# ULTRASONIC ABSORPTION IN AQUEOUS SUSPENSIONS OF SMALL ELASTIC PARTICLES

BY

STEPHEN ARTHUR HAWLEY
A.B., Knox College, 1961
M.S., University of Illinois, 1963

THESIS

Submitted in partial fulfillment of the requirements
for the degree of Doctor of Philosophy in Biophysics
in the Graduate College of the
University of Illinois, 1967

Urbana, Illinois

September 26, 1966

Dear Professor Dunn:

I understand that___Stephen Arthur Hawley_____
expects to complete his doctoral dissertation in the near
future. As chairman of his doctoral committee, will you
please be responsible for making the necessary arrangements
concerning the time and place of the examination?

The final certificate, a deferred grade report form, and a
card on which to notify the University Press of the examina-
tion have been sent to the departmental office. When the
thesis has been accepted in final form, the certificate,
signed by the committee, should be returned to the Graduate
College together with a deferred grade report for the candi-
date's research credit. A copy of the printed Certificate
of Approval, signed by the members of the committee and by
the department head, must accompany each of the two copies
of the thesis that are to be deposited with the Graduate
College.

Very truly yours,

Stanton Millet
Associate Dean

This committee is discharged as of January 31, 1967.

COMMITTEE

Floyd Dunn_____          H. S. Ducoff (Biophysics)_____
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K. E. Van Holde (Chem)_____          R. S. Wolfe (Micro)_____

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_____              _____

Department of Physiology
and Biophysics
524 Burrill Hall
September 26, 1966

Dr. H. S. Ducoff
Dr. K. E. Van Holde
Dr. R. S. Wolfe
Dr. F. J. Fry

Dear Colleagues:

The final oral examination for Mr. Stephen A. Hawley will be held

Tuesday, September 27, 1966, at 3:00 p.m. in Room 526 Burrill Hall.

Sincerely,

Floyd Dunn
Chairman

cc:  Graduate College
     Mr. S. A. Hawley
FD/ped

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Stephen Arthur Hawley, Ph.D.
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## CHAPTER I

## INTRODUCTION

The absorption coefficient, $\alpha$ , of an acoustic plane progressive wave is defined by the relation

$$I_x = I_o e^{-2\alpha x}$$

where $I_x$ and $I_o$ are the coordinate-dependent and reference intensities, respectively. Although ultrasound is defined in the entire frequency range beyond the upper limit of human auditory perception at about 20 KHz (Hz $\approx$ cycle/sec), the range in which the absorption coefficient may be efficaciously determined experimentally is somewhat less. In liquids, for example, $\alpha$ has been determined most successfully using a plane wave field configuration in the frequency range between about 100 KHz and 200 MHz. Because of the interest in the absorption coefficient, it is the latter frequency range that will be of primary concern in the ensuing discussions.

The use of ultrasonic propagation parameters, in particular the speed of sound and absorption coefficient, as a means of examining the molecular properties of matter represents a field of extensive experimental development over the last thirty years. Although the theoretical understanding of the problems has grown at a corresponding rate, much of the important ground work is attributed to workers of the classical era of physics, most notably, Stokes,

Kirchoff, and Rayleigh.

The recognition that acoustic parameters may be utilized to delineate the kinetics of rapid reactions near equilibrium is attributed to Einstein (1920). The principle of this method resides in the fact that the acoustic wave is an adiabatic and compressional phenomenon and its passage through a fluid results in rapid, periodic fluctuations of temperature and pressure which under appropriate circumstances may perturb a chemical reaction from equilibrium. The resulting relaxational interaction with the acoustic beam has a characteristic absorption behavior which will be described in more detail in Chapter II.

The investigation of the absorption properties of solutions of macromolecules is relatively recent and represents a point of convergence of at least two independent lines of research. One of these has arisen in conjunction with efforts to elucidate the mechanisms of absorption and interaction of high-intensity noncavitating ultrasound in biological materials. Another research interest has utilized ultrasonic absorption in solutions of polyamino acids in order to characterize helix-coil transitions which arise in the cooperative dissociation of hydrogen-bonding.

Although a number of investigations have been directed toward the characterization of ultrasonic absorption properties of solutions of macromolecules and a substantial amount of data accumulated, elucidation of the absorption mechanisms has been elusive. This is

primarily due to the disparity between the limited

information contained in the acoustic data and the

apparent complexity of the hydrodynamic properties of

macromolecules. Edmonds (1966b) has suggested that the

absorption in protein solutions may be due to the manifesta-

tion of as many as twenty-two molecular processes. This may

be contrasted to the fact that in most macromolecular

solutions studied thus far, the dependence of the absorption on

frequency may be described in a simple polynomial form.

Another problem may be traced to the fact that many of the

processes responsible for absorption, e.g., dynamic volume

viscosity, are unique to ultrasonic propagation. Thus, it

is not usually possible to refer to other methods to provide

parameters to delineate individual mechanisms.

The acoustic absorption properties of aqueous suspensions

of the following macromolecules have been observed over a

sufficient frequency range for comparison: hemoglobin

(Carstensen et al., 1953, 1959b; Edmonds, 1962; Dunn et al.,

1966), serum albumin (Carstensen et al., 1953), fractionated

dextran (Hawley et al., 1965; Kessler, 1966) helical polyglu-

tamic acid (Burke et al., 1965), and polyethylene glycol

(Hammes and Lewis, 1966). Although a variety of tertiary

structures are represented by these materials, the absorption

properties exhibit the following characteristic similarities.

1) The absorption coefficient is proportional to concentra-

tion. This seems to be valid to concentrations of about

15%. Because the displacement of particles relative to

the solvent due to the acoustic wave is of the order of a

few Angstrom units, it is not expected that the requirement of diluteness, to avoid intermolecular interactions, will be as stringent as in static flow experiments. 2) Extrapolation to zero concentration reveals that the absorption contributions of the polymer and the solvent are additive. 3) The excess absorption due to presence of the macromolecule is significantly greater than that observed in equivalent concentrations of monomer. Since only the soluble amino acids have been examined, there may be some uncertainty with regard to the natural proteins; however, it seems likely that the excess absorption is related to polymerization. 4) The excess frequency-free absorption $\alpha/f^2$ decreases with frequency and approaches the value of the solvent or slightly above it. Unfortunately this occurs in the region of the upper limit of frequency available by efficacious techniques so that high frequency behavior has not been determined precisely. 5) The frequency-free excess absorption for a significant portion of the available spectrum appears to be nearly proportional (excepting polyethylene glycol) to frequency raised to a small negative power. The apparent experimental error indicated for polyethylene observations does not, however, preclude it from this description.

Although an empirical description reveals a simple relation between absorption and frequency, it does not follow that the underlying mechanisms are also simple. For example, the absorption behavior has been attributed to a distribution of relaxation events whose corresponding

characteristic frequencies are distrubuted over the spectrum

of interest. Assuming a continuous distribution Carstensen

et al., (1959b), Edmonds (1962), and (Burke et al., 1965) have

been able to fit empirically their data to curves generated

by suitable distribution functions. The analyses are

complicated by the disparity that exists between the low

information content of the acoustic data and the highly

specific requirements of meaningful analysis. Since

a priori information concerning the molecular origin of the

distribution function is not used in these cases, the mean-

ing of such analyses is doubtful (as is recognized by the

authors).

This investigation considers the absorption by aqueous

suspensions of a flexible, linear polymer and an impermeable

spherical, elastic colloid. The selection of these materials

was predicated on the assumption that their absorption

character would yield information about specific mechanisms

that may be important in the absorption character of all

polymer solutions. The macromolecules investigated to date

have been almost exclusively polyelectrolytes whose absorp-

tion character may well be determined by such factors as the

hydrophobic bonding, rigid tertiary structure, or electro-

viscous interactions (Dunn et al., 1966). It was felt that by

employing a linear nonelectrolytic polymer, contributions

to absorption from such effects could be eliminated and a

more definable system provided.

Dextran, a linear (1-6) anhydroglucose polysaccharide

was chosen because of its chemical stability, inertness,

and availability in the relatively large quantities of fractionated material required for ultrasonic characterization, viz., approximately 100g in the frequency range of interest. The molecular species is essentially a linear structure with 5 - 10% of the glucose residues existing as branching elements. In aqueous solutions it behaves essentially as that of a random coil with a small additional molecular dilation attributable to excluded volume effects associated with the branching structures (Granath, 1958). The ultrasonic absorption was measured in solutions of several fractions corresponding to weight-average molecular weights ranging from $10^4$ to $3.7 \times 10^5$.

The passage of an acoustic wave through a suspension of particles which have a density different from that of the surrounding medium will produce relative motion between the particles and the fluid. The dissipation of acoustic energy that results from Stokes frictional losses has been described theoretically by Lamb, 1945; Epstein, 1941; Urick, 1948; Angerer et al., 1951; and Fry, 1952. For particles in the size range of macromolecules, this theory predicts a negligible contribution to the total absorption behavior. It has been found, however, that the propagation of high-intensity noncavitating ultrasound through solutions of DNA will produce molecular degradation (Frontali, 1962; Hawley et al., 1963). Both the mechanical stresses that appear in the liquid and the force gradient that is attributable to relative motion are too small to account for the degradation

(Macleod, 1966). In order to verify the magnitude of the viscous losses at small particle diameters, absorption measurements were conducted in dispersions of polystyrene spheres.

The absorption in five aqueous polystyrene lattices with particle radii ranging from .044 micron to 3.5 micron has been measured. Four of the suspensions are of the monodisperse species utilized as secondary standards in electron microscopy.

CHAPTER II

THEORY

A. Relaxation Phenomena

It seems appropriate to interpret first the acoustic implications of mechanical relaxation, a concept which is found to pervade virtually all mechanisms associated with acoustic attenuation. When a steady state stress applied to a real liquid is instantaneously released, it is found that a finite time is required for the disappearance of the associated strain. The time delay may arise from any of a number of mechanisms and is a reflection of the molecular properties of the liquid (Herzfeld and Litovitz, 1958). If the stress is associated with excess pressure and strain with excess density, the time dependence of the excess density, $\delta\rho$, may be written as

$$\delta\rho = \delta\rho_0 \, e^{-t/\tau}$$

where $\delta\rho_0$ is the steady state excess density and $\tau$ is the relaxation time.

When the stress is periodic, as is the case with the propagation of ultrasonic energy, the relaxation events are manifested as a frequency-dependent phase lag between pressure and density. The persistence of the phase difference requires work to be performed on the liquid at the expense of acoustic energy. The result is a frequency-dependent absorption coefficient, viz.,

$$\frac{\alpha}{\omega} = \frac{K\omega/\omega_r}{1 + (\omega/\omega_r)^2} \qquad (2\text{-}1)$$

where

$$\omega_r = 1/\tau$$

and $\omega$ is the angular frequency $2\pi f$, K is a constant and $\omega_r$ is the relaxation frequency (Kinsler and Frey, 1962). When $\omega_r \gg \omega$, it is evident that the excess density will closely follow the pressure variations with a small phase lag. When $\omega_r \ll \omega$, the relaxation process is not perturbed and again the associated phase retardation will be small. The variation of the quantity $\alpha/\omega$ as a function of frequency exhibits a peak when $\omega\tau = 1$, and K may be evaluated in terms of the maximum value, $(\alpha/\omega)^*$. Summarizing these ideas: when $\omega = \omega_r$

$$\frac{\alpha}{\omega} = \frac{K}{2}\left(\frac{\alpha}{\omega}\right)^*$$

thus

$$\frac{\alpha}{\omega} = 2\left(\frac{\alpha}{\omega}\right)^* \frac{\omega/\omega_r}{1 + (\omega/\omega_r)^2}$$

so that when $\omega \ll \omega_r$

$$\frac{\alpha}{\omega^2} = \frac{2}{\omega_r}\left(\frac{\alpha}{\omega}\right)^* = \text{CONSTANT}$$

and when $\omega \gg \omega_r$

$$\alpha\omega = 2\omega_r\left(\frac{\alpha}{\omega}\right)^* = \text{CONSTANT}$$

Examination of (2-1) reveals that a fairly broad peak is determined by a single relaxation event. Generally about a

two decade frequency span is required to delineate accurately

the relaxation event (Davies and Lamb, 1957).

The possible origins of relaxation events in terms of

molecular mechanisms are multifold.  For example, compressional

energy may be coupled to internal degrees of freedom

associated with specific heat (thermal relaxation) or it

may perturb configurational transitions between states

of different specific volumes (structural relaxation),

(Herzfeld and Litovitz, 1959).  Coupling of acoustic

energy to a molecular transition is an equilibrium process

described kinetically for the transition between two states

by the usual rate equation:

$$[A] \underset{K_2}{\overset{K_1}{\rightleftharpoons}} [B]$$
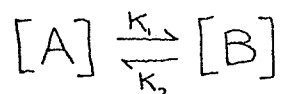
The equilibrium is most effectively perturbed at frequencies

determined by the rate constant.  For the above reaction the

relaxation frequency is

$$\omega_r = 1/\tau = \frac{1}{K_1 + K_2}$$

When a single relaxation event prevails, ultrasonic methods

may be gainfully employed in the analysis of molecular

transitions with time constants between about $10^{-5}$ and

$10^{-10}$ seconds (Hammes, 1966).  For a two state equilibrium

isothermal reaction, the expression for the molar free

energy change $\Delta G$ is

$$\Delta G = \Delta H - T\Delta S = \Delta U + P\Delta V - T\Delta S$$

where $\Delta H$, $\Delta U$, $\Delta V$, $\Delta S$, and  P are the molar changes in

enthalpy, internal energy, volume, entropy and pressure,
respectively. Determination of $\triangle G$ from acoustic parameters
is possible when either of two assumptions is valid: 1) when
$\triangle V \neq 0$, $\triangle U = 0$ and 2) when $\triangle U \neq 0$, $\triangle V = 0$ (Dunn, et al., 1966).
The processes described by conditions 1) and 2) are usually
referred to as structural and thermal relaxations, respec-
tively.

The most useful feature of the absorption spectrum
in delineating relaxation processes is the peak in
that occurs when $\omega T = 1$. If a simple two-state reaction
prevails in which the rate is determined by an activation
energy barrier $\triangle G^{\pm}$, it can be shown that the relaxation
time is of the form (Litovitz, 1959)

$$\tau = k e^{\triangle G^{\pm}/RT}$$

where K is a constant. Thus, by determining the temperature
dependence of $\tau$, it is possible to evaluate the associated
activation energy.

When more than one relaxation time is involved, the sit-
uation becomes more complicated and the quantity $\left( \alpha/\omega \right)$ is given
by

$$\frac{\alpha}{\omega} = 2 \sum_{n=1}^{N} \left( \frac{\alpha}{\omega} \right)_n^{*} \frac{\omega/\omega_n}{1 + (\omega/\omega_n)^2}$$

(2-2)

for N noninteracting relaxation events. Because (2-2) repre-
sents the summation of slowly varying functions, it can be
seen that the individual relaxation behaviors mask each other
at a relatively low density of relaxation events per frequency
interval. When the absorption can be traced to processes

which are governed by a continuum of relaxation times,
(2-2) takes the integral form (Carstensen and Schwan, 1959b)

$$\frac{\alpha}{\omega} = \int_{\omega_{r_1}}^{\omega_{r_2}} \frac{f(\omega_r)\,\omega/\omega_r}{1 + (\omega^2/\omega_r^2)}\, d(\omega_r) \qquad (2\text{-}3)$$

## B. Viscosity

At least a portion of the total absorption in all real
liquids is due to the finite viscosity of the fluid and since
viscous interactions impose a limit on the rate of molecular
flow, it may be considered a relaxation process. Passage of
a longitudinal acoustic wave through a liquid is accompanied
by both shear and volume deformations of the lattice. Energy
dissipation that occurs in the production of molecular trans-
lation is related to shear viscosity. Volume viscosity is
generally associated with the creation and extinction of va-
cancies in the liquid with a relaxation time related to the
rearrangement of molecules surrounding a vacancy (Herzfeld
and Litovitz, 1955). In addition, if the liquid exhibits
elastic response to mechanical stresses, the viscosities
include an imaginary component. Thus,

$$\eta_v^* = \eta_v' - i\eta_v''$$
$$\eta_s^* = \eta_s' - i\eta_s'' \qquad (2\text{-}4)$$

where the subscripts v and s refer to volume and shear
deformational modes, respectively. The viscosities can be
related to the corresponding bulk and shear moduli, i.e.,
the complex stress-strain ratios by

$$K^* = i\omega\eta^*_v$$

$$G^* = i\omega\eta^*_s \qquad (2\text{-}5)$$

so that $\eta'_v = K''/\omega$ ; $\eta''_v = K'/\omega$

$$\eta'_s = G''/\omega \; ; \; \eta''_s = G'/\omega \qquad (2\text{-}6)$$

The quantities G' and K' are usually referred to as the storage moduli as they are a measure of the stored dynamic energy. G" and K" are a measure of the heat loss per cycle and are correspondingly referred to as the loss moduli (Ferry, 1961). The dependence of the acoustic properties on the viscoelastic parameters are revealed by examination of the solution of the wave equation that can be derived from the Stokes-Navier hydrodynamic equations generalized to include a volume viscosity. The wave equation that arises is

$$\frac{\partial^2\xi}{\partial t^2} = \frac{[K^* + (4/3)G^*]}{\rho}\frac{\partial^2\xi}{\partial x^2}$$

where    is density and    is the displacement of a point in the medium. Using the procedure of Litovitz and Davis (1964), the velocity of a periodic disturbance is given by

$$V = \frac{[K^* + (4/3)G^*]^{1/2}}{\rho^{1/2}}$$

Assuming the presence of an absorption coefficient such that

$$\xi = \xi_0 \, e^{\{i\omega\tau - (\alpha - i\omega/v)x\}}$$

then

$$R_e\left\{K^* + (4/3)G^*\right\} = \rho V^2 \left\{\frac{1 - \left(\frac{\alpha V}{\omega}\right)^2}{[1 + \left(\frac{\alpha V}{\omega}\right)^2]^2}\right\}$$

$$I_m\left\{K^* + (4/3)G^*\right\} = \frac{2\rho V^2 \left(\frac{\alpha V}{\omega}\right)}{[1 + \left(\frac{\alpha V}{\omega}\right)^2]^2}$$

(2-7)

The quantity $\left(\frac{\alpha V}{\omega}\right)$ will be sufficiently small, viz., $< 10^{-2}$

for the materials of interest and equations (2-7) reduce to

$$K' + (4/3)G' = \rho V^2 \qquad (2-8)$$

$$K'' + (4/3)G'' = \frac{2\rho V^3 \alpha}{\omega} \qquad (2-9)$$

By combining (2-8) and (2-9) we have the relation

$$\frac{\alpha}{\omega} = \frac{\rho^{1/2} \left(K'' + (4/3)G''\right)}{2\left[K' + (4/3)G'\right]^{3/2}} \qquad (2-10)$$

Utilizing the relations between viscosities and moduli, (2-9)

becomes

$$\frac{\alpha}{\omega^2} = \frac{1}{2\rho V^3}\left[\eta'_v + (4/3)\eta'_s\right]$$

At sufficiently low frequencies $\eta'_s$ and $\eta'_v$ may be represented

by their static flow values and the result is essentially

the same as that derived by Stokes in 1945, viz., in the ab-

sence of experimental evidence, he assumed that

$$\eta_v' \approx 0$$

so that

$$\alpha = \frac{\omega^2}{2\rho V^3} \eta_s' \qquad (2-11)$$

In liquids, (2-11) constitutes what is usually referred to as the classical absorption coefficient. In general, the bulk viscosity does not vanish so that (2-11) often predicts values lower than are obtained experimentally.

The relaxation time associated with shear viscosity is given by (Kinsler and Frey, 1962)

$$\tau = (4/3) V^3 \rho \, \eta_s'$$

For water at $20.0^{\circ}C$, this corresponds to a relaxation frequency greater than $10^{12}$ Hz, which is out of range of the conventional experimental technology. If the excess absorption over the classical value is associated with bulk viscosity, the corresponding relaxation time is of the same order of magnitude as that due to shear processes.

## C. Dynamic Viscosities Of Solutions Of Random Coil Polymers

Generally the static flow viscosity does not describe adequately the dynamic properties of macromolecular suspensions. Equations (2-4) and (2-5) provide a more general form. Examination of these equations reveals, however, that determination of $\alpha$ and V for longitudinal wave propagation does not permit the separation of the contributions due to the bulk and shear moduli. Thus, without independent

information, assumptions concerning the relative behavior
of moduli are necessary.

Theoretical treatment of the dynamic shear viscoelastic
properties of dilute solutions of flexible polymers lead to
a distribution function (Rouse, 1953) that shows good
agreement with experimental evidence (Rouse and Sittel,
1953). The arguments evolve from a molecular model (Zimm,
1963) represented by a chain of massless beads connected
by Hookean springs which is suspended in a viscous liquid.
Free rotation of the segments is permitted about each bead
and the molecule is assumed to be long enough to permit a
Gaussian distribution of end-to-end chain lengths. Neglecting
hydrodynamic interaction of segments, a "free draining"
model is established (Rouse, 1953); the more general treat-
ment (Zimm, 1956) accounts for this interaction and is
referred to as the "non-free-draining" case. For the Rouse
theory, the prediction for the dynamic moduli are

$$G' = nkT \sum_{p=1}^{N} \frac{\omega^2 T_p^2}{1 + \omega^2 T_p^2} \qquad (2\text{-}12)$$

$$G'' = \omega \eta_0 + nkT \sum_{p=1}^{N} \frac{\omega T_p}{1 + \omega^2 T_p^2}$$

$$(2\text{-}13)$$

where $\eta_0$ is the solvent viscosity, k is Boltzman's constant,
T is absolute temperature, n is the number of polymer
molecules per unit volume and N is the minimum number of
submolecules such that random distribution of end-to-end
distances is observed (see for example, p. 159, Tanford,

1961).  The origin of the viscoelasticity is statistical.
Deformation of the molecule by an external force requires
energy since it results in a deviation from random behavior
and thus a decrease in configurational entropy.  Because
a finite time is required for reestablishment of random
orientation, some of the energy is stored in the transient
non-Gaussian average orientation.  In addition, the total
configurational behavior is described by a large number of
internal molecular modes and corresponding relaxation times
( $\tau_p$ ).  At low frequencies the viscosity is dominated by the
primary mode ($p = 1$) which corresponds to the translation of
the center of gravity (Rouse, 1953).  The corresponding
relaxation time is determined by

$$\tau_1 = \frac{6 M \eta_s [\eta]_0}{\pi^2 RT} \qquad (2-14)$$

where M is the molecular weight (Zimm, 1956).  This mode
contributes about 66% of the static viscosity.  The indivi-
dual importance to low frequency behavior of the other
modes decreases as the portion of the total molecule concerned
becomes shorter.

The contribution of the dynamic shear viscosity to the
ultrasonic absorption of solutions of random coil polymer
can be evaluated in terms of the Rouse theory.  By
combining (2-8) and (2-9) with (2-13) the following relation
results:

$$\frac{\alpha}{\omega} = \frac{\left[ K'' + 4/3 \left( \omega \eta_0 + nkT \sum_{p=1}^{N} \frac{\omega \tau_p}{1 + \omega^2 \tau_p^2} \right) \right]}{2 \rho V^3} \qquad (2-15)$$

Separating the shear from the volume viscous contributions

$$\frac{\alpha_s}{\omega} = \frac{2}{3} \frac{\left(\omega\eta_o + nkT\sum_{p=1}^{N}\frac{\omega\tau_p}{1+\omega^2\tau_p^2}\right)}{\rho V^3} \qquad (2\text{-}16)$$

where $\alpha_s$ is the absorption related to shear viscosity only. In the frequency range of interest in the present investigation (3-150 MHz), the period of the wave will be, generally, somewhat shorter than the primary relaxation time. In this regard (2-13) may be written as (Rouse, 1953)

$$G'' = \omega\eta_o + \frac{3\omega(\eta_s - \eta_o)}{\pi(2\omega\tau_1)^{1/2}} \qquad (2\text{-}17)$$

provided that $\quad 2 < \omega\tau_1 < N^2/250$

where $\eta_s$ is the steady state shear viscosity. For sufficiently dilute solutions (Zimm, 1956)

$$\frac{\eta_s - \eta_o}{\eta_o c} = [\eta]_o \qquad (2\text{-}18)$$

where $[\eta]_o$ is the intrinsic viscosity. Thus, by incorporating (2-17) and (2-18) into (2-16) the shear absorption is obtained in terms of steady state molecular parameters

$$\frac{\alpha_s}{\omega^2} = \frac{\rho_o V_o^3}{\rho V^3}\left[\frac{\alpha_{o,s}}{\omega^2} + \frac{3\eta_s[\eta]_o c}{\pi(\rho_o V_o^3)(2\tau_1\omega)^{1/2}}\right] \qquad (2\text{-}19)$$

$$2 < \omega\tau_1 < N^2/250$$

where $\rho_o$, $V_o$ and $\alpha_{o,s}$ are the density, velocity and Stokes absorption in the solvent, respectively. In dilute solutions $\frac{\rho_o V_o^3}{\rho V^3} \approx 1$, since the macroscopic com-

pressibility of a liquid will not be appreciably affected

by the presence of a small concentration polymer molecules

(Mason, 1961). Thus for dilute solutions we may write

$$\frac{\alpha_s}{\omega^2} = \frac{\alpha_{o,s}}{\omega^2} + \frac{3\eta_s[\eta]_o c}{\pi (2\omega\tau_1)^{1/2}\rho_o V_o^3} \qquad (2\text{-}20)$$

Unfortunately, a derivation of a quantitative relation

between the volume viscosity and molecular parameters is

not as clearly provided as it is in the case of shear

viscosity. For solutions of random coil polymers, however,

it is possible to make a few observations on the nature of

such a viscosity component. Any change in the apparent

volume viscosity of a liquid that may result from the addi-

tion of a random coil polymer must arise from an interaction

with the solvent. This is based on the premise that the

volume viscosity is related to the structural mobility of

clusters of neighboring molecules, a situation which is

clearly not provided by the polymer segments considered by

themselves. If the affinity of polymer segments for the

solvent is identical to the affinity between solvent

molecules, the presence of polymer would not be expected to

generate regions of high structural viscosity. Thus, a

significant volume viscosity can be developed only by the

formation of a polymer-solvent complex. The nature and

extent of the complex are determined in part by the inter-

action between monomer and the solvent as well as by any

cooperative effects that become manifest in polymerization,

e.g., arising from periodicity or immobilization of the monomer unit.

