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>0146</td>
<td>995 STOP</td>
<td></td>
</tr>
<tr>
<td>0147</td>
<td>END</td>
<td></td>
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</tbody>
</table>

```

-83-
C EVALUATE VISCOSITY AT EVERY OTHER TEMPERATURE POINT - ARRAY FNU

C

C = J=1
ARG = TUEFF-20.
ARG = J . 00067 / (1 + GJG62*ARG)
ARG = -7 . 6039 + ARG
MUC = EXP (ARG)
Rd = J J D JG 937
CMG = CMG+MUJ/MUO
VISC = RUD
EC J I = 1, NCF
J = J+2

C = J=3
ARG = TUEFF* (J) + TUEFF-20.
ARG = J . 00067 / (1 + GJG62*ARG)
ARG = -7 . 6035 + ARG
FNU (I) = EXP (ARG)/MUJ

C CONTINUE

C WRITE (6,11))
C WRITE (6,11) MUL, GCL, PCL, J, I, J
C WRITE (6,112) MUC, ELASTIC, HCCL, PCL, J
C WRITE (6,113) B, J, I, J
C WRITE (6,115) MAX
C WRITE (6,116) S FNP, FSP
C WRITE (6,117) FNP, FSP
C WRITE (6,118) S FNP, FSP
C WRITE (6,119) S FNP, FSP
C WRITE (6,120) C, J, I, J
C WRITE (6,121) (FNU(J), FNU(I), I = 1, 3)

C CUMULATIVE DENSITY INTEGRAL AS ARRAY FHCJ REPRESENTING DENSITY INTEGRAL 1-2
C Y = IC IC IC IC DENSITY 1 TEMPERATURE 1

C = 1/(STEMP-1.)
VHJ (1) = G.
&1 = 1
&2 = 3
EC = I = 2,3EP
A = E Y * (AHC (N1) + 4 * ahu (N1+1) + auC (N2))/J.
B = 2
C = 32
EC = &2,3EP
FHCJ (1) = A*FHCJ (1-1)

4 CONTINUE

C CUMULATIVE Y/MU INTEGRAL AS ARRAY REPRESENTING Y/MU INTEGRAL 1-2
C EVERY OTHER VISCOSITY POINT (1,..., EVERY FOURTH DENSITY-TEMPERATURE POINT)
1 FIRST EVALUATE Y/MU AS ARRAY S1.

C = 1/(STEMP-1.)
S1 (1) = G.
EC 5 I = 2,3EP
C evaluate FRCI/FNU as array U2 representing FRCI/FNU intermediate field Y(I)X(I).
C every other viscosity point. First evaluate FRCI/FNU at array Y(1).

C

0113  S1(I)=C.
0114  L=2,VEL
0115  S1(I)=FRCI(I)/FNU(I)
0116  7 CONTINUE
0117  J2(I)=C.
0118  N1=1
0119  N2=1+2
0120  9=2,VEL
0121  A=AXI*(S1(N1)+4.*S1(N1+1)+S1(N2))/3.
0122  N1=N2
0123  N2=N1+2
0124  J2(I)=A+U2(I-1)
0125  5 CONTINUE

C change U1 and U2 field integrals taken from also Y(I)X(I) to Y(I)X(I-1).
C velocity expressions--taken from Y(I).

C

0126  L=1,VEL
0127  J1(I)=U1(NVEL)-U1(I)
0128  U2(I)=U2(NVEL)-U2(I)

C evaluate the constant FNU by integrating -FNUU1+NU2U2 from X=1 to X=1.
C all setting result equal to CNL.

C

0129  CY=1./(NVEL-1.)
0130  N1=1
0131  CALL SYMS(DT,U1,N1,NVEL,A1)
0132  CALL SIMPS(DT,U2,N1,NVEL,A2)
0133  FNU=VISC/CNLY
0134  USCAL(5)=U1*DEPTH*DTU/TU/FNU/USCALL/4.?
0135  FNU=(-1.+FNU*47.)/A1

C evaluate th: al velocity

C

0137  USUN=0.
0138  UG 9 J=1,NVEL
0139  JU(J)=FNUU1(J)+FNUU2(J)
0140  9 USUN=USUN+U(J)

C
COMPUTE THE ELECTROOSMOTIC (Z-DIRECTION) VELOCITY. THIS HAS THE FORM OF \( \frac{1}{2} \).

So merely use integrated unit to determine constant, and then subtract 1.

```fortran
0141 #SCALE=LTNCB1*ECO*EC01
0142 FZ=1./AY
0143 #SUB=0.
0144 Jz=1
0145 LE 14 I=1,NVEL
0146 Jz=Jz+2
0147 CMCEL (1)=0.6*101*U1/(MU*FND (J))
0148 W (J)=FK*W1 (J)-1.
0149 12 *SUB=SUB+W (J)
```

PRINT ELECTRICAL AND U- AND Z- VELOCITY FIELDS AT VALUES OF I CONSERVATION.

TO USE VELOCITY POINTS.