## D. Lamb-Epstein-Urick Theory Of Acoustic Attenuation In Suspensions of Small Particles: Relative Motion

The absorption of sound by a suspension of small spherical objects is treated generally by Epstein (1941). By solving the Stokes-Navier hydrodynamic equations one can obtain the following expression for suspensions of particles that are small compared to a wavelength. It is additionally assumed that the shear rigidity of the particle is large with respect to the suspending fluid.

$$\alpha = \frac{c_v \left( \frac{\rho_0}{\rho_P} - 1 \right)}{2 (V/\omega)} R_q \left\{ \frac{i + b - i b^2/3}{\left( \frac{\rho_0}{\rho_P} \right) - i \left( \frac{\rho_0}{\rho_P} \right) b - \left( 2 + \frac{\rho_0}{\rho_P} \right) b^2/9} \right\}$$

where

$$b^2 = \frac{i \omega \rho_0 R^2}{\eta_0}$$

and R is the particle radius, $\rho_P$ is the particle density, $c_v$ is the volume fraction of the particles, $\rho_0$, $\eta_0$ are the density and viscosity of the suspending liquid, respectively. The real part of this expression may be extracted by using the parameter (Carstensen and Schwan, 1959a)

$$\gamma^2 = \rho_0 R^2 / 2 \eta_0$$

so that

$$b^2 = 2 i \gamma^2 \omega$$

The expression for the absorption becomes

$$\alpha = \frac{2}{9}\frac{C_v}{V}\left(1-\frac{\rho_o}{\rho_p}\right)^2\left\{\frac{\gamma^2\omega^2[1+\gamma\omega^{1/2}]}{[1+\gamma\omega^{1/2}]^2+\gamma^2\omega[1+\frac{2}{9}(\frac{2\rho_o}{\rho_p}+1)\gamma\omega^{1/2}]^2}\right\} \quad (2\text{-}21)$$

Epstein interprets the origin of this absorption as the result of conversion of longitudinal wave energy to shear waves at the interfaces between particle and the suspending fluid. Since shear waves are highly damped in fluids, energy converted to this form is degraded as heat. As the diameter of the particle is increased with respect to the wavelength, two addition terms are necessary: first, as a correction for (2-21) and second, as a scattering term. The first of these takes the form

$$\Delta\alpha = \frac{C_v\omega^3 R^2}{V^3} R_\rho\left\{(10/9b)+(23i/4b^2)\right\}$$

when

$$\frac{\rho_p}{\rho_o} \approx 1$$

Reducing this as above,

$$\Delta\alpha = \frac{C_v\omega^3 R^2}{V^3\gamma\omega^{1/2}}\left[\frac{5}{9}+\frac{23}{8\gamma\omega^{1/2}}\right] \quad (2\text{-}22)$$

This scattering term is

$$\alpha_s = \frac{C_v\omega^4 R^5}{V^4}\left[\frac{(\frac{\rho_p}{\rho_o}-1)^2}{(\frac{\rho_p}{\rho_o}+2)^2}+\frac{1}{3}\left(1-\frac{K_o'}{K_p'}\right)^2\right] \quad (2\text{-}23)$$

where $K_p'$ and $K_o'$ are the real parts bulk moduli of the

particle and liquid, respectively.

Although Epstein indicates that the losses due to relative motion of the liquid and the particle are included in (2-21), the extent of this contribution is not immediately obvious.

Urick (1947) developed an expression that relates the absorption coefficient to the relative motion losses. The identical result is obtained by extending the arguments of Lamb (1945) and also by considering the motion of a pendulum in a viscous liquid. In Urick's notation this is

$$\alpha' = \frac{2}{3}\pi R^3 \left(\frac{\rho_o}{\rho_p}-1\right)^2 \left\{ \frac{s}{s^2 + (\sigma+\tau)^2} \right\} \qquad (2-24)$$

where
$$\tau = \frac{1}{2} + \frac{9}{4}\beta R$$
$$s = \left(\frac{9}{4}\beta R\right)\left(1 + \frac{1}{\beta R}\right)$$
$$\beta = \left(\frac{\omega\rho}{\eta_o}\right)^{1/2}$$

This may be compared to the Epstein value by substituting

$$\beta = \frac{\gamma\omega^{1/2}}{R}$$

with the interesting result that expressions (2-21) and (2-24) are identical. Other approaches have led to an equivalent relation (Angerer, Barth and Guttner, 1951). Thus, as Carstensen and Schwan (1959a) point out, Epstein's concept of mode conversion is apparently a quantitative description of local shear flow of the suspending liquid relative to the particle surface.

The Lamb-Epstein-Urick theory has been found adequate

in describing the absorption in dispersions of rigid

particles such as glass beads (Busby and Richardson, 1956)

and san and Kaolin (Urick, 1948; Stakutis et al., 1955;

Hueter, 1958). Studies conducted in suspensions of polymethyl

methacrylate (Wada et al., 1960), however, reveal an absorption

coefficient somewhat higher than predicted by (2-21) and

(2-24). The disparity has been attributed by Wada et al.,

(1960) to the volume viscoelasticity of the particle with the

contention that the difference between the observed absorption

and that predicted by the Lamb-Epstein-Urick theory is given

by

$$\alpha_E = \frac{K_0 K_P'' C_V \omega}{2 V (K_P')}$$

where $\alpha_E$ is the absorption due to the volume viscosity of

the particle whose complex bulk modulus is determined by $K_P'$

and $K_P''$; $V$ is the velocity of sound in the suspension.

In arriving at this relation, the authors have assumed

that the shear rigidity of the particle is sufficiently

large that the shape deformation by the acoustic is

negligible.

CHAPTER III


EXPERIMENTAL


A.  Pulse Technique

The effective exploitation of ultrasonic spectroscopy
has developed over the last thirty years into a useful
technique for the characterization of matter.  Methodology
employed for liquids is diverse, however, and falls roughly
into two classes; continuous wave techniques and pulse
techniques.

Early investigators (before 1945) employed continuous
wave methods exclusively.  The absorption of a coherent,
plane longitudinal wave train in a liquid can be related
to both its standing wave interferometric behavior and
also to the ability of the wavefronts to act as an optical
diffraction grating (Debye-Sears effect), (Klein et al., 1965).
These techniques are not particularly effective over a wide
frequency range or whenever the attenuation is large
(Hueter and Bolt, 1960).  Because the existence of an
absorption coefficient requires the dissipation of acoustic
energy as heat, temperature fluctuations become a
serious problem when the absorption becomes large.  This
is particularly true of interferometric techniques in
which virtually all the acoustic energy is absorbed in
the sample (Pinkerton, 1949).  There are other problems
with continuous wave techniques (Hueter and Bolt, 1960)
which restrict their usefulness in liquids and as such will

not be dealt with further here.

Pulse techniques were introduced to ultrasonic absorption and velocity measurements in liquids by Pellam and Galt (1946), Pinkerton (1947, 1949), and others as a natural outgrowth of the radar technology developed during World War II. At the basis of this method is the determination of the relative pressure amplitude of a short burst of ultrasonic energy after it has been allowed to propagate for specified distances in a test medium. The generation and detection of pulses can be accomplished in either of two ways; by a transmission method requiring two transducers such that one acts as the source and the other as receiver; or by a reflection method requiring a single transducer and a reflecting surface so that the transducer becomes both source and detector. The immediate advantage of this technique over continuous wave methods is that while the peak intensity of the pulse may be maintained at a high level, the average intensity of the signal propagated in the medium is kept sufficiently small by using a low duty cycle $(10^{-3})$ so that temperature stability problems are avoided. Further, the short acoustic transients that are characteristic of pulsed methods preclude problems that may develop in connection with the appearance of undesirable standing waves. Increased frequency range and less stringent restrictions on the absorption property of the sample are the net result.

There are a number of adaptations of the pulse technique available which permit the exploration of the

frequency range from 300 KHz to 500 MHz (Carstensen, 1954;
Edmonds et al., 1962; Andreae and Joyce, 1962; Hunter and
Dardy, 1965). A single experimental configuration,
however, is limited to somewhat less than two decades of
frequency. The frequency limitations imposed on this
method are essential in absorption cell design consideration
and are discussed below. For the purposes of acoustic
spectroscopy, ultrasonic energy can be both generated
and detected by proper accomodation of the piezoelectric
phenomenon. Longitudinal waves are conveniently produced
from a wafer of quartz, cut perpendicular to the crystalline
x-axis and with metal electrodes deposited on the major
faces (Mason, 1950). Effective transduction of electrical
energy applied to the electrodes to mechanical vibrations
occurs at frequencies such that the thickness of the
wafer corresponds to an odd number of half wavelengths.
Inversely, an oscillatory mechanical stress of the appropriate
frequency results in the development of an electrical
potential of the same periodicity. The relation between
fundamental resonant frequency and thickness for generation
of longitudinal waves in liquids is given (for quartz) by

$$f = \frac{c}{2t} \qquad \frac{2.875 \times 10^5}{t} \quad \sec^{-1},$$

where f is frequency, c is speed of sound in quartz
($5.75 \times 10^5$ cm/sec) and t is the thickness in cm.
Transducers may also be operated at the odd harmonies of
the fundamental frequency. Thus, frequency cannot be

used effectively as a continuous variable and is limited
to discrete values. This is not a serious handicap,
however, since acoustic variables in general do not exhibit
rapid fluctuation with frequency (Litovitz, 1959).

B.  Experimental Design Considerations

    1.  Diffraction

    The diffraction pattern of a plane transducer acting
as a piston source is similar to that of a beam of light
passing through a circular aperture (Kinsler and Frey, 1962).
The field for a source radiating into a semi-infinite
medium is composed of a near Fresnel region and the far or
Fraunhofer field.  Demarcation between these two regions
is usually set at the zero order maximum of the near field
at a distance z from the source such that $z = R^2/2\lambda$
(Hueter and Bolt, 1960) (where R is the radius of the
source and $\lambda$ is the wavelength in the medium).  The
important features of the Fresnel region for the purposes
of pulse measurements are that for a source large with
respect to the wavelength only a small amount of beam
divergence occurs in the Fresnel region and the criteria
for plane waves are satisfied.  In the Fraunhofer region
the beam intensity becomes more diffuse as the main portion of
the beam spreads at an angle $\Theta$ determined by $\sin\Theta = 0.61\lambda/R$.
In an infinite medium, the wavefronts become spherical
at large distances from the source with the intensity
decreasing as $\frac{1}{z^2}$.

    Since the receiving transducers are of finite di-
mensions, the experimental design is such that measurements

are usually confined to the Fresnel region where the
entire beam may be intercepted by the detector. In
practice this restricts the maximum acoustic path length
to less than $R^2/2\lambda$ (Pinkerton, 1949). The requirement
that the radius of the source be large with respect to
wavelength is satisfied by making its diameter at least
50 wavelengths (Nozdrev, 1965). For a cylinderical
geometry with the direction of propagation along the axis,
the requirement of an infinite baffle (semi-infinite
medium) is satisfied by making the radius of the container
at least twice that of the source transducer (Del Grosso,
1965). If this condition is not met, analyses become
complicated by guided wave propagation resulting from
reflections off the walls of the container. If a single
path system is used, the receiving transducer should be
at least as large as the source so that beam spread losses
are kept at a minimum (Andreae et al., 1958). If the
above conditions are met, the resulting diffraction will
be of a free field form (Del Grosso, 1964). At low
frequencies, however, the diffraction losses may still
be large relative to the intrinsic absorption in the
liquid. This is particularly true of water, aqueous
solutions, and other materials with a small absorption
coefficient below about 5 MHz (Del Grosso, 1964). The
rather complete characterization free field diffraction
by Del Grosso (1964, 1965) has made it possible to define
adequately the system under these conditions and apply
suitable corrections, thereby allowing the useful frequency

range of the apparatus to be extended below that where

diffraction losses are negligible.

Satisfaction of the above requirements for free field

diffraction place a nominal lower limit on the frequency

range at which meaningful measurement of absorption can be

made.  For water and other low absorbing materials, this

limit is about 500 KHz (see Appendix II).  In addition,

and more important for the study of some biological

materials is that a minimum sample volume is required.

It is found (Appendix II) that at 1 MHz about 5-10 liters

of sample are needed to determine the absorption coefficient

in water with the volume required decreasing with the

square of frequency.

The volume requirement is a particularly stringent

one in the consideration of some biological materials.  This

is especially true of purified proteins and highly

fractionated macromolecular preparations.  An additional

difficulty is that the absorption coefficient for aqueous

suspensions is usually rather low so that concentrations

of 3 - 10% are generally required for characterization.

Thus the investigation of purified enzyme preparations over

a wide frequency range is prohibited by the expense required

for materials.  There are a few materials like bovine

serum albumin and hemoglobin which are somewhat more readily

available.  It was thus decided that a maximum volume of

about one liter of appropriate concentration could be

tolerated, corresponding to a lower frequency limit of

about 3 MHz.  The low frequency characteristics of the

chamber were thereby fixed and are tabulated in Table 3-1.

TABLE 3-1

LOW FREQUENCY CHARACTERISTICS OF CELL AT

3MHz WITH WATER ($20^{\circ}$C) AS TEST MATERIAL.

| | |
|---|---|
| $\lambda$ | 0.05 cm |
| $R/\lambda$ | 25.4 |
| $R^2/\lambda$ | 32.2 cm |
| $R'/R$ | 3.5 |

MAXIMUM TRANSDUCER SEPARATION 21.3 cm

## 2. High Frequency Limitations

As frequency is increased, the problems associated
with diffraction are alleviated. This follows from the
fact that as the limit ($R^2/\lambda$) of the Fresnel field
extends further into the medium, measurements are made
over a smaller fraction of the diffraction pattern where
the associated losses become negligible. Diffraction
corrections are usually unnecessary above 15 MHz (Carome
and Witting, 1960).

There are, however, a definite set of problems associated
with high frequency measurements. The maximum acoustic
pulse amplitude available is limited by the dielectric
strength of the transducer material and mounting. For
the quartz, the maximum voltage gradient that can be applied
is about 1.5 kv/mm (Nozdrev, 1965). In a Stokes liquid
the absorption coefficient increases with the square of

frequency so that with a limitation of pulse amplitude
measurements at higher frequencies must be made at corre-
spondingly shorter path lengths. With the transducers exposed
directly into the medium, it is found that when the
transit time of the pulse in the medium is less than
about 10 microseconds, difficulty is encountered due to the
time proximity of the rf driving pulse and the received
signal. This is particularly true when the single transducer
system is used since the oscillator and receiver are not
electrically isolated. Even with the double transducer
system it is extremely difficult to prevent some rf pickup
by the high gain receiver. Thus, in order to produce a
suitable time separation of the driving pulse and the
received signal, the procedure of introducing an acoustic
delay line is usually employed. This is accomplished by
attaching the transducers to a rod of low loss material
of the same acoustic impedance in such a way as to increase
the acoustic path length without introducing attenuation
(Andreae, 1958). Generally, fused quartz or silica is used
for this purpose. Application of this technique allows
the path length in the sample to be decreased to virtually
zero without introducing electronic difficulties.

The high frequency limit of the complete apparatus
depends ultimately on the mechanical precision of the moving
parts. 510 MHz has been achieved (Hunter and Dardy, 1965)
by maintaining transducer parallelism to better than
$2.5 \times 10^{-4}$ cm/cm (about one wavelength in their test medium)
and by measuring distance increments of 0.25 microns.

For the purposes of this work, an upper frequency limit of about 170 MHz was decided to be satisfactory. This eases the mechanical precision requirements to the extent that satisfactory parts can be fabricated in the laboratory machine shop and also obviated the need for a separate high frequency chamber. It has been found that with extreme care the final apparatus is capable of satisfactory operation at 200 MHz.

### 3. Velocity

Because of the importance of the absorption coefficient, velocity was given secondary consideration in the design of the apparatus. For the materials of interest here, the magnitude of the velocity dispersion requires that velocity measurements be accurate to at least one part in $10^4$ (Carstensen and Schwan, 1953). Accuracies of better than one part in 30,000 have been achieved (Del Grosso, et al, 1954), but because it would require a sacrifice of versatility of absorption measuring capability, the apparatus was constructed with the usual pulse ranging method of velocity measurement in mind (i.e., an accuracy of about 1%) (Pellam and Galt, 1946). Late in the investigations a somewhat new technique of velocity measurement was discovered which permits an accuracy of about 3.5 parts per 1,000. This method, in which the apparatus is used as an automatic recording interferometer, is found to be extremely useful in cross-checking other characteristics of the system such as distance measurements and temperature stability. Salient

aspects of this interferometric method will be discussed in detail below.

### 4. Temperature Control

The maintenance of accurate temperature control for absorption measurements is particularly crucial when aqueous materials are concerned. This may be attributed to the relatively large temperature dependence of the absorption coefficient that occurs for water (Figure 3-11). If the per cent deviation of the absorption coefficient per degree centigrade ($\frac{1}{\alpha} \frac{\partial \alpha}{\partial T}$ x 100) is computed, it is found that for water at 20°C there exists a 10% per degree dependence. Thus, to preclude an $\pm$ 1% error due to temperature uncertainty, knowledge and control of temperature to $\pm$ 0.1°C is required.

In addition, local fluctuations in temperature may result in refraction of the acoustic beam and lead to deterioration of instrument stability and accuracy (Edmonds, 1966a). Constant mixing of the liquid is generally recommended.

### 5. Semi-Automatic Recording Facility

The effective measurement of absorption usually involves point to point determination of pulse amplitude and acoustic path lengths (see for example, Pellam and Galt, 1946). The amplitude is measured at a large number of increments over an appropriate distance. Resulting points are usually plotted in a semi-logarithmic fashion so that where $p = p_0 e^{-2\alpha x}$ a straight line results with the attenuation per unit path length as the slope. It was found in the preliminary

measurements associated with this study that this technique is both tedious and time consuming and thus prompted a consideration of a semi-automatic recording facility.

Andreae and Joyce (1962) describe an instrument which allows the direct display of the attenuation of the pulse amplitude as a function of distance. This is accomplished by inserting a servo-motor driven piston attenuator in such a manner that the amplitude of the pulse that is applied to the receiver is maintained at a constant level. The attenuator provides a constant attenuation insertion per unit distance and its movement is mechanically coupled to a recorder pen so that the deflection is a direct indication of pulse attenuation. The movement of the recorder paper is linked mechanically to the drive system which governs the acoustic path length so that an accurate correspondence is maintained. Because of the complexity of this apparatus its fabrication was not attempted. Using this system as a starting point, however, a somewhat simpler instrument was developed which differs from the above described apparatus in several respects.

C. Design

1. Mechanical

The instrument is designed to be used in two configurations corresponding to two frequency ranges; a low and intermediate range (LI) from 3-87 MHz and a high frequency range (H) covering 75-165 MHz. Operation in both ranges was accomplished for the most part with the same mechanical components in slightly different orientation.

Appropriate transducer displacement was found to be most conveniently achieved by employing a piston-cylinder arrangement (Edmonds et al, 1962). A piston with a transducer centered on the face (Figure 3-4) is driven within a cylindrical chamber that contains an identical transducer mounted in the base. By using the walls of the cylinder as a guiding surface, it was found that stable and uniform motion can be obtained.

All machined parts which come in contact with the sample are fabricated from stainless steel 316. The chamber (Figure 3-1) is a precision ground cylinder with a bore of 3.5 inches and effective length of 9 inches. The piston assembly is driven vertically within the cylinder. The piston rod is a one inch stainless tube which is precision ground ($\pm$ 0.0002 in.) along its entire length. Both ends of the rod are internally threaded so as to accomodate a transducer assembly at one end and an actuator screw at the other. The rod has a half inch internal diameter to allow the passage of RG-62-U cable and associated rf connectors to effect a stable electrical connection to the transducer. A small transverse hole near the top provides the necessary cable exit.

The piston head is a cylinderical block of stainless steel having a length of two and one half inches. Because stainless steel provides a notably poor bearing surface, Teflon O-rings are employed in order that a small clearance with the cylinder wall could be tolerated without fear of mechanical seizure. A one inch diameter hole bored along
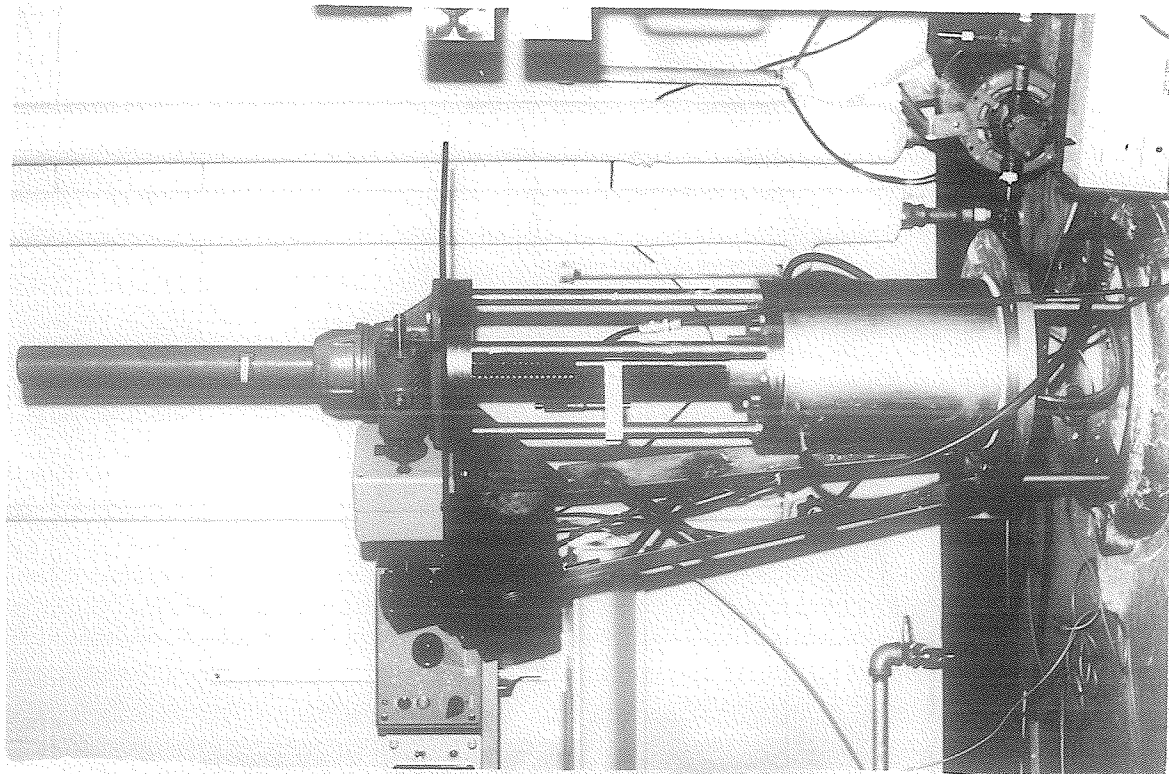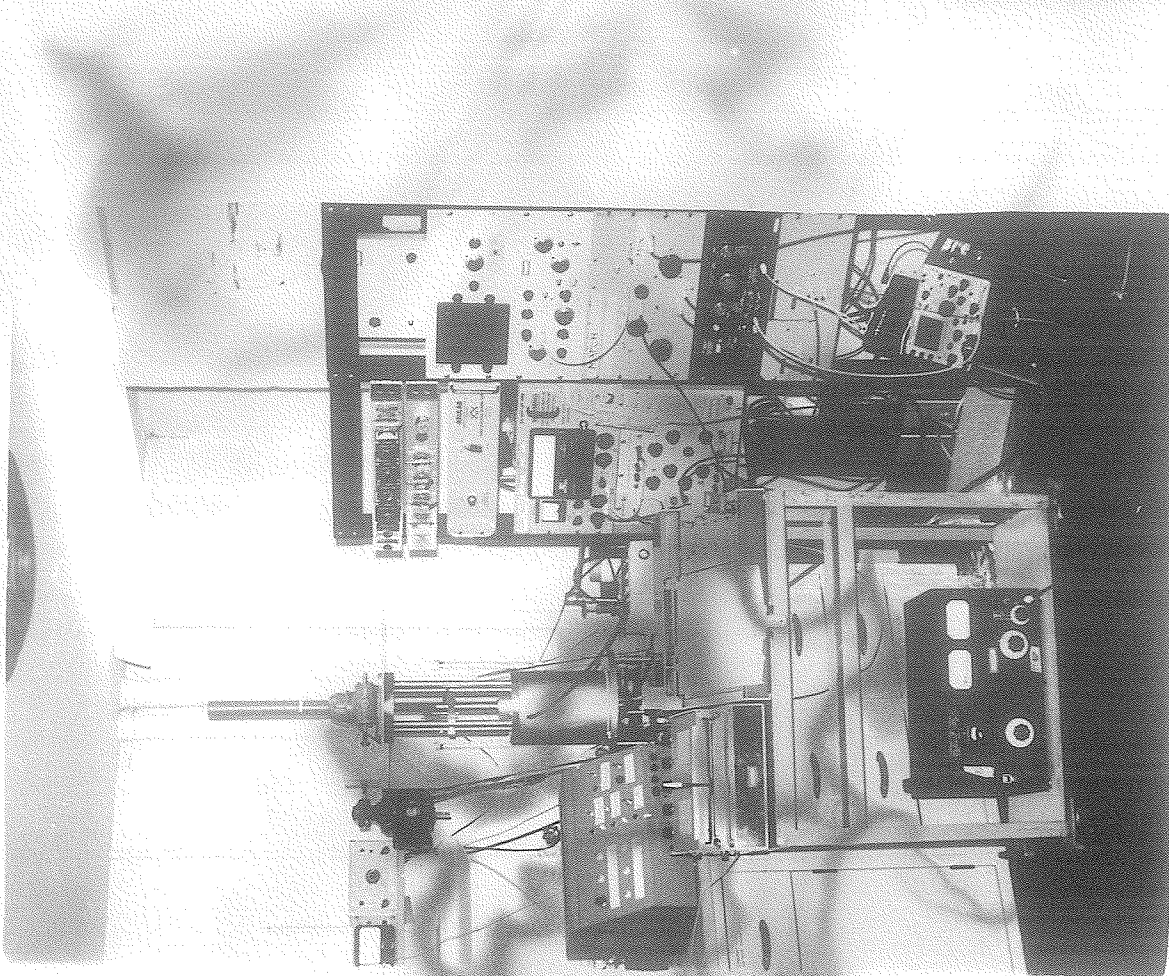
FIGURE 3-2 ABSORPTION CHAMBER



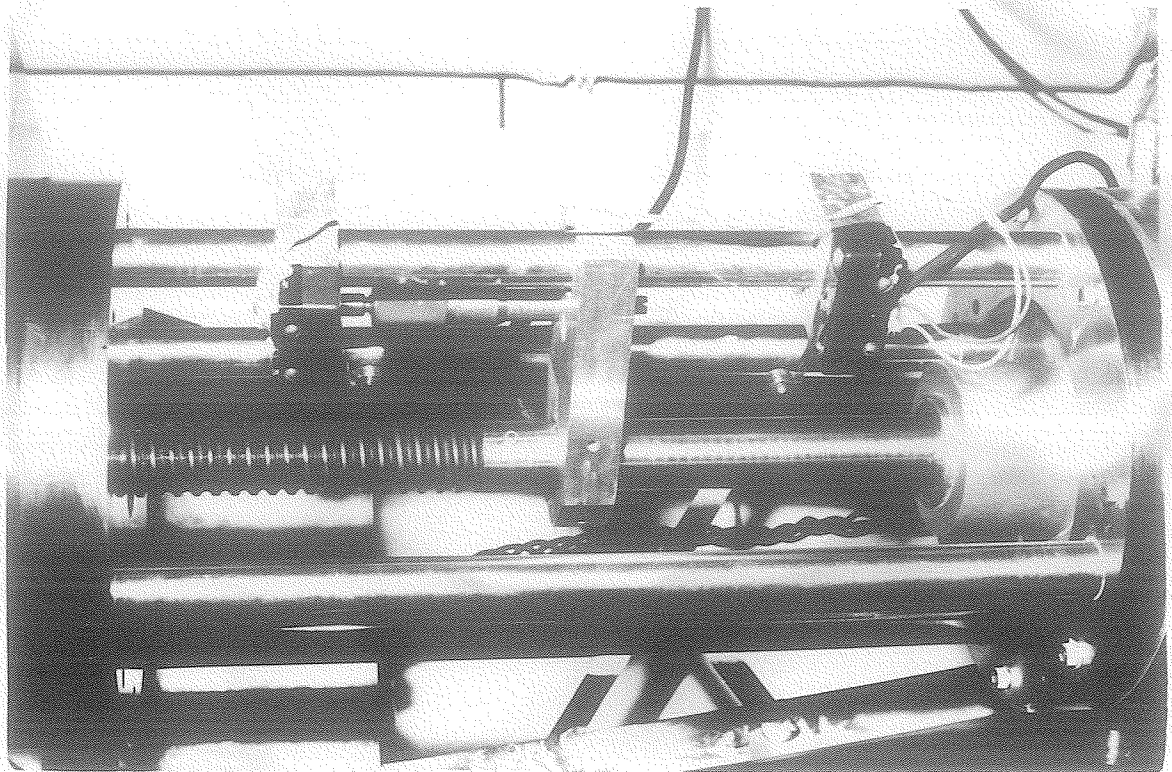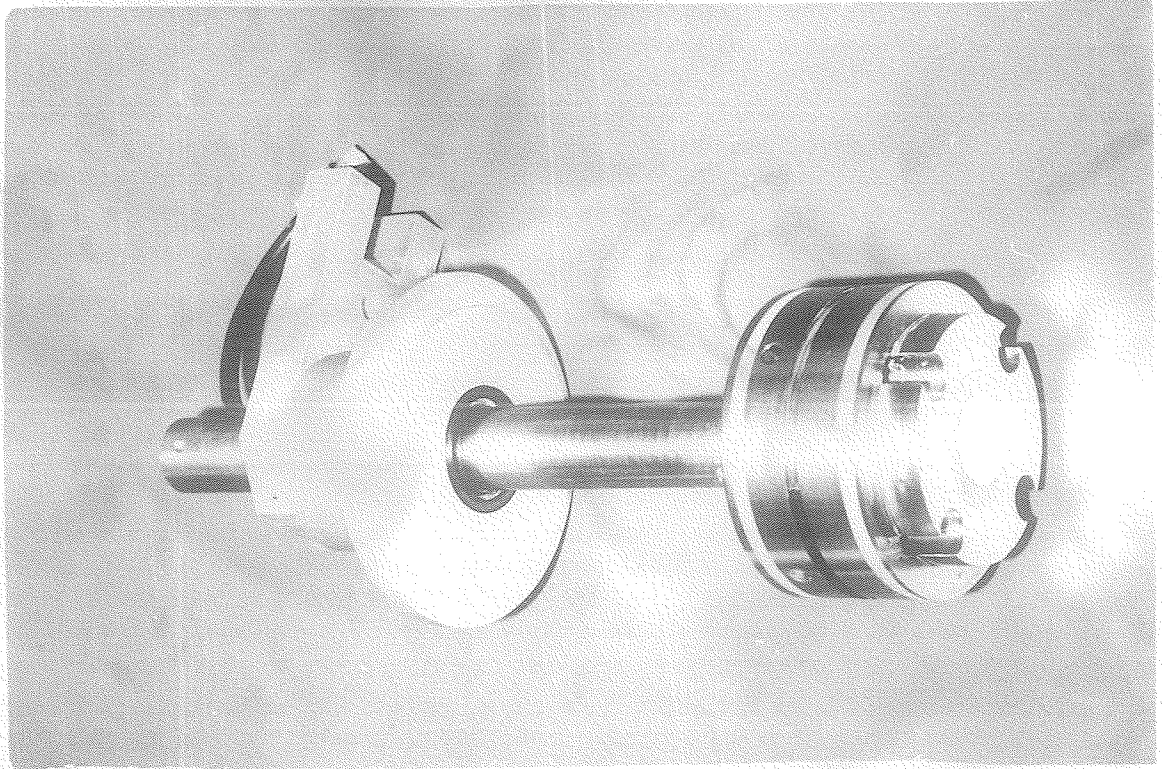FIGURE 3-1 ABSORPTION DETERMINING APPARATUS

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FIGURE 3-4 PISTON ASSEMBLY (LI) CONFIGURATION

FIGURE 3-3 DETAIL OF ACTUATOR - PISTON ROD
JUNCTURE. SHOWING ANTIROTATION
GUIDE AND REMOTE SWITCHES.

the axis of the piston head accomodates the piston rod. A counter-bored recession along the exterior of this hole receives the transducer housing. The piston head is rigidly fixed in position on the rod by the transducer assembly on one end and an adjustable lock nut on the other.

The transducer housing provides a sealed unit with an rf connector to provide access to the crystal. Contact to the internal crystal electrode is accomplished with a short length of beryllium-copper wire with a small solder bead on the end. The length of the wire is adjusted to be just long enough to provide a stable mechanical contact when the connector is turned tightly into the housing. The solder bead inhibits scratching of the surface electrode.

The upper end of the piston rod is threaded into the lead screw of an actuator unit[1]. The actuator provides transformation of rotary motion to linear displacement via a worm gear and geared lead nut. Reworking of the actuator components was found to necessary to eliminate lateral motion of the end of the lead screw. Additional assurance that lateral forces do not act upon transducer alignment is guaranteed by the use of a bushing at the point of entry into the chamber by the piston. A ball bushing[2] is used so that difficulties with sample contamination by lubricants are avoided. An anodized aluminum housing supports the bushing and permits alignment with the top inside surface of the cylinder, thereby serving the additional function of chamber cover. A rubber seal prevents fouling of the bushing by liquid adhering to the rod. A

small vent provided accomodates a thermometer and also

prevents the development of internal pressure.

The actuator is supported fourteen inches above

the absorption chamber by four 3/4 inch stainless pillars.

Rotation of the lead screw is restrained by a notched

aluminum block fixed to the piston rod in such a way that

vertical motion is guided by one of these pillars. The

aluminum block serves an additional function as support

for a micrometer with which small vertical displacements

can be accurately measured.

A Bodine 1/25 HP synchronous, four lead, capacitor

motor[3] is used to drive the actuator. The motor is

reversible and includes an internal gear reducer so that

a speed of 180 rpm at 11.4 lb-in. is obtained at the output

shaft. Connection to the actuator occurs through a variable

speed gear box which permits speed reduction in ten integral

ratios from 1 : 1 to 200 : 1. The actuator provides one

inch of linear displacement for every ninety-six revolutions

of the input shaft so that ten piston speeds between 1.875

in/min to 0.009375 in/min are available.

The direction of the motor is controlled both at a

remote panel and by switches that may be mounted optionally

on the vertical pillars, activated by the micrometer

support (Figure 3-1). The use of these switches permits the

operation to be self-cycling over a predetermined displacement.

Rigid mechanical connections couple the motor shaft to

the piston rod so that the displacement may represent the

synchronous action of the motor as nearly as possible. The ratio of the output torque to that required to turn the actuator shaft under load condition is about 5 : 1. This load condition was selected to prevent slippage of the armature relative to the driving field.

In the high frequency range a reassembly of components is necessary to provide the additional precision required. Although the piston-cylinder configuration is retained, the guiding surface for the transducer displacement is the precision bored hole in the piston block (Figure 3-6) that is also used for the (LI) frequency range measurements. This block is inverted and placed near the bottom of the chamber.

The transducer assemblies are each composed of a 3/4 in. diameter fused quartz delay rod and associated quartz transducer mounted in a stainless steel housing. Matched transducers, 1.5 in. in diameter, with a fundamental frequency of 15 MHz are employed. The rf connectors are fitted to the housing and electrical contact is made in the same manner as described previously. The housings are machined to provide a precision ($\pm$0.0002 in.) sliding fit for the delay rods which are sealed in place by a thin filet of epoxy resin. Because of the difference of thermal coefficients of expansion between quartz and stainless steel these assemblies are not usable much below room temperature. The lower transducer remains stationary and its alignment is maintained by penetration of the housing through the ball joint and 0.5 in. into the guiding bore.

TO ACTUATOR

PILLAR

VENT

BALL
BUSHING

SEAL

PISTON ROD

LOCK NUT

TEFLON
O-RINGS

PISTON
HEAD

BYPASS

RF CONNECTOR

PORT

X-TAL

BALL SOCKET
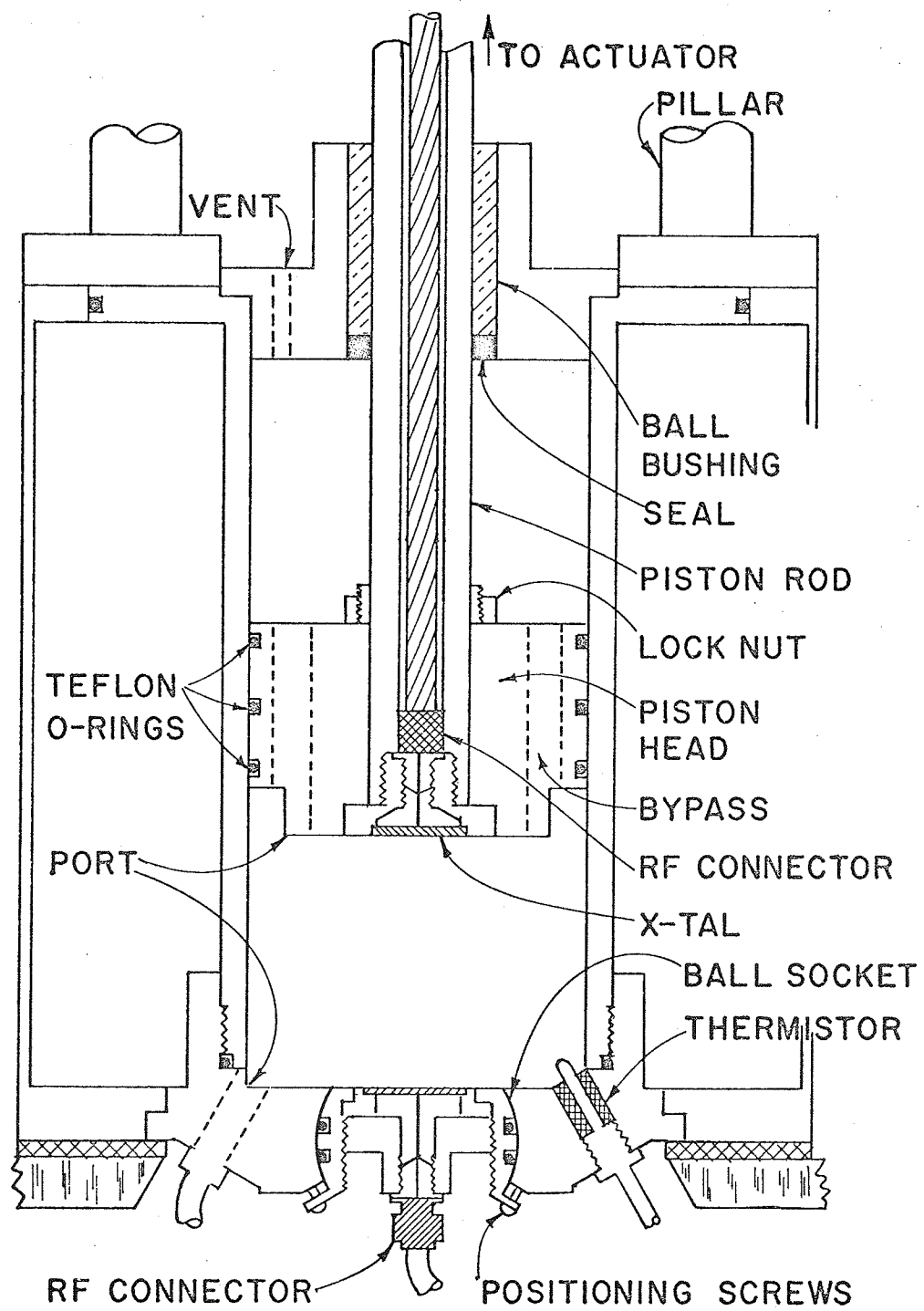
THERMISTOR

RF CONNECTOR

POSITIONING SCREWS

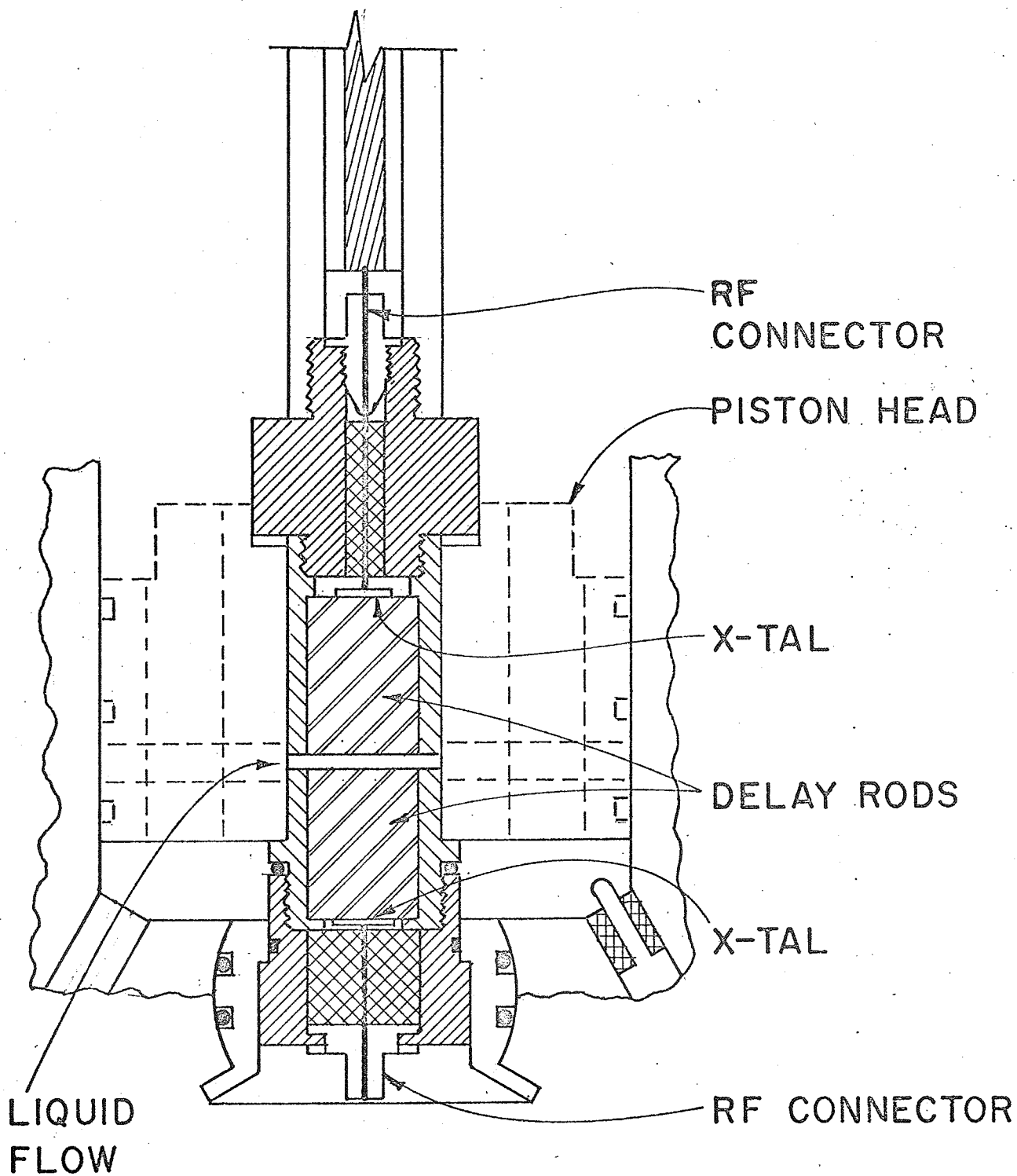FIGURE 3-5   DETAIL, ABSORPTION CHAMBER (LI) CONFIGURATION

FIGURE 3-6   DETAIL OF (H) CHAMBER CONFIGURATION

The upper transducer housing is driven in the same
manner as described above. Smooth translation of the rod is
maintained by the use of a single O-ring about the exterior
of the housing.

2. Transducers And Transducer Mounting

Optically polished X-cut quartz crystals are obtained
in matched pairs ($\pm$0.01%) from the Valpey Crystal Corporation,
Holliston, Mass.. Mounting of the crystals for the low
and intermediate frequency measurements is similar to that
described by Carome and Witting (1960). The transducers
are edge-supported by a thin annulus of epoxy resin and a
narrow (0.005 in.) guide ledge counter-faced into the holder.
Chromium on gold electrodes are electro-deposited to the
faces of the quartz. With the transducer fixed in the
housing, the exposed face is electro-deposited to form a
continuous contact to the holder. The 15 MHz transducers
are cemented to optical grade, polished, fused, quartz rods
also obtained from Valpey Crystal Corporation. The end
and cylinderical surface of the delay rod is electro-deposited
prior to the fixation of the transducer to provide a
contact with the hidden surface of the transducer. A
small drop of resin is applied to the rod and squeezed
out while wringing the two surfaces together. Satisfactory
mating of the surfaces was determined acoustically at
285 MHz using a Matec PR201 Attenuation Comparator. The
generation of a suitable bond results in an exponentially
decreasing echo pattern with negligible echo deterioration
near the baseline. When a good bond is achieved, the crystal

is held in place under pressure with a Teflon die and the assembly cured at $150^{\circ}C$ for ten hours.