```fortran
0150 Lyft=1./(NVEL-1)
0151 WRITE (6,135)
0152 WRITE (6,134)
0153 WRITE (6,125)
0154 LC 3) I=1,NVEL
0155 LY=LY*(I-1)
0156 Jz=Jz*(I-1)*4
0157 Jz=Jz*(I-1)*4
0158 LY=LY*LEFT/2.
0159 WRITE (6,135)
0160 WRITE (6,124)
0161 WRITE (6,125)
0162 LC 7) I=1,NVEL
0163 Y=LY*(I-1)
0164 Jz=Jz*(I-1)*4
0165 U (1)=U (1)*SCALE
0166 W (1)=W (1)*SCALE
0167 T (J)=T (J)*ISCALE+STEIFF
0168 1:
```

```fortran
Jz=Jz*SCALE
0170 Jz=Jz-NVEL
0171 CMCEL (J)=CMCEL (NVEL+1-J)
0172 U (J)=U (NVEL+1-J)
0173 14 W (J)=W (NVEL+1-J)
0175 NVEL=NVEL-1
0176 CMCEL (NVEL-I)=CMCEL (NVEL+I)
0177 U (NVEL-I)=U (NVEL+I)
0179 (NVEL-I)=U (NVEL+I)
```

```fortran
0180 WRITE (6,126) 1,1 (J),U (I),W (I)
0181 WRITE (6,127)
0182 WRITE (6,128)
0183 WRITE (6,129)
0184 WRITE (6,130)
0195 YY(J)=Y
```

20

```fortran
16 CONTINUE
```
FC8TBN IV G LEVEL 21

CALL SIMES(CY,AM,1,WKB,D)

PB8E(I)=D/USCAll/DEPTH

23C CONTINUE

C FIND THE AVERAGE MOBILITY DISTRIBUTION IN EACH SAMPLE COLLECTED.

NLIV=NCCL-1

DC 25C I=1,NCCL

JLMT=(I-1)*NLIV

TO 240 K=1,1,NCCL

L=K+JLMT

24C EP(K)=PB8E(L)

CALL SIMPS(DZ,2N,1,NCCL,C)

C=C/2CCL

C PRINT THE COLLECTED MOBILITY DISTRIBUTIONS.

WRITE(6,127) FMCCL(J),A,C

24C CONTINUE

31C CONTINUE

320 5> STCF

1247 XL

ORIGINAL PAGE IS OF POOR QUALITY
SUBROUTINE TEMP(NL,N3,F5,FK,T)
C THE TEMP SUBROUTINE EVALUATES THE TEMPERATURE FIELD T = T(Z) = f(z) + F(z)*exp(z)
C T(Z) IS THE ZERO ORDER FIELD. THE FORM IS GIVEN BELOW.
C THE TEMPERATURE AT MTEMP POINTS FROM Z=0 TO Z=1
C
0002 DIMENSION T(1111)
C CALCULATE ZERO ORDER FIELD

0003 FZ = 3.1415926
0004 F5 = F5** (0.5)
0005 CFZ1 = CC3(FZ)
0006 IZ = 1.2*FZ1
0007 ET = 1./ (11-1)
0008 FC 2 J = 1,11
0009 IZ = IZ*(J-1)
0010 IF* (1.+Z)*FZ/2.
0011 T(0) = CC3((FZ *(1+Z) - CFZ1)/CFZ1/F5)
C CALCULATE FIRST ORDER FIELD

0012 SUB = C
0013 FAK = 4.*F5/PJ/PJ
0014 DO 3 J = 1,11
0015 PJ = J*PJ/4.
0016 FK = C3*(PJ)
0017 FC = TFK1+FK2
0018 FC2 = (FC - FK1)/10.
0019 A = SIN((FC1)*4./FC1*PI*F2)/4./F4+FK1/F5/2.
0020 A = A*F1*F1
0021 PC1 = F1*PJ
0022 FC = J*J*PJ
0023 D = SIN((FC1)/2.)/FC1*PI*F2/2./PJ
0024 EM = E2*EM*F1*FC
0025 MC = 1.(J = 1)
0026 MC = C3*CF2*MC/J
0027 FJ3 = FJ1
0028 ASUM = (A+B+C)/FJ3/2.+(FJ1-FJ3)
0029 ASUB = ASUM*ASIN(J1A)
0030 SUB = SUB + ASUM
0031 S101 = SUB/F5/F5/CFZ1/CFZ1
0032 IG1 = ILNK2*Z/F5
0033 D = 1.*TZEFC*FK3*IG1
0034 RETURN
0035 END
FORTRAN IV G LEVEL 21

SUBROUTINE SIMPS(H,A,N1,N2,AREA)

C THE SIMPS SUBROUTINE USES SAMPSON'S RULE TO INTEGRATE AN ARRAY

C

0002 REAL PI,N SUM

0003 DIMENSION A(1001)

0004 END SUM=0.

0005 N SUM=0.

0006 M2=(N2-N1)/2

0007 J=N1-1

0008 IC 1 I=M1

0009 IC 1 J=M1

0010 IC 1 ENDSUM=ENDSUM+A(J)

0011 IC 1 J=J+1

0012 IC 1 M SUM=M SUM+A(J)

0013 IC 1 AREA=H*(2.*ENDSUM+M SUM-A(N1)+A(N2))/3.

0014 RETURN

0015 END

ORIGINAL PAGE IS OF POOR QUALITY