### 3. Electronic: Principle Of Operation

Figure 3-7 is a block diagram of the arrangement of the electronic components used for absorption and velocity measurements. The sequence of events begins at the pulsed oscillator with the formation of a rf modulated pulse with a peak to peak amplitude of 10 - 200 volts. The pulse is applied to the quartz transducer via a network that permits the output impedance of the oscillator to be properly matched to that of the crystal. A variable attenuator is inserted between the oscillator and the source in instances when the minimum stable voltage available at the oscillator is too great to be tolerated by the receiver. The ultrasonic pulse is generated, detected and the small resulting rf signal ( < 50mV) is sent to an electronic receiver. At frequencies below 10 MHz, a broadband receiver is used and above 10 MHz a high gain heterodyne receiver is employed. After appropriate amplification, the pulse is detected and rectified so that only the pulse envelope remains for further analysis. This signal is then applied to the input of a coincidence gate. The portion of signal that is allowed to pass the gate is determined by the coincidence in time by a square wave pulse from the pulse generator which is triggered at a variable time after the rf driving pulse. The important feature of this device is that it permits the removal of undesirable events from the signal sequence.
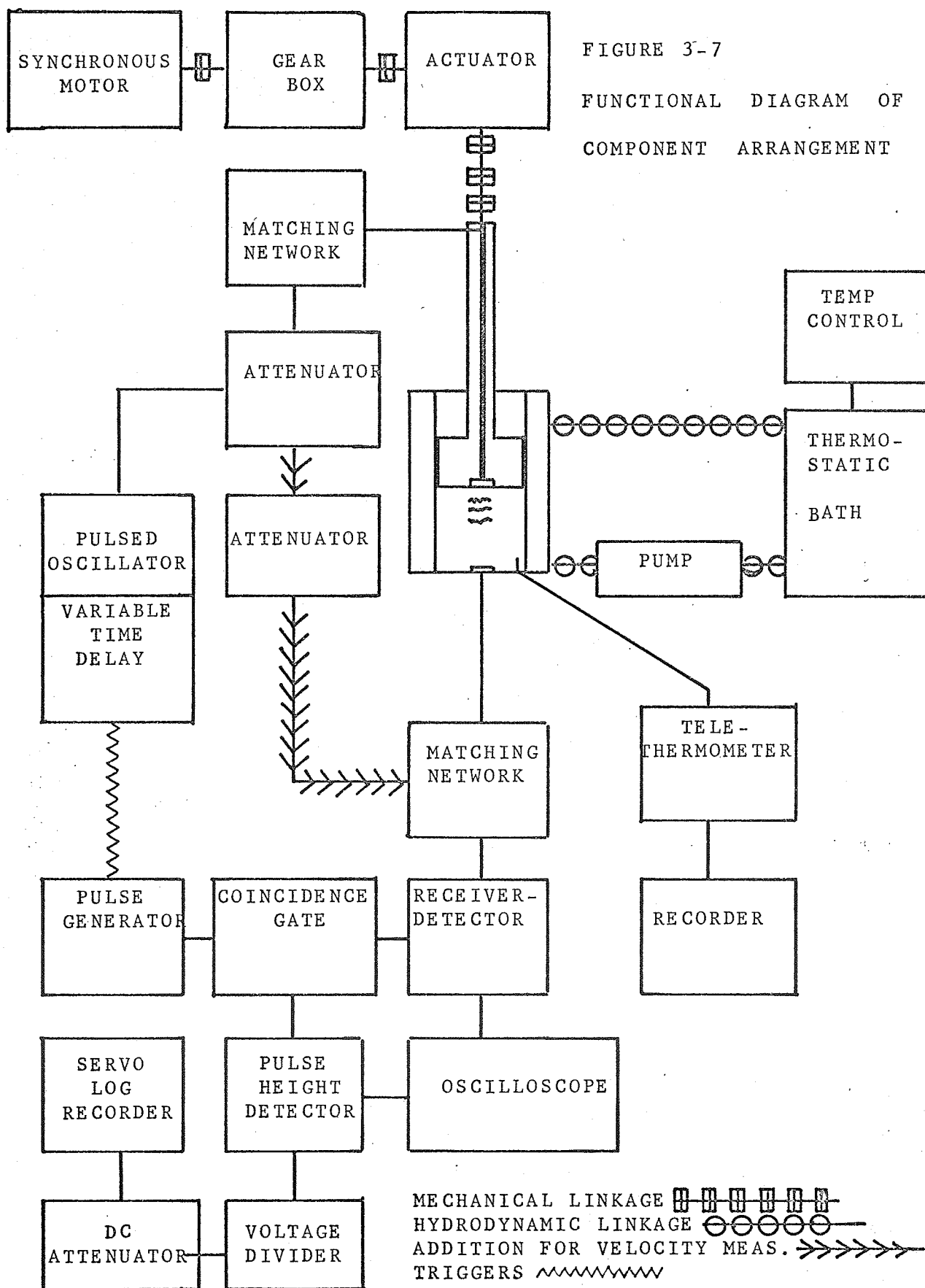
FIGURE 3-7

FUNCTIONAL DIAGRAM OF

COMPONENT ARRANGEMENT

Particularly of interest here is the elimination of rf feed-through that may occur. The inclusion of the gate permits measurements at higher frequencies than would normally be possible. By varying the gating pulse length and the time delay, it is possible to "gate out" for analysis all or any portion of the signal train. At low frequencies, where a number of echos are present, the analysis and tracking of any echo is easily accomodated.

The maximum voltage that is allowed to appear at the output of the gate is determined by the amplitude of the gating pulse. Appropriate selection of the pulse voltage makes it impossible to overload subsequent detecting devices. The pulse envelope is next fed to a pulse height detector which produces a dc voltage proportional to the amplitude of the largest pulse in the signal sequency, independent of other repetitive signals that may be present. Thus, when a multiple echo pattern appears at the output of the gate, it is not necessary to track continuously the first echo but merely to remove the rf feed-through. The input to the detector ranges from about 0.2 to 10 V and the voltage gain is -1, so that a fixed dc attenuator is necessary to reduce the signal to a level appropriate ($\sim$50 mV) for the recorder. A servo-logarithmic recorder is employed with a variable dc attenuator at the input. Precise ($\pm$0.01dB) units of attenuation may be inserted here in order to calibrate the recorder scale.

The measurement of the attenuation in a liquid is

accomplished by changing the separation of the transducers

at a constant speed so that a straight line is generated at

the recorder with slope of $\pm\alpha$ depending on the direction

of the displacement.

Pulse lengths from 0.5 to 20 microseconds are employed,

depending upon frequency and acoustic path length. For

absorption measurements, the criteria that the pulse length

be compatible with the band width of the transducer (Nozdrev,

1965) is always observed, i.e.,

$$T < \frac{2}{\Delta f} = \frac{2Q}{f_o}$$

where T is pulse length, Q is the mechanical quality

factor, $f_o$ is the center frequency and $\Delta f$ is the bandwidth.

The frequency of maximum acoustic response is deter-

mined by a variation of method recommended by Arenberg

(1966). A digital counter is used to resolve the frequency

of a signal generator signal necessary to produce a zero

beat with the received acoustic pulse. It is found that

for both sets of transducers the use of maximum acoustic

response to determine the frequency of operation is suffi-

ciently accurate ($\pm 0.2\%$) so that precise determination

before each measurement is not necessary.

4. Electronic: Components

PULSED OSCILLATORS: A model PG-650-C pulsed oscillator

obtained from the Arenberg Ultrasonic Laboratory, Jamaica

Plain, Mass. is employed as the rf modulated pulse source

for the range between 3 MHz and 105 MHz. In addition,

this instrument provides the time base and associated
trigger pulses for synchronizing the other electronic
components for all measurements.  Pulse widths are
continuously variable from 0.5 to 20 microseconds.  Pulse
amplitude is adjustable from 0 to about 600 volts into a
93 ohm load and the pulse repetition rate is variable
from 50 pps to 2400 pps utilizing the internal triggering
circuits.  Double pulses can be obtained in phase stable pairs
of identical duration and adjustable time separation from
20 to 11,000 microseconds.  Both direct and delayed trigger
outputs are provided.

For frequencies above 105 MHz, the source of the rf
pulses is an Ultrasonic Attenuation Comparator model PR-201
manufactured by Matec Inc., Providence, R. I., (see Chick
et al, 1959 for details of circuitry of this instrument).
This device contains pulsed oscillators, superheterodyne
receiver, and video display circuits for ultrasonic
measurements between 1 and 380 MHz.  The oscillator provides
variable (0 to about 100 volts, peak to peak) amplitude
pulses into 50 ohms.  Discrete pulse widths of 0.5, 1, and
2 microseconds at a repetition rate of 100 pps are provided
by the instrument as supplied from the manufacturer.
Longer pulses can be obtained by substituting additional
pulse transformers with correspondingly smaller pulse ampli-
tudes.  Higher repetition rates are most conveniently achieved
by triggering the oscillator externally.  Although the PR-201
provides an input for an external triggering mode, it is

found that stable operation is only accomodated by disconnecting the internal triggers. Because of the limitation in pulse length, the Arenberg is the preferred pulse source and is used exclusively below 100 MHz.

ATTENUATORS: Two 50 ohm attenuators with ranges of 0 to 12 dB and 0 to 80 dB manufactured by Daven Div., Manchester, N. H. are employed in situations where 50 ohm terminations are required. Ninety-three ohm attenuators manufactured by Arenberg Labs with a range of 0 to 122 dB are used for corresponding 93 ohm loads. Both types of attenuator have rated uncertainties at $\pm$ ½ dB (1%).

MATCHING NETWORKS: Approximate matching from 3 MHz to 42 MHz is achieved by insertion of a tuned transformer section in parallel with the transducer. By operating the transformer in a partial resonance condition, i. e., resonating either primary or secondary with a shunt capacitance, an appropriate "magnitude" impedance transformation may be accomplished (see for example, p. 435, Everitt and Anner). Rotary air core inductors provide a convenient autotransformer with a continuously variable tap. Either of two transformer sections are used at each transducer to provide matching up to a frequency of about 42 MHz, limited by the self-resonance of the coils. At higher frequencies, matching is accomplished with sections of coaxial cable of various lengths in conjunction with double-stub tuners.

RECEIVERS: A Matec PR 603 broadband untuned receiver is employed at frequencies below 10 MHz. This unit provides nearly 70 dB gain. Above 5 MHz the internal superheterodyne receiver of the Matec PR 201 is used (Chick et al, 1959). This receiver employs a synchronously tuned, four stage 60 MHz i.f. section with a gain of about 80 dB and bandwidth of 1.5 MHz. In order to prevent stray signals generated by the local oscillator from entering the i.f. strip, a variable and a fixed frequency trap are employed at the output of the rf mixer. The variable trap tracks the local oscillator frequency and the fixed trap is set at 65 MHz, the lower limit of the local oscillator. Thus, operation of the receiver is avoided at frequencies where this trap is active (63 MHz).

COINCIDENCE GATE: A simple two transistor, two diode logic module is employed as the gate (Figure 3-8). The emitter-follower ($Q_3$) was included as part of the preliminary design to provide sufficient current gain so that peak rectification techniques of determining pulse amplitude could be investigated. The emitter-follower is not essential when the gate is followed by a high impedance such as that provided by the pulse-height detector described below. Signals with amplitudes between 0 and 10 V are conveniently handled by the gate with a common mode of two positive coincident signals appearing at the output. In addition, the signal of interest is accompanied by a pedestal of about 1 V. The pedestal, which is characteristic
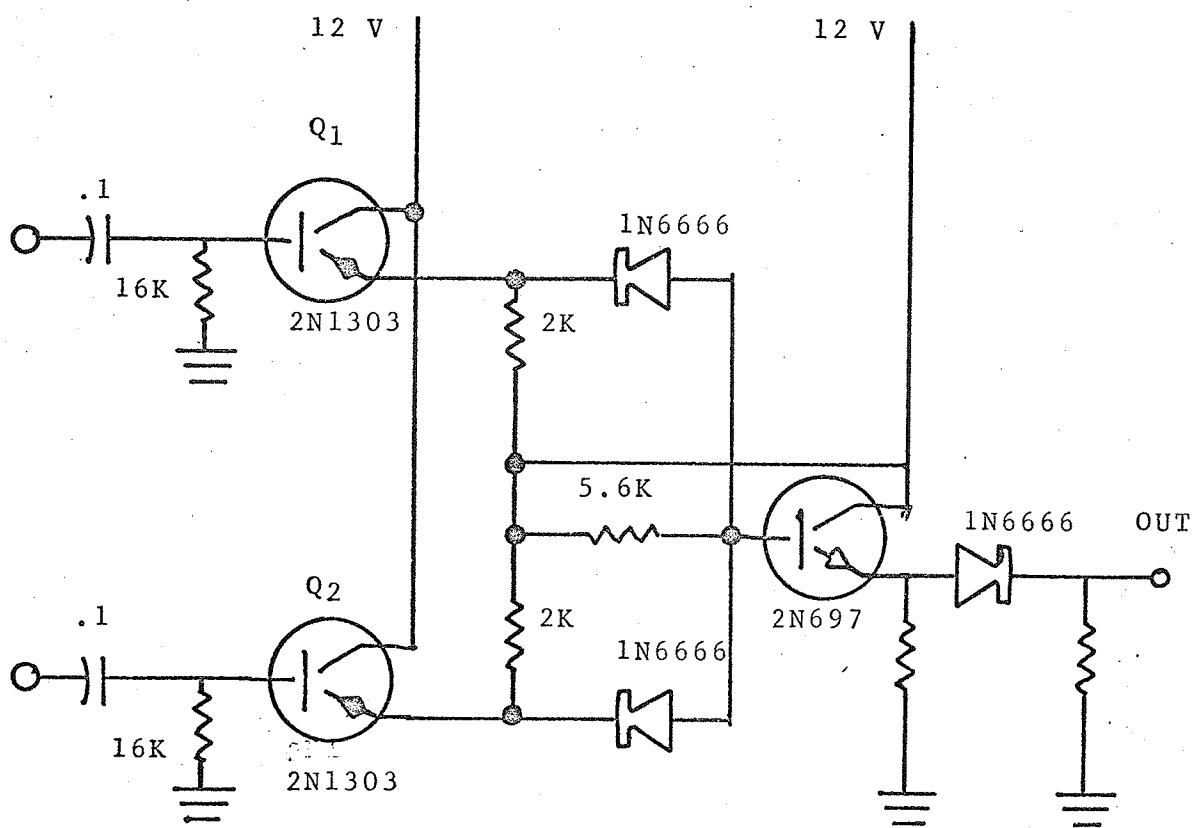
Figure 3-8: COINCIDENCE GATE

of this type of gate, carries the entire signal at sufficiently high potential such that difficulty with threshold devices (diodes, etc.) is obviated at small signal amplitudes.

PULSE HEIGHT DETECTOR: Detailed description of this circuit and its characteristics may be found elsewhere (Sylvan, 1963; G. E. Transistor Manual, 1964). Basically, this is a peak-sensitive operational amplifier which accurately transforms pulse amplitude to dc, utilizing a self-adjusting bias in a tunnel diode threshold sensing element. A high degree of flexibility in circuit parameters is possible. The pulse height detector used in the present investigation has the following characteristics: input impedance, 100 k$\Omega$ ; voltage gain, -1; output ripple voltage less than 40 mV peak to peak; output impedance, 10 k$\Omega$ . The bias current of the tunnel diode is adjusted so that the output voltage is zero when the pedestal voltage of the gate appears at the input.

OSCILLOSCOPE: A Tektronix Type 453 transistorized oscilloscope is employed. This instrument has dual channels each having vertical amplifiers with a dc to 50 MHz bandwidth. For the most part, the oscilloscope is used to monitor the position relationship in the time domain of the various signals. One particularly useful feature is the ability to display the sum or difference of two signals with common mode rejection ratio of nearly 100 : 1 for signals up to 5 MHz. Thus, if one observes the difference

mode of the signal at the input and output of a circuit

element, e. g., the pulse height detector, appropriate

adjustment of the relative channel gain will produce a

null for a linear element.  In addition, the dynamic

range may be indicated by the dependence of the null on

signal amplitude.

PULSE GENERATOR:  A Type 1217-C General Radio Co.,

Unit Pulse Generator is employed as a source for the gating

pulse.

RECORDER:  The acoustic data recorder is a Sergent

Model SRL, which displays the electric potential as a

function of time.  Conversion of the signal to the logarith-

mic form is accomplished mechanically with single cycle

(20dB) logarithmic gears.  Full scale deflection is 24 cm

and the maximum balancing speed requires one second for

full scale travel.  Full scale potential is variable from

1 mV to 120 mV.  The chart paper is driven by a three

speed synchronous motor at 0.2, 1, and 5 inches per minute.

Deviation from a true logarithmic response is of the order

of the width of the pen line ( $\pm$ 0.03dB).  Calibration of

the scale is most conveniently achieved by insertion of

precisely ( $\pm$ 0.01dB) known dc attenuation units at the

input of the recorder.

5.  Temperature Control And Measurement

Maintenance of constant temperature is achieved

by the circulation of water from a 15 liter thermostatic

bath through the annular space about the acoustic chamber. The bath temperature is controlled using a thermistor in conjunction with a temperature controlling unit and a 1,000 watt immersion heater balanced against the circulation of $16^{\circ}C$ tap water through a heat exchanger. The control thermistor is imbedded in the bath liquid rather than the sample chamber to avoid large oscillations that result from the high thermal mass of the latter.

A second thermistor sealed in a stainless steel fixture is mounted in the base of the chamber so that the tip extends about ½ inch into the sample. This temperature is monitored by two means: 1) by a meter reading, resistance bridge which allows direct observation of the temperature to $\pm 0.2^{\circ}C$ and 2) by a servo recorder which provides a continuous record of the temperature with accuracy of $\pm 0.03^{\circ}C$. Both of these systems are standardized against a National Bureau of Standards calibrated thermometer.

The sample may be continuously mixed by circulation through an external metering pump. Although it is not desirable to remove the liquid from the chamber, circumstances did not permit the inclusion of a somewhat more elaborate internal mixing scheme. Since practically all of the investigations are carried out at temperatures near that of the external environment, this method did not cause a significant loss of stability. It is found that with the above scheme the temperature can be maintained

and determined with an accuracy of $0.01^{\circ}C$ at $20.0^{\circ}C$.

## D.  Determination Of The Absorption Coefficient

At frequencies above 3 MHz the absorption coefficient is determined directly from the slope of the recorder tracing.  In order to correct for diffraction losses, it is assumed that the intrinsic absorption and that due to diffraction are noninteracting and additive (Del Grosso, 1964).  In addition, it is found at frequencies above 3 MHz in the suspensions studied that the attenuation attributable to the presence of suspended matter, $\alpha_1$ , and that due to the suspending fluid, $\alpha_0$ , are also additive (Chapter IV).  Thus, the total absorption, appears to comprise three terms, viz.,

$$\alpha_T = \alpha_1 + \alpha_0 + \alpha_D(x)$$

In the solvent $\alpha_1 = 0$    so that

$$\alpha_{T,0} = \alpha_0 + \alpha_{D,0}(x)$$

The diffraction losses at a given point in the suspension, $\alpha_D(x)$ , and in the solvent, $\alpha_{D,0}(x)$  , will be the same if the wavelengths are identical.  It is found in the suspensions investigated that $\alpha_T/\alpha_{T,0}$ increases with concentration more rapidly than the ratio of wavelengths. Typically, at a concentration of 10% it is found that $\alpha_T/\alpha_{T,0} \approx 8-10$ while the difference in wavelength has changed about 4% (Chapter IV).  Thus, by assuming

$\alpha_{D,o}(x) \approx \alpha_D(x)$, the error introduced by this assumption

will always be small relative to the total absorption.

Therefore,

$$\alpha_I = \alpha_T - \alpha_{T,o}$$

and the curve obtained by subtracting the absorption trac-

ing in water from that in the suspension will yield the

desired parameter.

### E. Velocity Measurements, Tracking Error, And Temperature Stability Characterization

In the early stages of this work, velocity was deter-

mined by measurement of the time delay of an acoustic

transient occuring in a sample of known thickness (Pellam

and Galt, 1946). This technique has the intrinsic limitation

that group velocity rather than phase velocity is the para-

meter being examined (Del Grosso, 1964). This difficulty

proved to be problematical, however, as the error that

arose experimentally ($\pm$ 1%) proved to be more than an order

of magnitude greater than those introduced when dispersion

is neglected. A more interesting technique was discovered

in the course of investigation which permitted a somewhat

more accurate measurement of the velocity as well as

characterization of tracking errors (deviation from linear

correspondence between piston displacement and paper move-

ment) and temperature stability.

The arrangement of the electronic and mechanical

components is basically the same as that for the low and

intermediate frequency absorption measurements. An additional

attenuator provides a direct rf path (see Figure 3-7) from

the oscillator to the receiver. The oscillator is operated

in a double pulsed mode with the time delay between pulses

adjusted to be nearly equal to the time required for the

acoustic pulse to complete a transit in the absorption cell.

If this condition is satisfied, the first pulse traveling

through the absorption chamber will arrive at the receiver

input at the same instant in time as the delayed pulse

through the shorting attenuator (Figure 3-9). If the ampli-

tudes are equal, the resulting phase interaction in the

region of overlap is such that a null is produced whenever

the difference in phase angle $\delta$ between the rf modulations

is $180^\circ$. The relative phase is directly determined by the

delay of the acoustic pulse in the sample such that phase

cancellation occurs whenever the transducer separation is

varied through one wave length. Thus, by using the recorder

to depict the phase interaction, an interference pattern is

generated as the piston is driven through the sample (Figure

3-10). Since the acoustic wavelength in the medium is

constant, any nonuniformity in the distance between minima

represents the inaccuracy in the tracking system. The

velocity is then determined from the knowledge of wavelength

and frequency to an accuracy limited by this nonuniformity

only.

If the pulse height detector is to be used to detect

the null for purposes of automatic recording, it is essential

that the non-zero components of the signal be removed. This

DIRECT SIGNAL THROUGH ATTENUATOR

INCREASING TIME

DELAYED SIGNAL
THROUGH SAMPLE

←———TIME DELAY———→

SIGNALS MIXED $\delta = 180°$

NULL

SIGNALS MIXED $\delta = 0°$
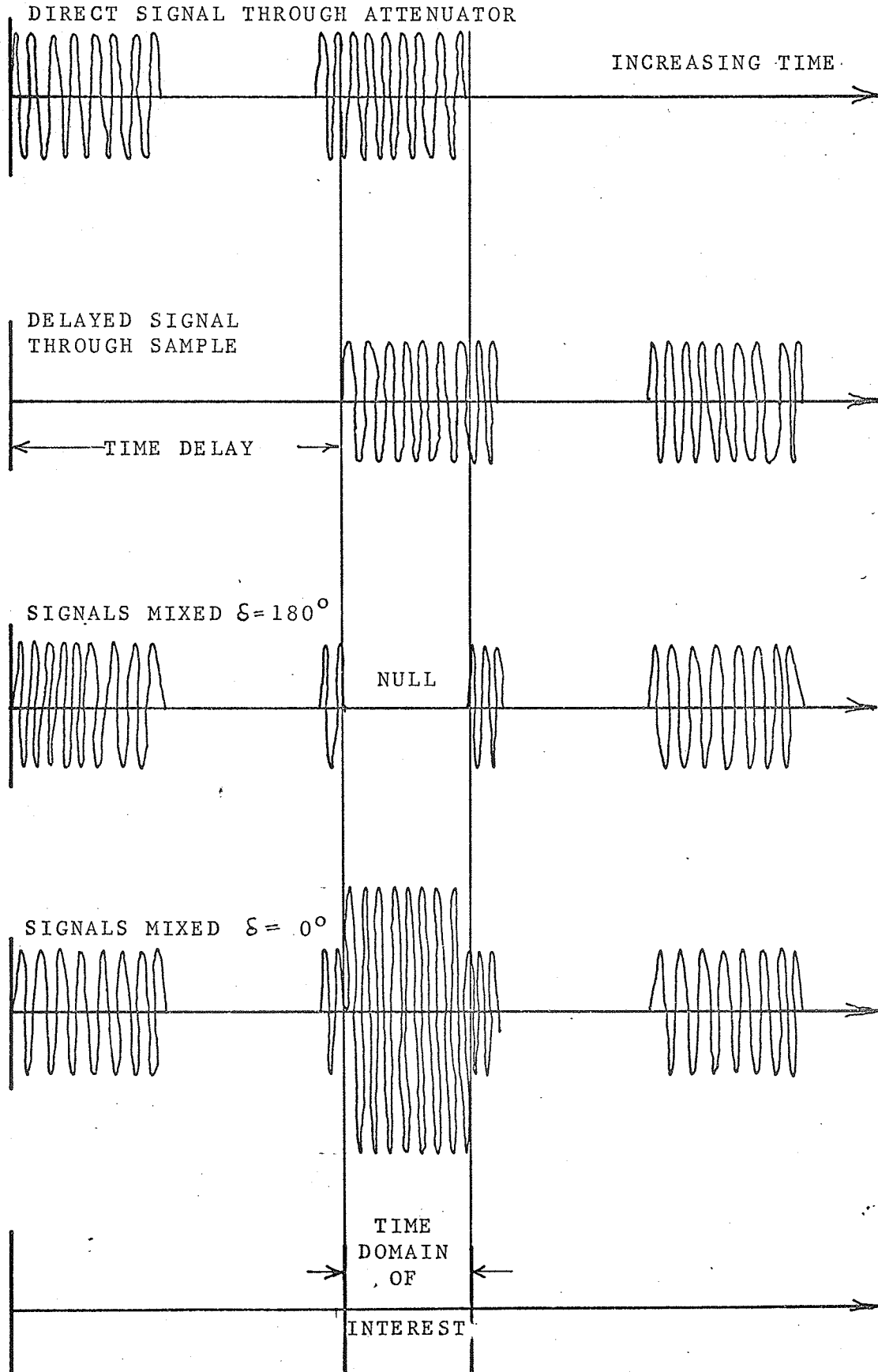
TIME
DOMAIN
OF
INTEREST

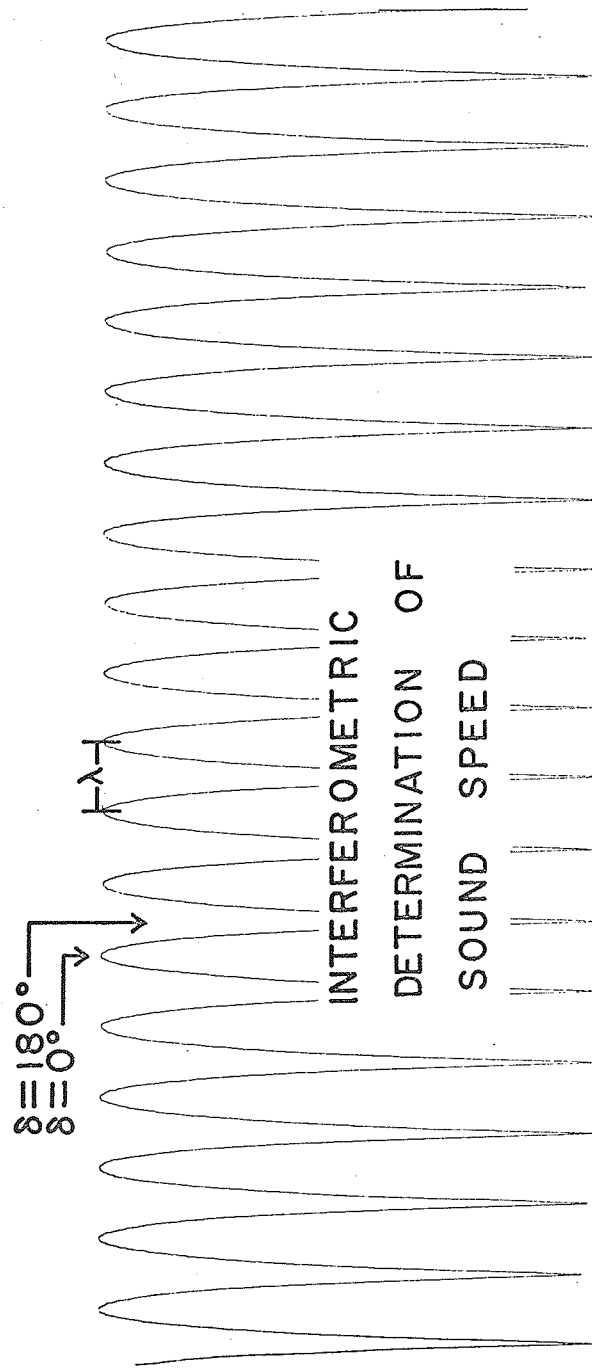Figure 3-9: PULSE CONFIGURATION; VELOCITY MEAS.

FIGURE 3-10: INTERFEROMETRIC DETERMINATION OF SPEED OF SOUND IN WATER; RECORDER TRACING, REDUCED TO ONE-THIRD ACTUAL SIZE.

is accomplished in the same manner as for absorption measurements. An additional variable time delay, however, is required to control the time position of the delayed rf pulse from the oscillator. The Arenberg pulse oscillator, which is employed for these measurements, permits a maximum pulse length of about 20 microsecond. At 3 MHz this allows a 60 cycle pulse to be used. Ideally, one would like to examine the interference of two long pulses over a large number of cycles. At 3 MHz a compromise is necessary between the length of the overlap and the number of nulls examined. A 30 cycle overlap is used over about 30 cycles. Interferometric patterns up to 21 MHz have been produced in the present instrument. Above 9 MHz, however, jitter in the relative time and phase between the two pulses produced at the oscillator becomes comparable to a cycle so that the sharpness of the null becomes impaired. The extent of the jitter, estimated on the basis of the interference pattern, is found to be less than a quarter of a cycle at 15 MHz or about 15 nsec. Stability of this magnitude is representative of that which can be achieved with a double pulsed system.

The use of a gated amplifier in such a way that the acoustic pulse is mixed with the modulating rf is better suited for this technique. Since a portion of a continuous rf signal is compared to itself, problems with pulse jitter and finite overlap could be, to a large extent, eliminated. The present oscillator does not include this facility, and this particular aspect of the experimental technique was not

pursued further.

The use of the instrumentation as an interferometer
allows the investigator to determine accurately the error
encountered when the recorder is assumed to be an accurate
transducer of piston displacement. In the course of velocity
determinations, measurements were made a 3 MHz and over
several wavelengths so that the error introduced by the
finite width of the null is minimized. By variation of
speed reduction, it is possible to separate the contribu-
tions to the error by the actuator from the gear box-motor
combination. The acoustic system was thus found to have
a distance uncertainty associated with it of approximately
3.5 parts per thousand. Virtually all of this uncertainty
was attributable to the actuator. Additionally, it is
found that the error is introduced as a complex oscillatory
deviation with a period corresponding to about 3 cm.

Since the velocity of sound in aqueous liquids has a
relatively high temperature coefficient, the interferometric
technique described above allows a means for characterizing
the temperature stability of the system. One may question
the advantage of making such a determination when means for
thermistor measurement of temperature has already been
provided. The justification resides in the fact that the
acoustic method is a reflection of the average temperature
of a large volume of medium where as the thermistor is
representative of a single point only. The technique is as
follows. With the acoustic path length set at a fixed

distance, the phase difference of the interfering pulses is adjusted so the dependence of the recorder response on wavelength is maximal. This occurs at a phase difference of $120^\circ$. The deviation in time of the recorder pen from this initial position is representative of the maximum temperature deviation. The sensitivity of this technique is limited by the phase stability of the oscillator ($\sim$15 nsec). Using water ($20.0^\circ$C), the dependence of velocity on temperature is about 3.5 m/sec/$^\circ$C (Nomoto, 1958). At 9 MHz it is found that a phase change corresponding to 30 nsec is easily detected. The velocity of sound is nominally about $1.5 \times 10^5$ cm/sec or $1.5 \times 10^{-4}$ cm/nsec. A path length of 15 cm results in a transit time of $10^5$ nsec. A $1^\circ$C change in temperature results in 250 nsec change in transit time. Thus, under these conditions $\pm$ $0.1^\circ$C deviations are easily monitored.

## F.   Experimental Error

The error that arises in the determination of the absorption coefficient is usually assessed by comparing values obtained in an extensively characterized liquid for which published values exist. For purposes of this investigation, water is used as a standard liquid. The instrument has been repeatedly checked with distilled water resulting in the accumulation of a large amount of data. It is instructive to compare these results with the accepted values of Pinkerton (1947, 1949) and their implications with regard to system error.

The absorption as a function of temperature for

three frequencies is compared with the accepted behavior
(Figure 3-11). The lowest equilibrium temperature provided
by the temperature control system is $17.7^{\circ}C$. Although lower
temperatures are accessible by the addition of ice to the
thermostatic bath, the lack of temperature control and the
instability of the acoustic signal made analytical measure-
ments at low temperatures unfeasible. The highest tempera-
ture at which this system has been operated is about $60^{\circ}C$.
Although acceptable performance is encountered at this
temperature, the use of higher temperatures was not
pursued to avoid damaging the absorption chamber. In
particular, it was feared that the epoxy resin supporting
the transducer would deteriorate.

The frequency-free absorption between $17.7^{\circ}C$ and $55^{\circ}C$
at three frequencies shows some deviation from the accepted
values but the general temperature dependence is reproduced.
Each point represents the mean value of ten consecutive
determinations and all measurements are made with the (LI)
configuration of the cell.

Most measurements were conducted at $20.0^{\circ}C$. The
results of a large number of measurements of the absorption
in water at $20.0^{\circ}C$ are tabulated in Table 3-2. Because
a smaller number of determinations at each frequency are
used in the actual measurement of experimental liquids, the
standard deviations and the fractional standard deviations
are used to characterize the experimental reproducibility.
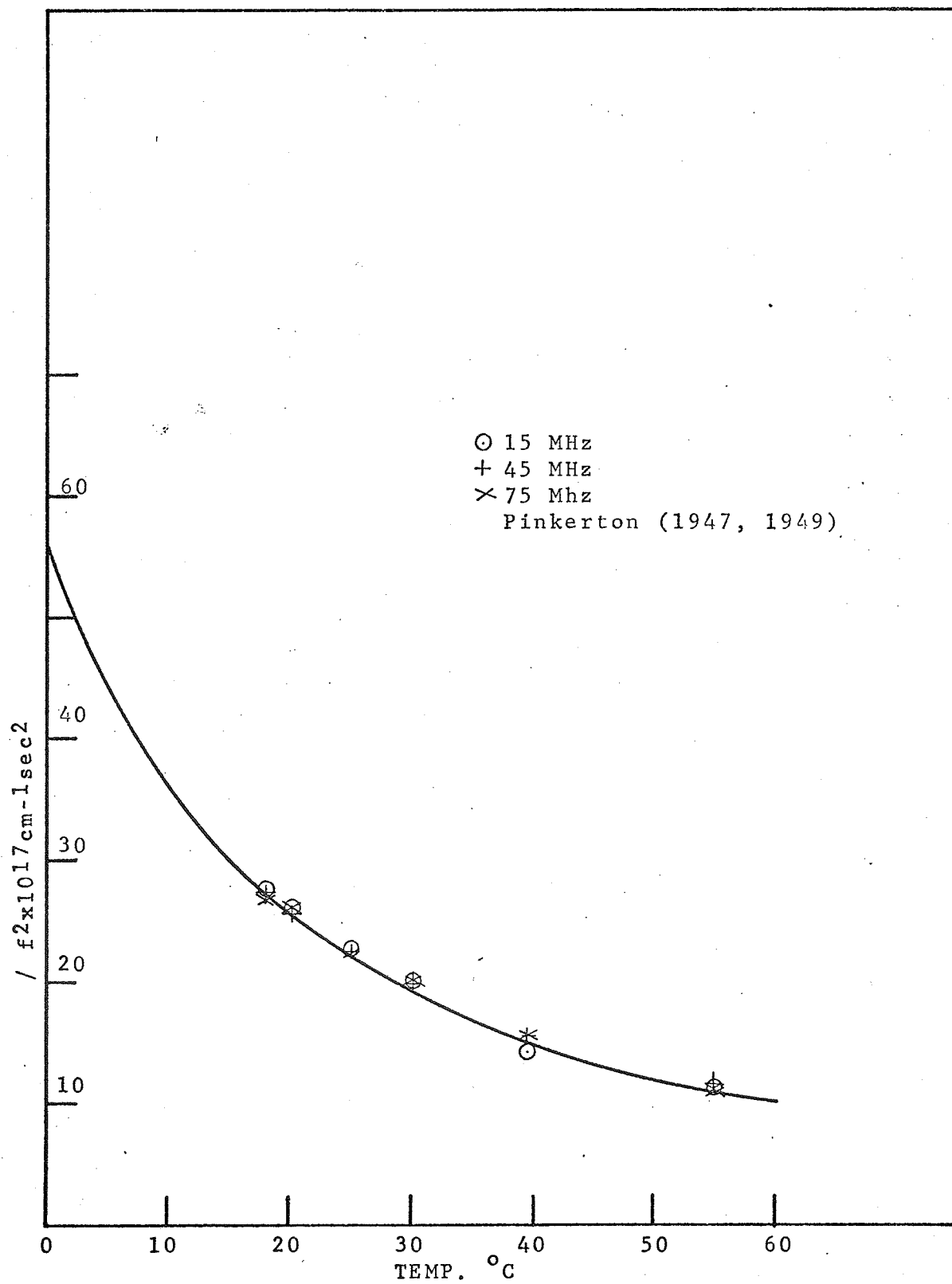It is seen that systematic deviation from the Pinkerton
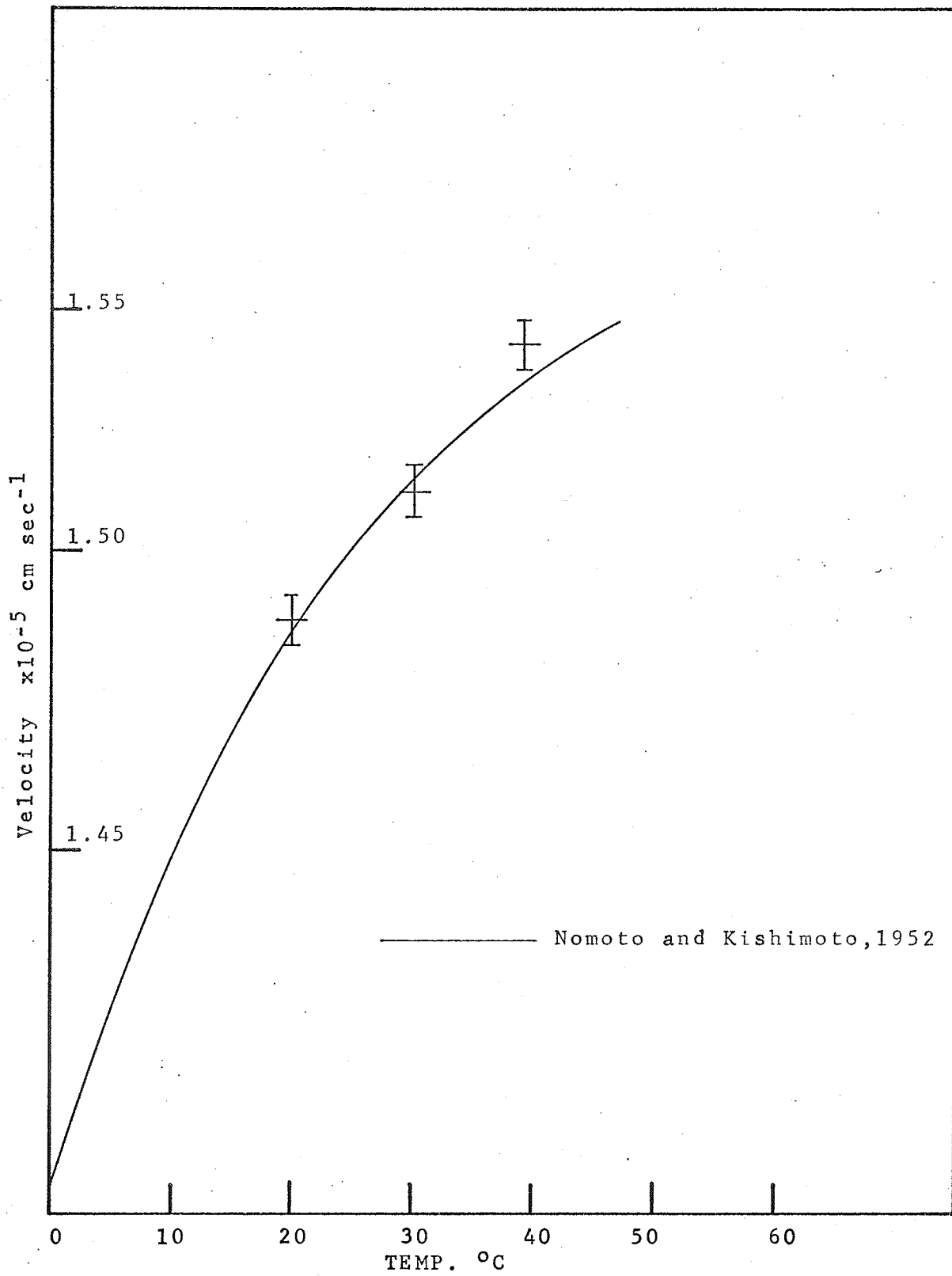
Figure 3-11  Absorption vs Temperature, $H_2O$

Figure 3-12   Speed of Sound in $H_2O$, 3 MHz

TABLE 3-2

ERROR DATA

SYSTEM PERFORMANCE:  WATER 20.0°C

| f MHz | SYSTEM | $\alpha/f^2$ $cm^{-1}sec^2$ | STD. DEVIATION $cm^{-1}sec^2$ | FRACT. STANDARD DEVIATION % | NUMBER OF MEASUREMENTS |
|---|---|---|---|---|---|
| 9 | LI | 27.09 | 1.01 | 3.72 | 68 |
| 15 | LI | 25.87 | 0.84 | 3.24 | 76 |
| 21 | LI | 25.63 | 0.63 | 2.46 | 32 |
| 27 | LI | 25.67 | 0.77 | 2.99 | 77 |
| 33 | LI | 25.25 | 0.72 | 2.85 | 11 |
| 37 | LI | 26.08 | 1.06 | 4.06 | 68 |
| 45 | LI | 25.83 | 1.18 | 4.56 | 45 |
| 51 | LI | 26.33 | 1.18 | 4.48 | 82 |
| 75 | LI | 26.11 | 1.46 | 5.59 | 37 |
| 87 | LI | 26.83 | 1.76 | 6.56 | 4 |
| 105 | H | 26.03 | 1.02 | 3.92 | 37 |

TABLE 3-2 cont'd

| f MHz | SYSTEM | $\alpha/f^2$ cm$^{-1}$sec$^2$ | STD. DEVIATION cm$^{-1}$sec$^2$ | FRACT. DEVIATION % | NUMBER OF MEASUREMENTS |
|---|---|---|---|---|---|
| 135 | H | 25.88 | 1.33 | 5.14 | 41 |
| 165 | H | 26.22 | 2.03 | 7.74 | 23 |
| 195 | H | 27.11 | 4.88 | 18.00 | 12 |

ESTIMATED ERROR IN OTHER PARAMETERS:

| | % ERROR |
|---|---|
| FREQUENCY | 0.2 |
| VELOCITY | 0.35 |
| CONCENTRATION (DEXTRAN) | 0.5 |
| CONCENTRATION (POLYSTYRENE) | 0.3 |
| DENSITY | 0.1 |

value ($25.3 \times 10^{-17} \text{cm}^{-1}\text{sec}^2$) occurs. This may be the result

of a systematic error and some difference in the water used.

Also notable is the 4% fractional deviation that exists

at 9 MHz. This is presumably due to diffraction losses

and experimental values are corrected by this amount

where appropriate. The general performance at high frequen-

cies deteriorates with both configurations. Although the

(H) configuration never achieves the reliability of the (LI)

system, it does allow the continuation of acceptable

measurements to about 165 MHz. The results at 195 MHz

represent two sets of determinations made at an early and

at a late stage in the investigation. While earlier measure-

ments indicated that perfectly acceptable results could

be obtained at this frequency with moderate care, the later

determinations include a somewhat larger error. This is

presumably due to attrition of the bearing surfaces. In

addition, it is felt that all the high frequency measure-

ments are to some degree influenced by this effect. However,

since most of the data concerning water are not dated,

further characterization is not possible.

Without attempting further to resolve the systematic

deviation from the Pinkerton value, an $\alpha$ at 20.0°C of

$26.0 \times 10^{-17} \text{cm}^{-1}\text{sec}^2$ is used in all further computations.

In the light of the above data, it is not felt that the error

introduced by this assumption will be significant.

Because water is in all cases the lowest absorbing

material studied, brief consideration of the influence

of the absorption coefficient on error is warranted.
For all the materials studied (with one exception) the
frequency-free excess absorption decreases with increasing
frequency and above 50 MHz a relatively small variation of
absorption from that of the solvent is observed. At low
frequencies, the excess frequency-free absorption may become
appreciable. In all circumstances, however, this has
the effect of decreasing the relative magnitude of the losses
attributable to diffraction. In general it is felt that the
values appearing in Table 3-2 are representative of the maxi-
mum error associated with the instrument.

A list of estimated error values that arise in the
computation of acoustic parameters is also shown in Table 3-2.

## G. Materials and Methods

WATER: Singly distilled water obtained from the
laboratory still was employed in the preparation of all
solutions and also for instrument characterization. Deuterium
oxide (99.5%) was obtained from the Columbia Organic Chemical
Co., Columbia, South Carolina.

DEXTRAN: The dextran was obtained in a fractionated and
characterized form from the Pharmacia Fine Chemical Co.,
Piscataway, New Jersey. The weight and number of average
molecular weights as well as the intrinsic viscosity of
each fraction was supplied by the manufacturer (Table 3-3).
No further characterization was attempted other than
determination of the intrinsic viscosity as a check on the
manufacturer's values, wherein excellent agreement ($\pm$ 0.5%)

TABLE 3-3

DEXTRAN:  Pharmacia Fine Chemical Company

Source:  _Leuconostoc mesenteroides_, NRRL-B512

| $\overline{M}_w$ x $10^{-3}$ | $\overline{M}_N$ x $10^{-3}$ | $[\eta]_0$ dl/g | $\overline{M}_w/\overline{M}_N$ |
|---|---|---|---|
| 11.2 | 5.70 | 0.098 | 1.97 |
| 21.8 | 14.5 | 0.147 | 1.50 |
| 39.8 | 25.6 | 0.198 | 1.55 |
| 72.0 | 40.5 | 0.256 | 1.77 |
| 167.0 | 105.0 | 0.38 | 1.59 |
| 370.0 | 185.0 | 0.50 | 2.00 |

Other Properties (Granath, 1958)

Partial specific volume $\overline{V}$ : 0.61 cc/g

Statistical segment length ($\overline{M}_w = 10^5$), 8.8 Å

$$2.43 \times 10^{-3} \overline{M}_w^{0.42} \quad dl/g$$

Hydrodynamic radius, $R_h = 0.32 \overline{M}_w^{0.50}$ Å

BOVINE SERUM ALBUMIN:  Sigma Chemical Company

Properties in 0.1 M KCl (Tanford and Buzzell, 1956)

$M_w$ : 65,000

$\overline{V}$ : 0.734 cc/g

R : 33.7 Å

TABLE 3-3 cont'd

LATEX PARTICLES: Dow Chemical Company

| Radius x $10^4$ cm | Standard Deviation x $10^4$ cm | Number of Measurements | Comments |
|---|---|---|---|
| .044 | .004 | 1164 | Monodisperse |
| .110 | − | − | Distribution |
| .178 | .003 | 655 | Monodisperse |
| .504 | .003 | 106 | Monodisperse |
| 3 − 7 | − | − | Distribution |

Partial specific volume : 0.95 cc/g (Cheng and Schachman, 1955)

was observed for all samples used in this investigation.
Tabulation of the weight average molecular weight $\overline{M}_W$ ,
number average molecular weight $\overline{M}_N$ , and intrinsic viscosity
$[\eta]_o$ , are found in Table 3-3.

The dextran is produced by Pharmacia from <u>Leuconostoc</u>
<u>Mesenteroides</u>, strain NRRL-B512. The hydrodynamic properties
of aqueous solutions of this material have been studied by
Granath (1958), who finds that the statistical segment length
is not much larger than the diameter of the glucose molecule
(8.8Å vs 5.15Å at a $\overline{M}_W$ of $10^5$) indicating that dextran
molecules are highly flexible.

Enough polysaccharide is dissolved in distilled water
to form a one liter stock solution with a concentration of
10 - 15%. Solutions are sterilized and clarified by filter-
ing through membrane filters (pore size, $0.3\mu$) and then
refrigerated at $5^{o}C$ until acoustic measurements are made.

Generally, a small amount of distilled water remains
in the absorption vessel as a residue from the preliminary
performance check. Thus, the polysaccharide concentration
is most effectively determined after the solution has been
introduced into the acoustic chamber and vigorously pumped
through the vessel for several minutes.

Concentration of aqueous dextran solutions is conven-
iently determined polarimetrically with an error not exceeding
1%. The specific rotation $[\alpha]_D^{20}$ was determined to be $199^{o}$.
This compares to a value of $+198^{o}$ by Senti <u>et al</u> (1955) and
$199^{o}$ listed by Pharmacia. A Rudolf model 70 polarimeter
capable of resolving $\pm.01^{o}$ was employed for optical rotation
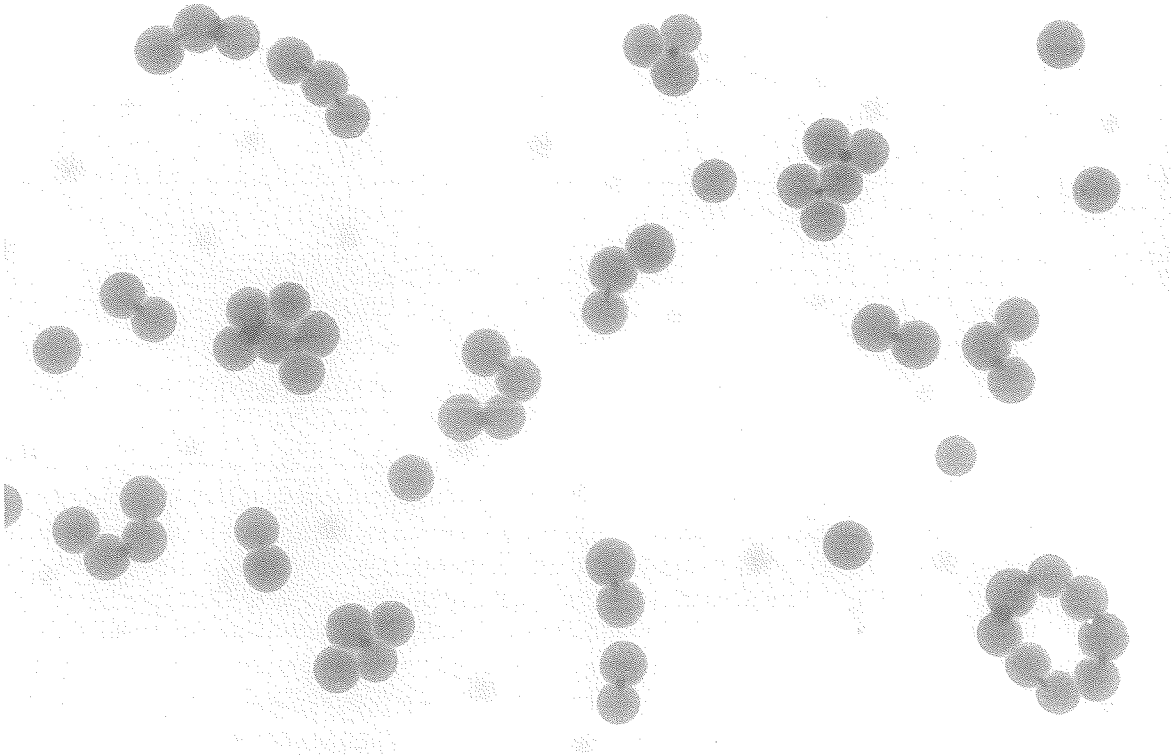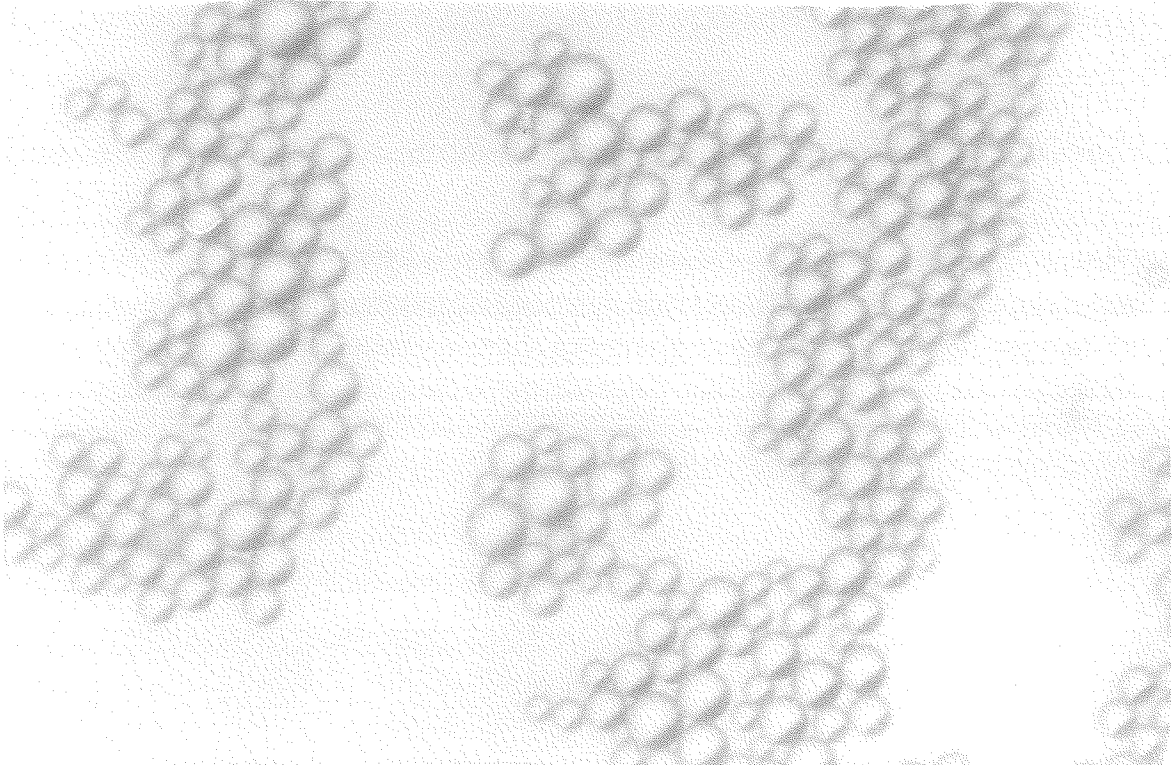
FIGURE 3-13   ELECTRON MICROGRAPH, POLYSTYRENE PARTICLES
              RADIUS:   0.044 micron


FIGURE 3-14   OPTICAL MICROGRAPH, COPOLYMER PARTICLES
              AVERAGE RADIUS:   3.5 micron

measurements. Room temperature was sufficiently stable ($\pm 1^{\circ}C$) that no special temperature control was necessary.

Intrinsic viscosities are determined using Cannon-Ubbelohde semi-micro-dilution viscometers suspended in a temperature control bath ($\pm 0.01^{\circ}C$). The viscometers were selected such that the flow time is all cases was sufficiently long (> 100 sec) to avoid kinetic energy errors. Values obtained agreed with the Pharmacia values in all cases.

BOVINE SERUM ALBUMIN: Fraction V, bovine albumin was obtained from Sigma Chemical Co., St. Louis, Missouri. A one liter solution with a nominal concentration of 10% was prepared by suspending the protein in 0.1M KCl, pH 6.8. This solution was membrane filtered prior to acoustic measurements.

Concentration is determined on the basis of an extinction coefficient of 6.6/g albumin/100mg/cm (Cohn et al., 1947). Observation of the behavior of this material in the ultracentrifuge revealed a single sharply defined peak with a sedimentation coefficient, $S_{20,w}$ equal to $4.07 \times 10^{-13}$ sec at a concentration of 5.4 mg/ml. This compares favorably to the accepted value and is a reasonable indication that the material under study is native protein.

POLYSTYRENE LATTICES: Spherical polystyrene and styrene-divinylbenzene copolymer particles suspended in deionized water were supplied by the Dow Chemical Co., Midland, Michigan. Six different dispersions are investigated.

Four suspensions are monodisperse polystyrene lattices,

the type used extensively as a secondary standard in optical

and electron microscopy. The sizes of the particles have

been characterized accurately by the manufacturer (see Table

3-3). Although these materials are usually only available in

10 ml lots, one liter of each latex was provided through the

generous interest of the manufacturer. Particle concentra-

tion as supplied in each case is nominally 10% solids by

weight.

The other suspensions are heterodisperse. One of

these is a styrene-divinylbenzene copolymer latex (DC642-19)

with a nominal average particle radius of $3.6\mu$. The

distribution of particle sizes has been characterized by

the manufacturer using optical microscopy. The distribu-

tion curve is roughly Gaussian with about 99% of the

particles having radii between $1.5\mu$ and $6.5\mu$. Copolymeri-

zation of the styrene with divinylbenzene has the effect of

increasing the particle rigidity (Dow Chemical, 1966).

This suspension is also supplied with a concentration of

about 10%.

The other dispersion investigated is Dow 586 poly-

styrene latex. This material is a heterodisperse industrial

preparation supplied with a concentration of 50% solids by

weight. Nominal particle radius is 0.11 micron.

Particle concentration is determined by weighing the

residue obtained by evaporating a known volume of suspension

to constant weight. Since the polystyrene is very hydro-

phobic no special drying procedures are necessary.

The concentration of free surface active agent is

determined by centrifuging the particles from a known volume
of suspension, membrane filtering the supernatant and
weighing the residue that remain after drying. This determin-
ation is limited to the suspensions containing the $0.5044\mu$
and $3.6\mu$ particles. Removing the smaller particles from
sufficiently large quantities of suspension (50 ml) is hinder-
ed by the small density difference between the particle and
water ( $\rho_P/\rho_o \approx 1.05$ ).

The apparent partial specific volume, $\bar{V}$, of polystyrene
latex particles has been determined to be 0.950 cc/g by Cheng
and Schachman (1955). In addition, the particles appear to
be impermeable to water and thus are not internally hydrated.
The density is then given by $1/\bar{V} = 1.052$ which agrees with
the value for bulk polystyrene.

Because of the importance of the density, independent
determinations were made. Two methods are employed. Assuming
impermeability of the particle, the following expression
may be derived readily to describe the density of the two
component system:

$$\rho_P = \frac{c\,\rho_T\,\rho_o}{\rho_o + (c-1)\rho_T}$$

where c is concentration and $\rho_T$, $\rho_P$, $\rho_o$ are the densities of
the suspension, particle and suspending liquid, respectively.
Using standard pycnometric techniques and allowing an
accuracy in measurement of 0.0001 g/cc, the density of all
the polystyrene latex particles is found to be in agreement
with the previously determined value. The copolymer particles

are also found to have an average density of about 1.052 g/cc. It is found, however, in centrifuging these particles in sucrose solutions that neither complete sedimentation or floatation occurs when the suspending fluid has a density between 1.050 and 1.053 g/cc. In addition, observation of the behavior in a sucrose gradient reveals that while most of the particles accumulate as in a narrow band, isolated particles exist with densities as high as about 1.058 g/cc. This probably is due to some variation of the copolymerization from particle to particle. No further effort was made, however, to characterize the density of this material and the average value will be employed in subsequent discussions.

Contact of the latex with glass and stainless steel results in the deposition of a thin layer of polystyrene. This is avoided to a large extent by treating the contact surfaces with Desicote.

For the most part, the acoustic procedures involved in the use of the particles are the same as with homogeneous materials, with one notable exception. It was found that the piston transducer could not be driven through suspensions of the largest particle size without mechanical seizure due to "wedging" of the particles between the bearing surfaces. This problem can be alleviated by inserting spacers between the transducer housing and piston head so that the transducer may move through the suspension while the piston bears on the sur- face near the top of the absorption cylinder. This modifica- tion, however, limits the maximum transducer separation to the extent that the absorption coefficient and 3 MHz could not

be obtained.

---

1. Manufactured by Duff Norton Corp., Charlotte, North
   Carolina.

2. Manufactured by Thomson Industries, Manhasset, New
   York.

3. Obtained from the Minarik Electric Co., Los Angeles,
   California.

CHAPTER IV

## RESULTS

The dependence of the absorption coefficient on con-
centration in aqueous solutions of dextran at $20.0^{\circ}C$ for
six molecular weight fractions has been measured. Figure
4-1 depicts the behavior at $M_w$  72,000, which is represen-
tative of that observed at all the molecular weights
investigated ( 11,200; 21,800; 39,800; 167,000; 370,000).
It may be observed that at frequencies greater than 3 MHz a
common zero-concentration intercept on the ordinate axis
exists which reflects the Stokes $f^2$-dependence of the
absorption of the solvent. Thus, by employing a normalized
ordinate parameter of $\alpha/f^2$, the data at a number of
frequencies are conveniently compared in a single plot.
At 3 MHz, the concentration-independent term is removed
along with the diffraction losses (Chapter III) so that
for this frequency the ordinate is $\Delta\alpha/f^2$. The value of
$\alpha$ at each frequency at every concentration represents the
mean value of at least five individual determinations.

Effective comparison of the absorption properties of
different macromolecular species is facilitated by defining
a reduced coefficient that results upon removing the
contribution of the solvent and the concentration dependence,
viz.,

$$A = \frac{1}{c}\frac{\Delta\alpha}{f^2} = \frac{1}{c}\left[\frac{\alpha_T - \alpha_o}{f^2}\right]$$

where $\alpha_T$ is the observed absorption coefficient and $\alpha_o$ is
the concentration independent term. Figure 4-2 is a
composite log-log plot of A as a function of frequency for
all molecular weights investigated. The results of measure-
ments of the absorption in aqueous solutions of glucose are
also represented. Shear viscosity contributions to the
absorption as calculated by the Rouse theory for the
highest and lowest molecular weights are depicted within
the range of applicability of the high frequency approxima-
tion (cf Appendix I). Values of A for the various fractions
are shown in Table 4-1. Similar measurements of absorption
in aqueous solutions of bovine serum albumin in 0.1 M KCl
at $20.0^{\circ}C$ are shown in Figures 4-3 and 4-4. The absorption
characteristics of aqueous suspensions of beef hemoglobin
(Carstensen, 1959b, Edmonds, 1966b) are compared with the
albumin data in Figure 4-4. Tabulation of the values of
A of serum albumin are also shown in Table 4-1.

The absorption in the five aqueous polystyrene lattices
has been measured at $20.0^{\circ}C$ in the following concentration
ranges.

| R (micron) | Conc. Range (g/cc) |
|---|---|
| 0.044 | 0.0 - 0.1 |
| ∼ 0.11 | 0.0 - 0.5 |
| 0.178 | 0.0 - 0.1 |
| 0.504 | 0.0 - 0.1 |
| ∼ 3.5 | decreasing 0.0 - 0.1 at 9 MHz |
| | to 0.05- 0.01 at 81 MHz. |

In every case, the absorption has appeared as a linear function of concentration to at least 0.1 g/cc. Representative of the dependence of absorption on concentration is that observed in DC-586 (R: ∼0.11 microns) in which the concentration range is the greatest (Figure 4-5). The corresponding value of A at each frequency as above is taken from the slope in the concentration range where the dependence is linear. A minimum of three concentrations were used in each case to determine A. A log-log plot of A vs frequency for the lattices is shown in Figures 4-6, 4-7, 4-8, 4-9, 4-10.

The temperature variation of the absorption parameter A, in DC-586 from about 17°C to 48°C at four frequencies from 9 MHz to 51 MHz is shown in Figure 4-11.

The addition of NaCl in an amount sufficient to make a 10% suspension of DC-586 0.05 M in this salt did not result in a detectable change in the absorption properties. Higher concentrations of salt results in flocculation of the latex particles and further variation of the ionic environment of the particle was not attempted. Log-log plots of A vs frequency for the particles investigated are shown in Figures 4-5 to 4-10. A minimum of three concentrations is each case were used to determine the values of A. The theoretical absorption character predicted on the basis of the Lamb, Epstein, Urick relations is shown for reference for each particle size.

In order to further examine experimentally the

importance of the relative motion contribution, it is

of interest to investigate the absorption character in the

absence of relative motion which is the case when the

particle and surrounding medium possess the same density.

Since the specific gravity of the particle is fairly close

to unity (1.055 at 20.0$^{\circ}$C), this situation may be conven-

iently achieved by employing a $D_2O$- $H_2O$ mixture as the

suspending fluid. The results of absorption measurements

at 20.0$^{\circ}$C in suspensions of 0.504 $\mu$ particles in a $D_2O$-$H_2O$

mexture with a specific gravity of 1.053 are shown in

Figure 4-9 along with those obtained with $H_2O$ as the

suspending liquid.

Although velocity determinations were not a primary

consideration in this investigation, a number of measure-

ments were made using the method described in Chapter III.

A composite plot of the relative velocity $V/V_o$, $V_o$ being

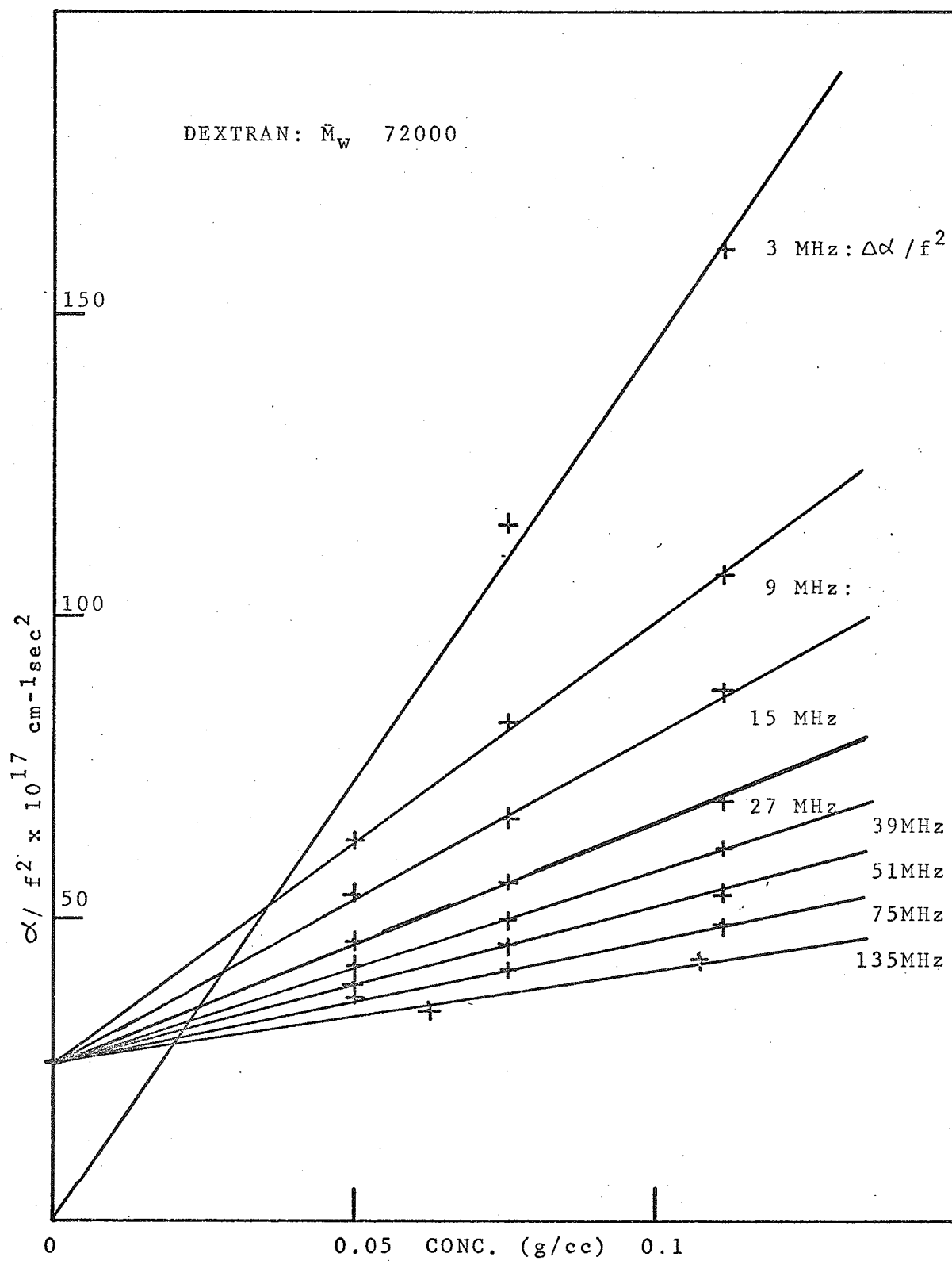the velocity of water, for the materials investigated is

found in Figure 4-12.

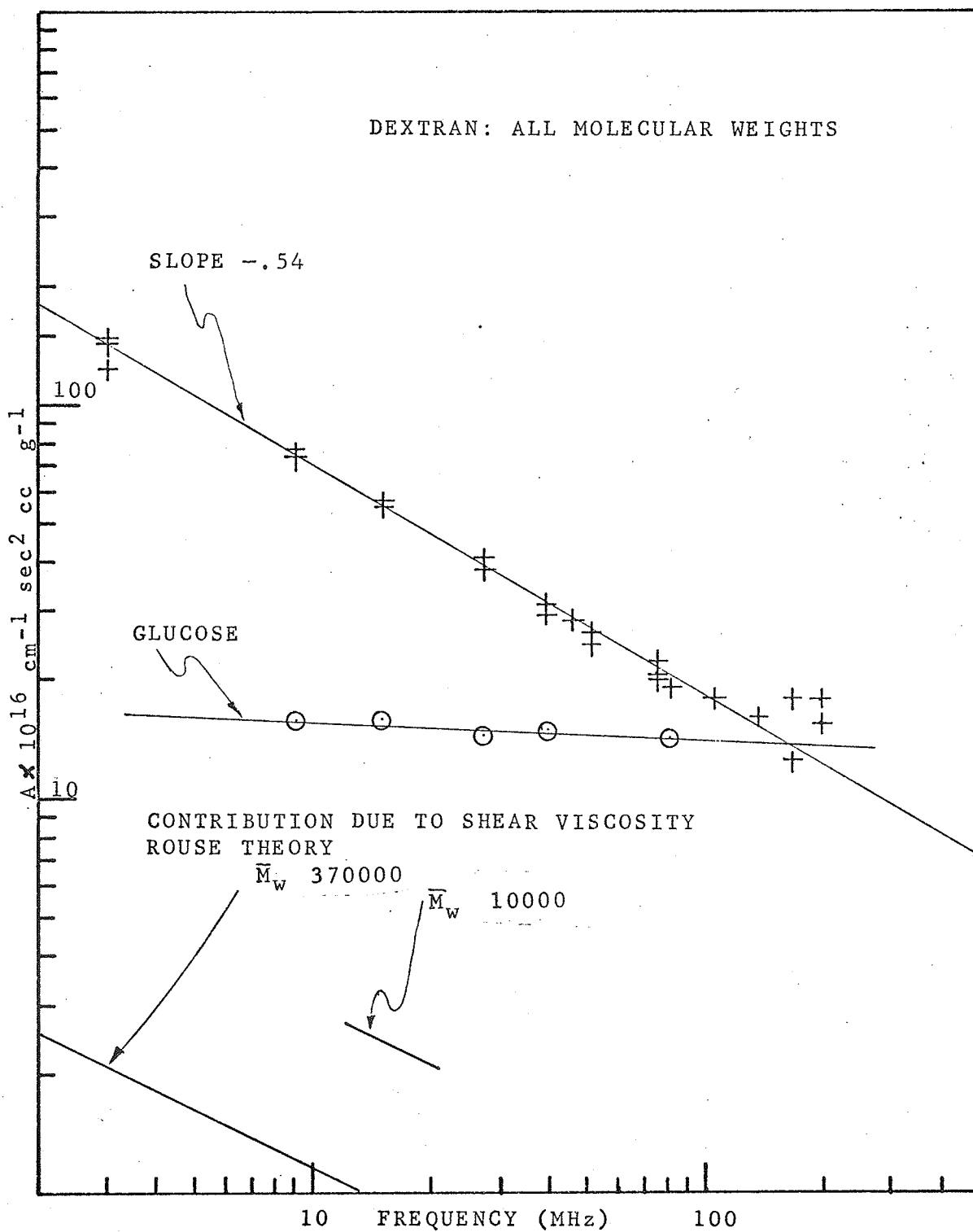Figure 4-1  Absorption vs Concentration, Dextran  $\overline{M}_W$  72000
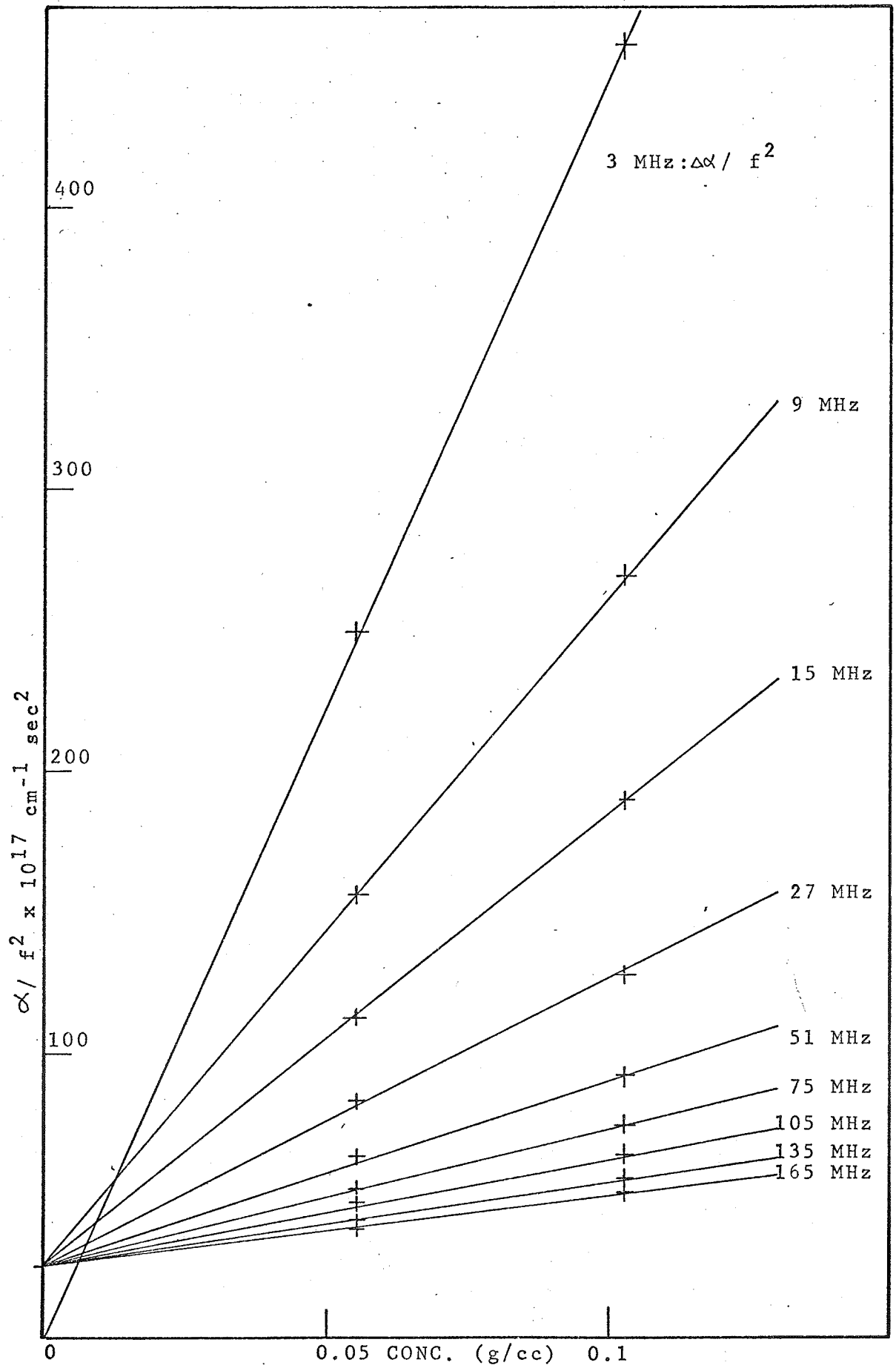
Figure 4-2: EXCESS ABSORPTION IN DEXTRAN

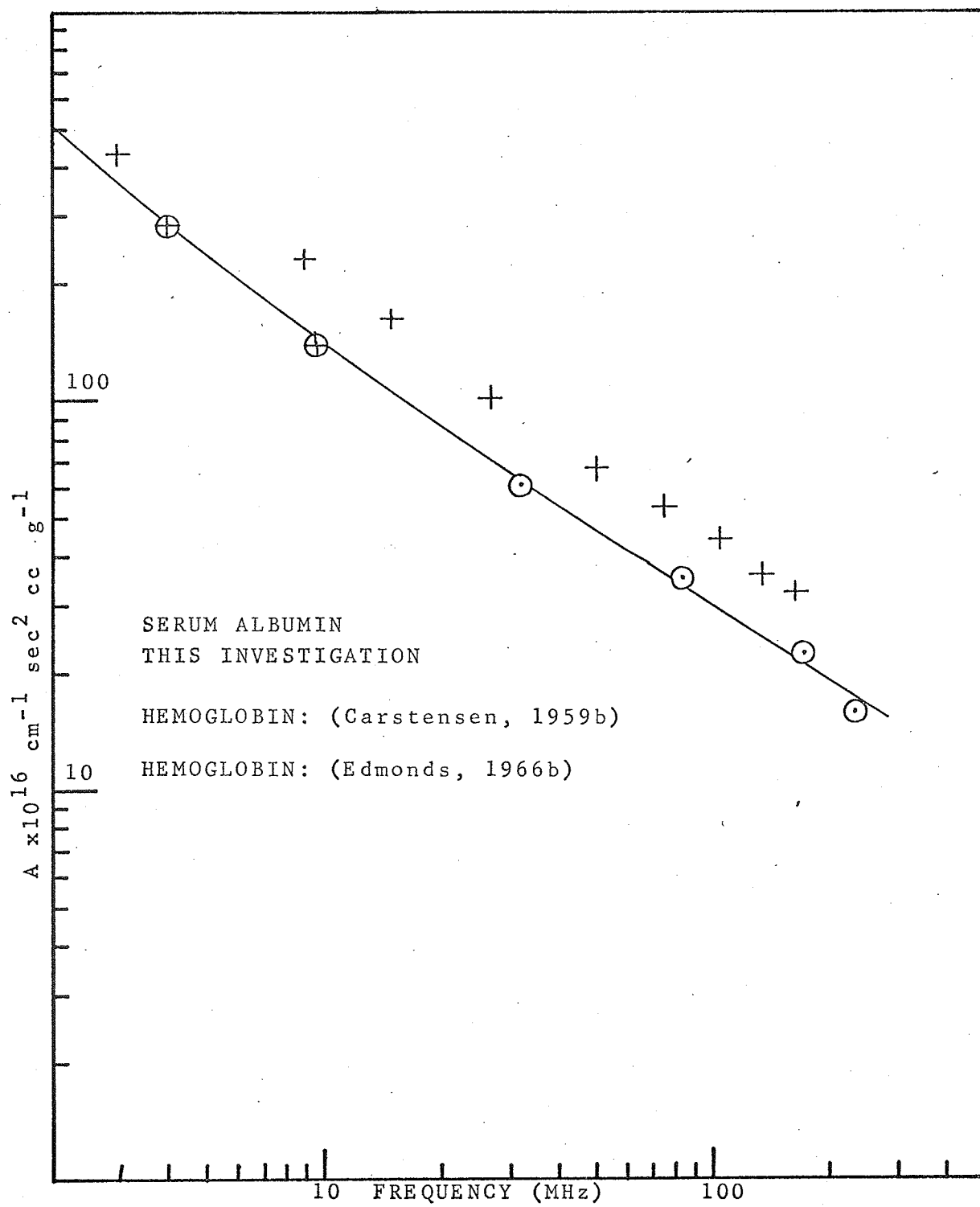Figure 4-3   Absorption vs Concentration, Bovine Serum Albumin

Figure 4-4: Bovine Serum Albumin

TABLE 4-1

Values of A for DEXTRAN and SERUM ALBUMIN

$A \times 10^{16} \ cm^{-1} \ sec^2 \ cc \ g^{-1}$

$$T = 20.0^\circ C$$

| $M_w \times 10^{-3}$ | 11.2 | 21.8 | 39.8 | 72.0 | 167 | 370 | BSA |
|---|---|---|---|---|---|---|---|
| f(MHz) | | | | | | | |
| 3 | 122. | 143. | <u>147.</u> | <u>147.</u> | - | - | 430 |
| 9 | 74. | <u>75.</u> | <u>77.</u> | <u>73.</u> | <u>71.</u> | <u>68.</u> | 235 |
| 15 | <u>55.</u> | <u>57.</u> | <u>56.</u> | <u>56.</u> | - | <u>51.</u> | 163 |
| 27 | 41. | 38. | <u>38.</u> | 40. | <u>37.</u> | <u>39.</u> | 1?2 |
| 39 | 29. | - | <u>30.</u> | - | - | - | - |
| 45 | - | 28. | - | - | - | <u>25.</u> | — |
| 51 | 26. | 24. | 26. | <u>25.</u> | - | - | 68 |
| 75 | 20. | 21. | 22. | 20. | <u>18.</u> | <u>17.</u> | 53 |
| 81 | - | - | 19. | - | - | - | - |
| 105 | - | - | - | 18. | - | - | 44 |
| 135 | - | 17. | - | 16. | - | <u>16.</u> | 36 |
| 165 | - | 18. | - | 15.3 | - | <u>12.6</u> | 32 |
| 195 | - | 18 | - | - | - | <u>15.3</u> | - |

Underlined values occur at frequencies such that

$2 < \omega \tau_1 < \dfrac{N^2}{250}$ ; see Appendix I.
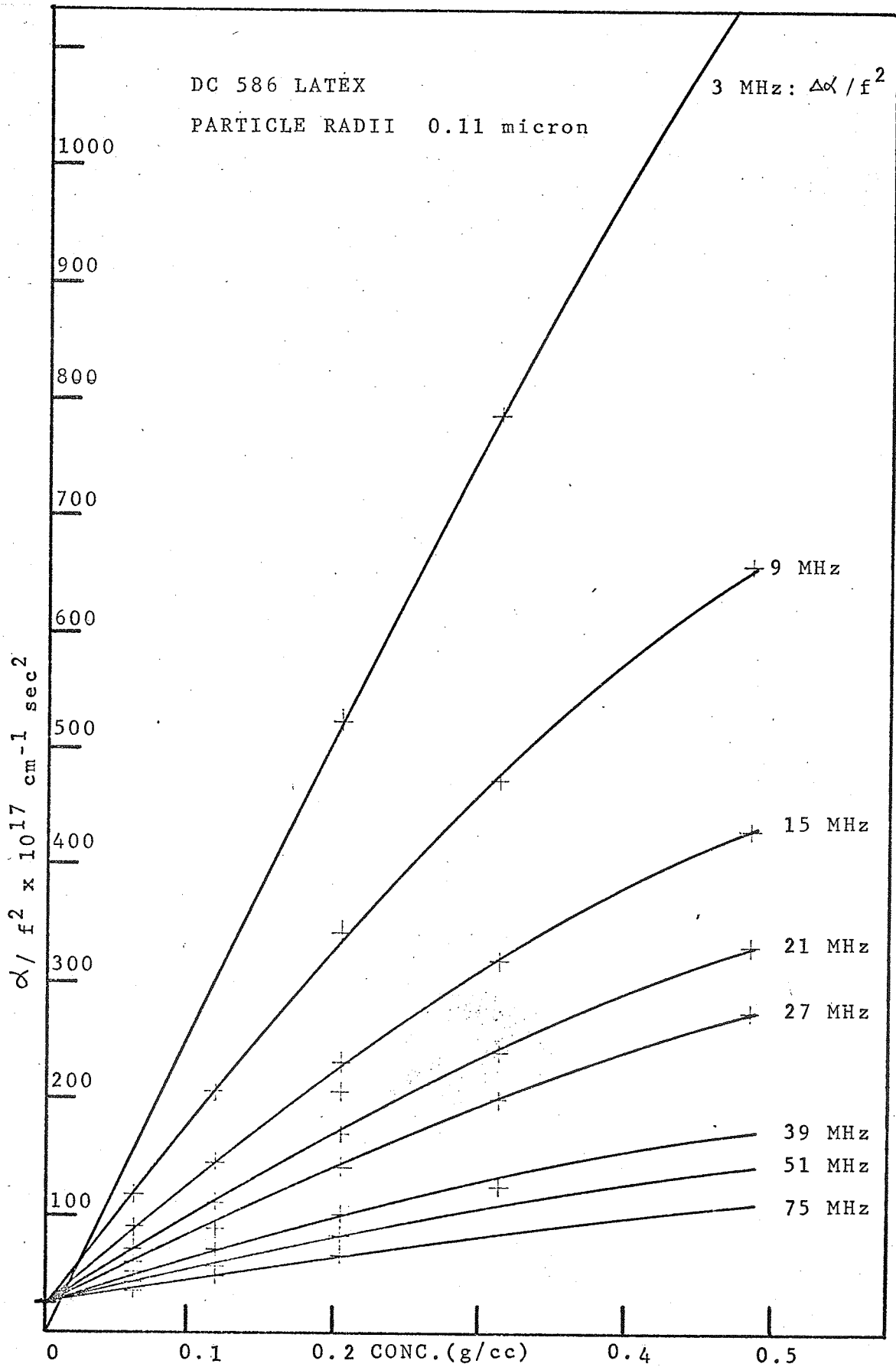
Figure 4-5   Absorption vs Concentration, DC 586 Latex

PARTICLE RADIUS: 0.044 micron

$A \times 10^{16}$ $cm^{-1}$ $sec^2$ $cc$ $g^{-1}$

100

10

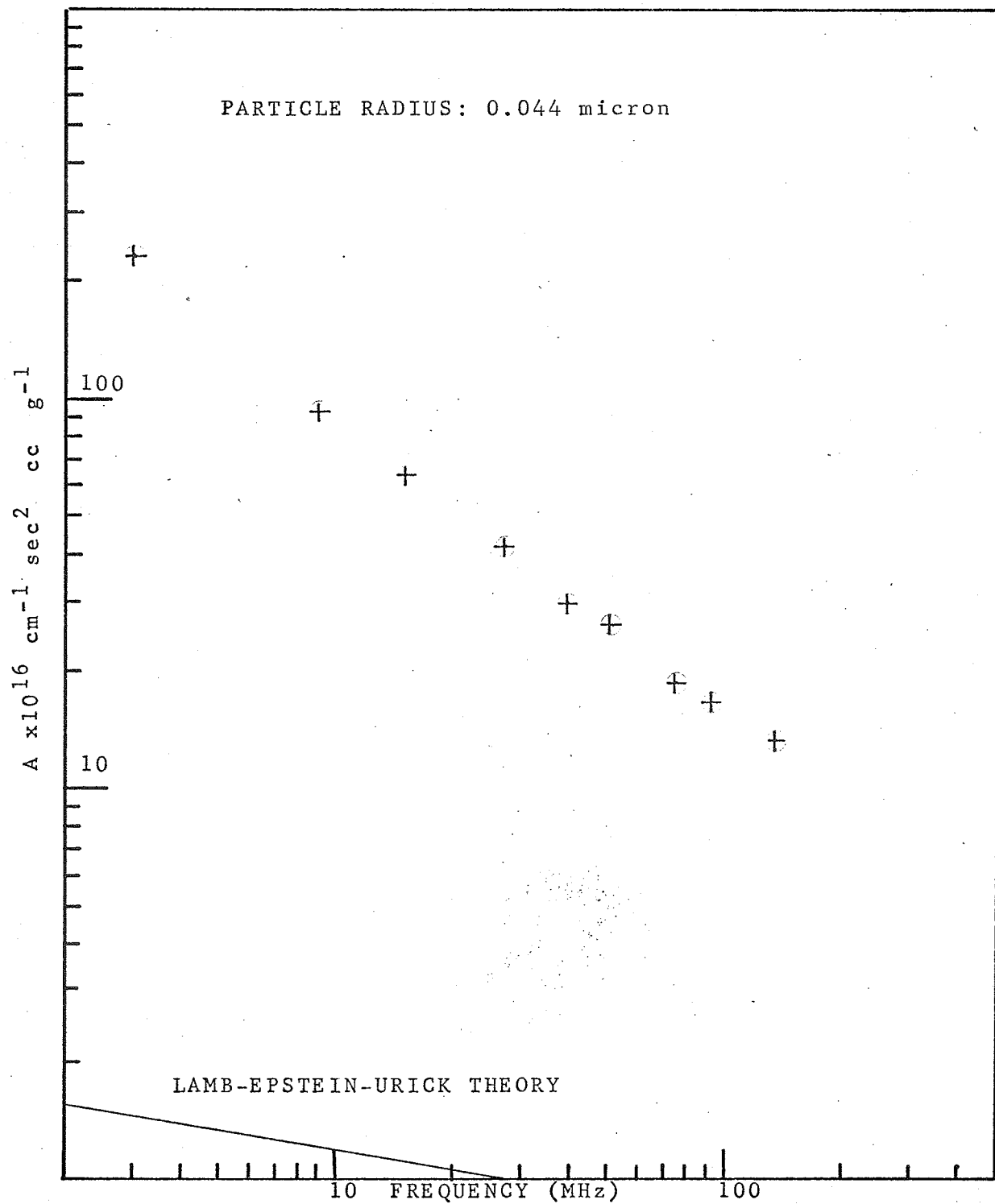LAMB-EPSTEIN-URICK THEORY

10    FREQUENCY (MHz)    100
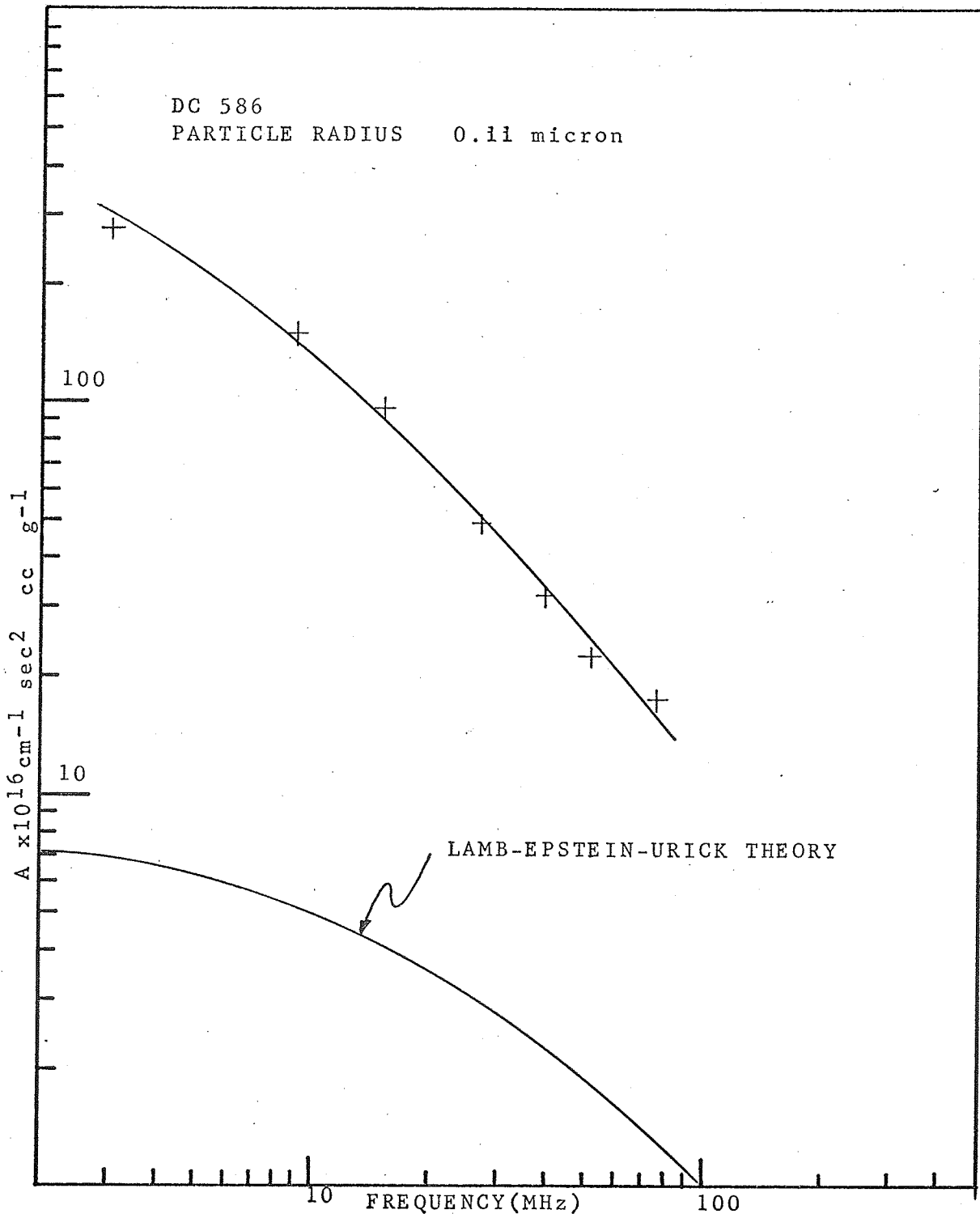
Figure 4-6: Particle Radius; 0.044 micron

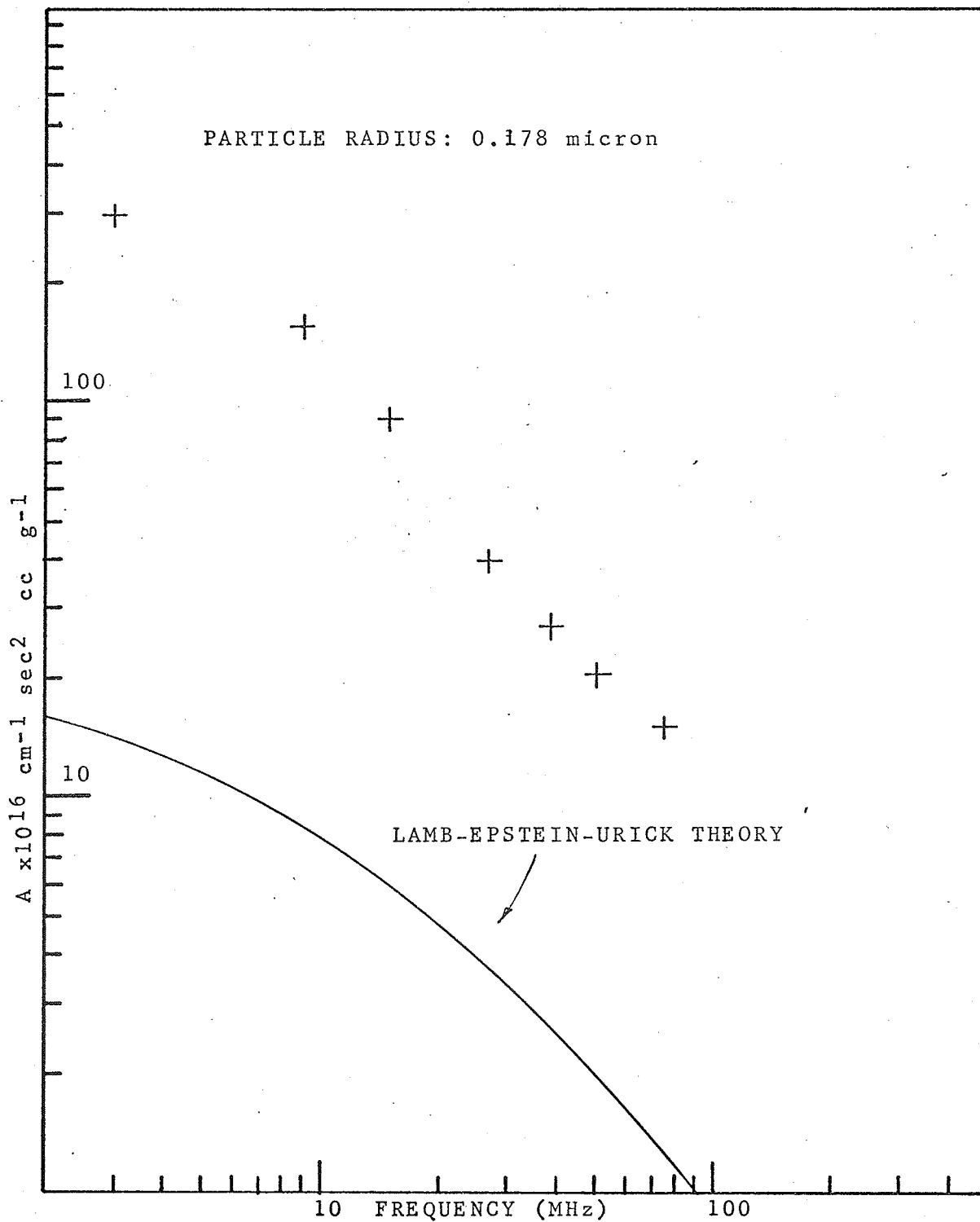Figure 4-7: DC 586    Particle Radius;    0.11 micron

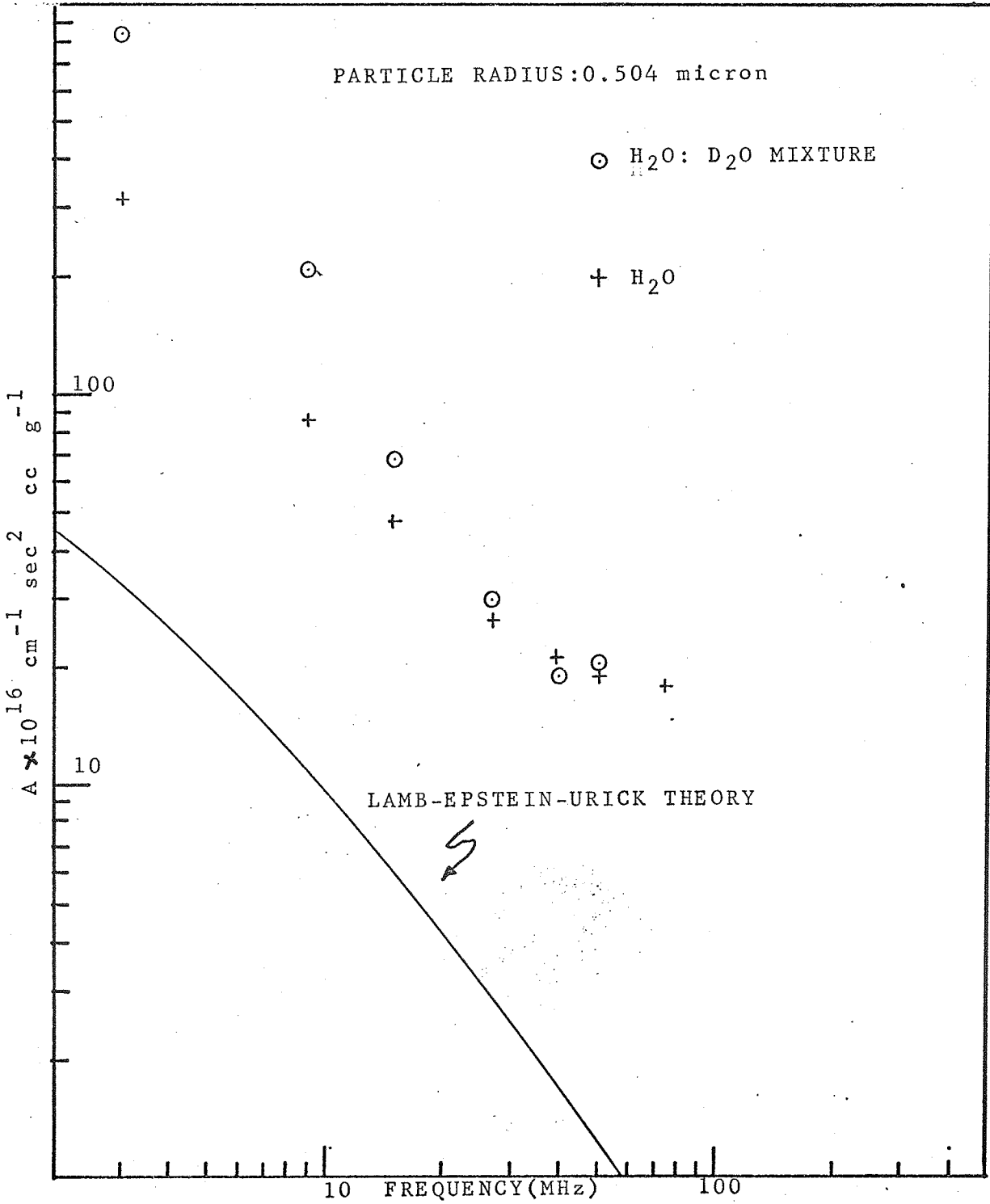Figure 4-8: Particle Radius; 0.178 micron
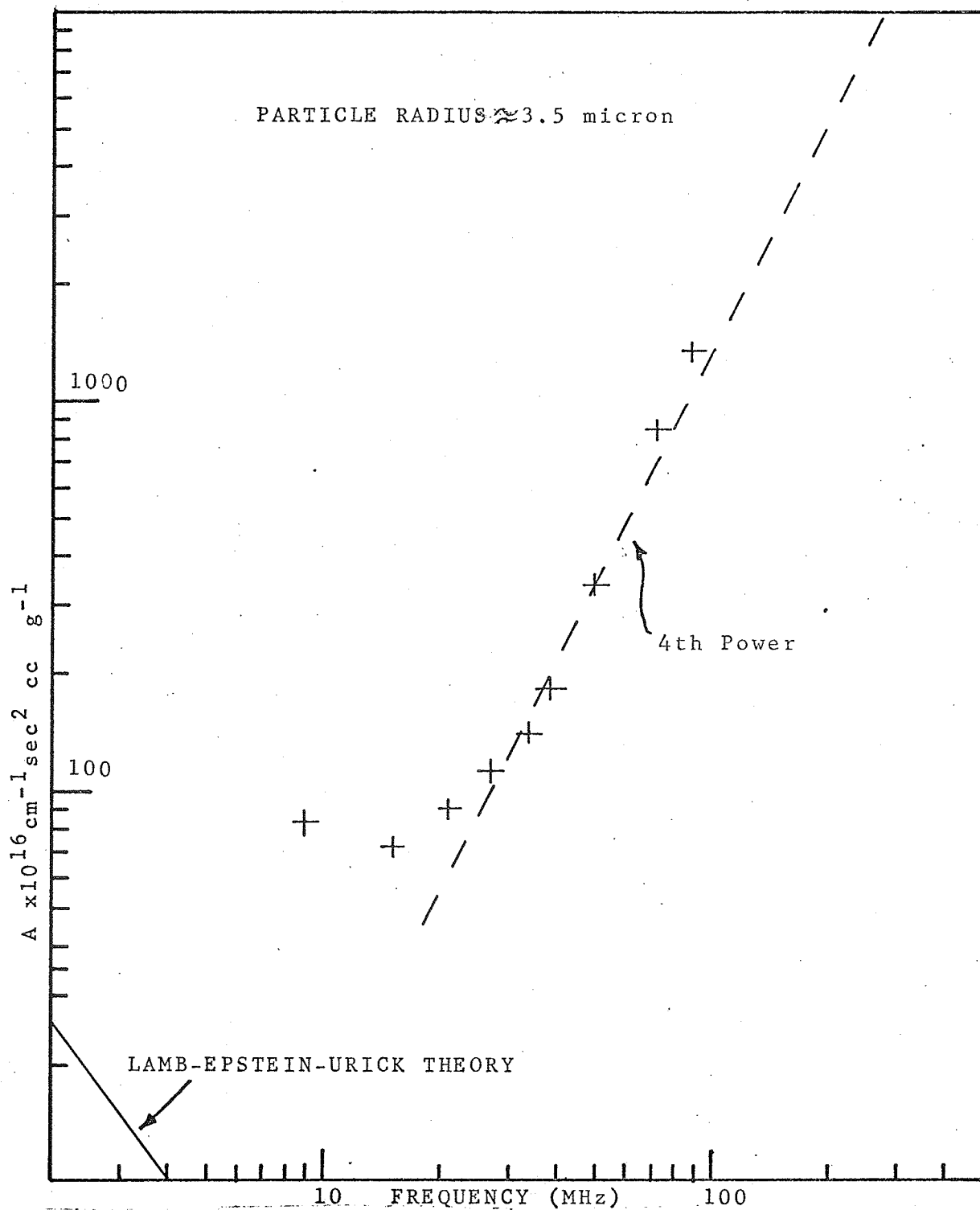
Figure 4-9: Particle Radius; 0.504 micron
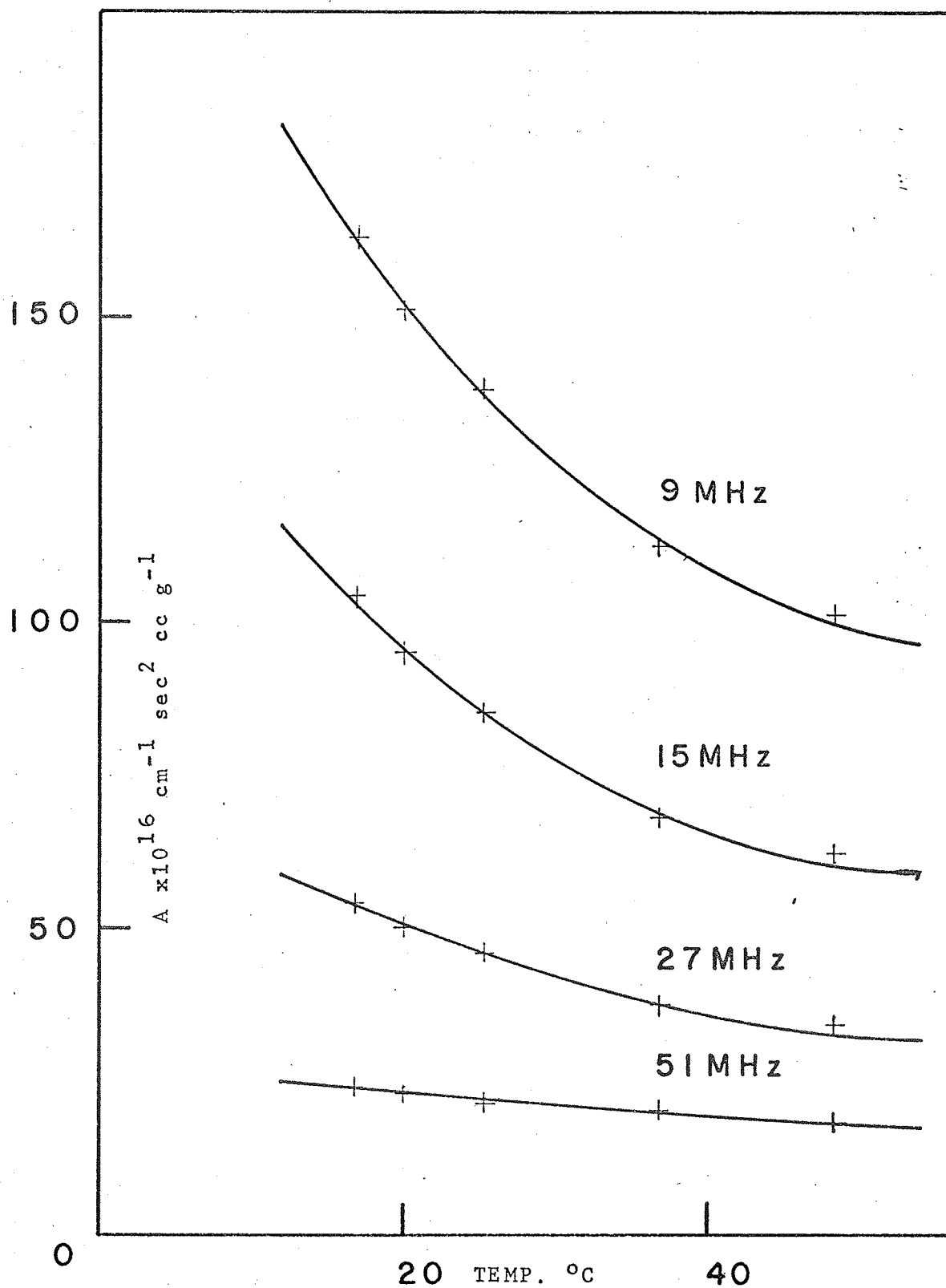
Figure 4-10: Particle Radius; 3.5 micron
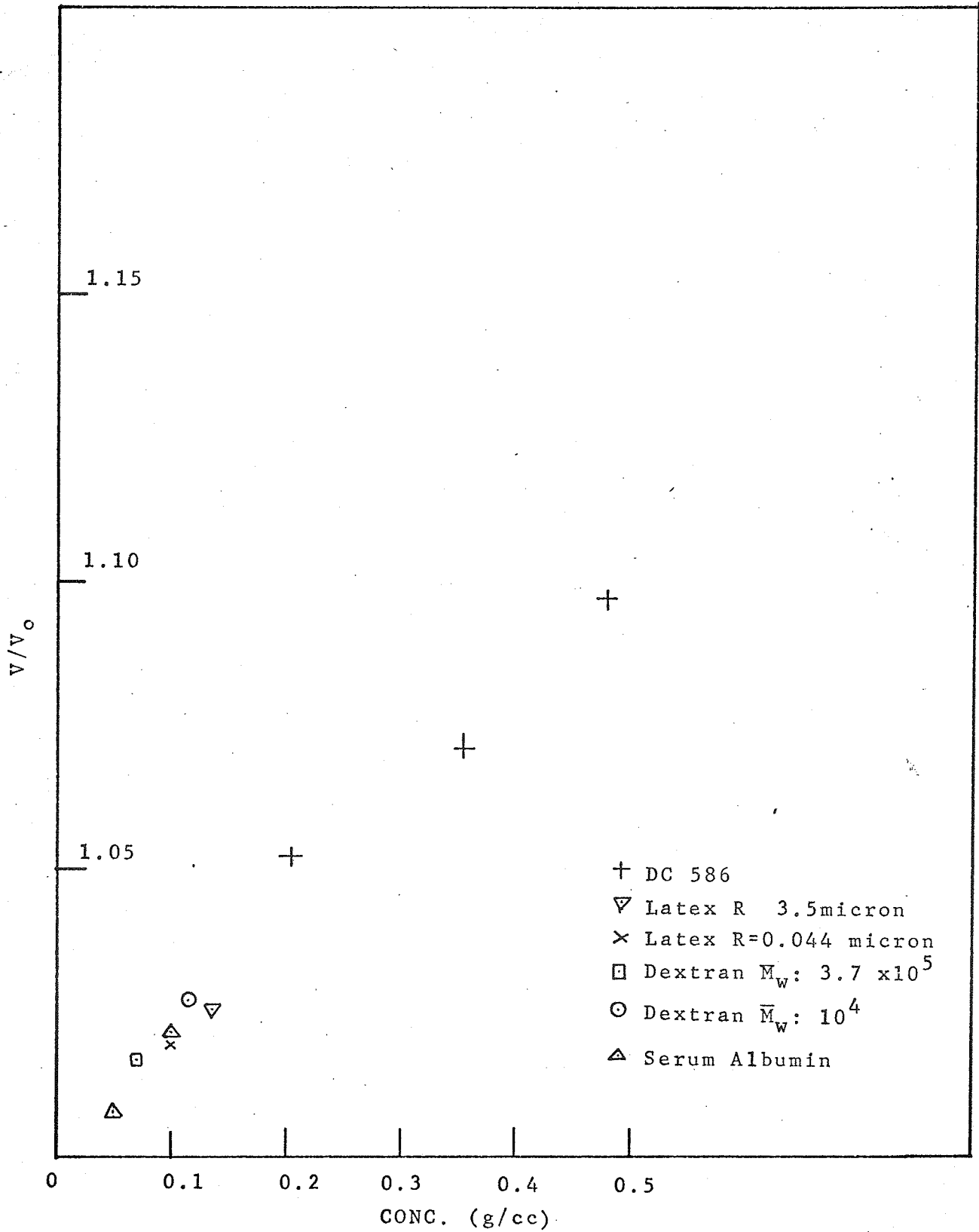
Figure 4-11  Excess Absorption vs Temp., DC 586 Latex

Figure 4-12    Relative Velocity vs Concentration

CHAPTER V

DISCUSSION

The results presented in the previous section indicate
that the ultrasonic absorption properties of both the dextran
fractions and the aqueous polystyrene lattices (excluding
that containing the largest particle sizes which will be
discussed separately) have three essential similarities in
common with the properties observed in other macromolecules
in this frequency region:  1) The absorption coefficient
is related linearly to the concentration of suspended
material and extrapolates to the value of the solvent at
zero concentration.  2) The frequency-free excess absorption,
A, decreases with increasing frequency, approaching zero or
a small limiting value.  3) It is observed that the speed of
sound at 3 MHz in all cases is increased in approximately
the same way by the presence of suspended matter, indicating
that their incorporation into the fluid is accompanied by
a decrease in the macroscopic density and compressibility.

The linearity of absorption with concentration implies
that the absorption coefficient associated with individual
particles or molecules is constant and independent of
effects related to proximity or interactions between neigh-
boring molecules or particles.  In this light, the ensuing
discussion will attempt to evaluate the dissipational
processes that may occur either internally, externally, or
at the surface in terms of the mechanisms described in
Chapter II.

A.  Dextran

Computation of the shear viscosity from the Rouse theory
(Appendix I) is based on the assumption that the affinity
between solvent molecule and polymer segment is similar to
that between adjacent polymer segments.  While this
assumption may not be a precise molecular description of the
dextran solutions, it does guarantee consideration of vis-
cosity under conditions of maximum extension of the macro-
molecule into the solvent (Tanford, 1958, p. 163) and minimum
damping of internal modes resulting from molecular constric-
tion.  Thus, the computed value represents the maximum
dynamic shear viscosity in the system.

It can be seen in Figure 4-2 that the absorption co-
efficient computed from the Rouse theory leads to values
which are at least an order of magnitude too small to
account for the observed behavior.  This is a reasonable
indication that the attenuation of shear stresses by the
solute molecule are not important in determining the excess
absorption.  On the molecular level this has the following
implications:  1) At the highest molecular weight (370,000),
the longest internal modes excited by the acoustic energy
correspond to segments which have a length less than 2%
of the total polymer so that the statistical sphere
represented by the entire molecule will very nearly appear
as a rigid body to the oscillatory shear stresses.  2) If
the polymer still behaves as a random coil at a molecular
weight of 10,000, some excitation of the primary mode,

i.e., distortion of the center of gravity, is predicted. At this chain length, however, the viscosity imparted to the liquid at virtually zero frequency is too small to be an important factor in determining the large excess absorption involved.

In order to discuss the importance of volume viscosity, let it be assumed that the acoustic losses related to this process are the result of the formation and extinction of vacancies in the liquid lattice imposed by the oscillatory mechanical stresses (Hirai and Eyring, 1958). The absorption will be a manifestation of the time required for structural rearrangement of the six or eight molecules surrounding a vacancy (cf Chapter II).

Because of the random coil nature of the polymer, the glucose residues will not be in close proximity to each other and considered by themselves cannot account for the viscosity on the basis of the vacancy theory. It is necessary then to postulate that the presence of the polymer gives rise to a structural viscosity in conjunction with neighboring water molecules. While a water-polymer complex undoubtedly results from the preponderance of hydrogen-bonding sites concommitant with the glucose monomer, it is not possible on the basis of the experimental evidence alone, to state unequivocally that such a complex has a significantly higher volume viscosity than that found in solutions of glucose. If it is assumed, however, that the excess absorption is related to a volume viscosity of bound water; it is possible to derive a model which will

permit at least an empirical explanation of the observed behavior. Let it be assumed that the polymer-solvent complex exists in the form of a cylindrical layer surrounding the polymer backbone. The thickness and degree of association of the hydration layer would be in part due to the fundamental interaction of water with glucose. In addition, some cooperative effects may be attributed to periodicity and immobilization of the monomer.

It is found that the excess frequency-free absorption is nearly proportional to the reciprocal square root of frequency, i.e., the dependence predicted by theRouse theory. Thus, if the relaxation times associated with volume viscosity are the same as related in a linear way to those important to shear deformation, the same frequency dependence will be observed at high frequencies. In associated liquids, such as water, this is often the case since both shear and volume viscosity are related to the average time between movement of molecules into vacancies (Herzfeld and Litovitz, 1958). The problem that arises is the disparity between the magnitudes of the different viscosity contributions. In water and other associated liquids $\eta_v' / \eta_s' \approx 3$ , whereas the volume viscosity required to explain the absorption in dextran would require $\eta_v' / \eta_s' \approx 20$ . To apply this criterion to the postulated model would presuppose that the concentrations of the polysaccharide-water complex involved in excess volume losses and excess shear losses are identical. Whereas this is a reasonable assumption in homogeneous liquids, it is possible that the effect of the polymer may penetrate the

solvent to a greater extent in relation to volume viscosity.
Thus, if the polymer chain is assumed to have a "cylinder
of influence" associated with it, the effect on volume
viscosity would have to extend about twice as far into the
medium as that associated with shear viscosity, in order
that $\eta'_v / \eta'_s \approx 3$.

On the basis of this model it is also possible to
explain two other observations:  1) the independence of
excess absorption on molecular weight and consequently
hydrodynamic volume and 2) the apparent asymptotic approach
of the excess absorption to that of the monomer at high
frequencies.  The first of these is the result anticipated
on the basis of a cylindrical hydration layer with the
total volume involved related to the number of monomer units
rather than the hydrodynamic volume.  The second is consistent
with the Rouse model of the polymer behavior.  As the fre-
quency increases, the modes important in determining viscosity
correspond to decreasing lengths of polymer chain and at
the high frequency limit, only the residual losses associated
with the monomer prevail.  Noting that at about 200 MHz the
excess absorption in the polymer becomes indistinguishable from
that of the monomer, it becomes possible to obtain an
approximate indication of the minimum chain length required
to establish an acoustically detectable polymer-solvent complex.
By extrapolating the Rouse theory to this frequency, it is
found that a molecular weight of roughly 2,000, or about
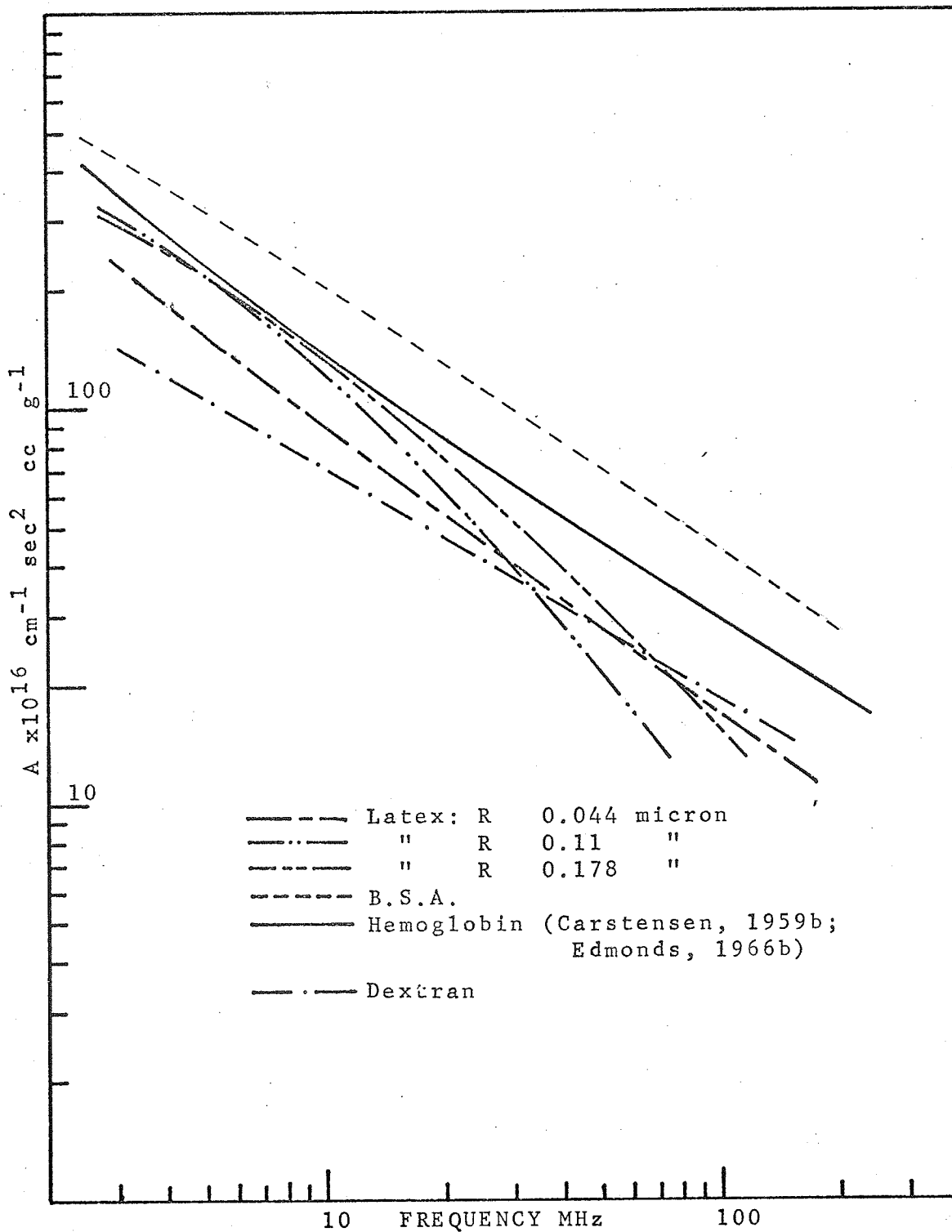11 monomer units, is required.

Figure 5-1  Composite, Absorption Data

B. Polystyrene Latex

From an acoustic standpoint, the polystyrene lattices appear to be generally similar to aqueous solutions of serum albumin and hemoglobin. This extends to the magnitude and frequency disposition of the excess absorption (Figure 5-1) as well as to the concentration dependence of the speed of sound in the suspensions (Figure 4-12). While these observations are suggestive of similarities in the absorption processes, it is necessary to examine the origin of the excess absorption in the polystyrene lattices before drawing further conclusions.

A linear dependence of the excess absorption on concentration is also observed in the polystyrene lattices to concentrations of about 10%. As is the case with macromolecular suspensions, this indicates that the excess absorption is associated with processes concerning single particles rather than particle-particle interactions.

Because of the hydrophobic character of polystyrene, emulsification of the particles can be effected only by the addition of a surface-active agent. This material resides as an adsorbed surface layer and imparts an ionic character to the particle. Disturbance of the ionic layer by the acoustic wave will result in the production of oscillatory dipoles (Enderby, 1952). If this interaction leads to net flow of charge in the region of the particle over the period of a cycle, a contribution to the attenuation will ensue. It would be expected that the observed absorption

losses would be contingent upon the ionic character of the

pervading liquid. Thus, the addition of small ions to the

liquid phase provides migratory charge carriers and

should result in the enhancement of this effect. This

is not observed experimentally, as the absorption character

is not affected by the presence of a neutral salt. In

addition, it has been observed by previous investigators

that the absorption in dispersions of kaolin platelets

is decreased by deflocculation with a surface active

agent and that the absorption character that results is in

good agreement with that predicted by relative motion of

the particles (Hueter, 1958). Thus, it seems likely that

the observed excess absorption is not attributable to

acoustic disturbances of the adsorbed layer.

The observations of previous investigators has

confirmed the validity of the Lamb-Epstein-Urick relation

(Equation 2-21) in predicting the absorption in particulate

suspensions due to relative motion (Chapter II). The

contribution computed on this basis, however, accounts at

the most for 10% of the observed absorption in the poly-

styrene lattices (Figures 4-6, 4-7, 4-8, 4-9, 4-10). As A

further check of this theory, measurements were conducted

employing a $D_2O-H_2O$ mixture as the suspending fluid with the

composition adjusted so that the density of the liquid

phase and the particle were nearly equal. Since the relation

describing this contribution contains the factor $(1 - \frac{\rho_o}{\rho_p})^2$

as a multiplicative coefficient, the relative motion

contribution should disappear in this circumstance (Chapter II). It is found, however, that the excess absorption increases despite the fact that there should be no relative motion between the particle and liquid (Figure 4-9). These observations, therefore, preclude the importance of relative motion in determining the observed absorption character.

If the excess absorption is equated to the volume viscosity of the particle, (2-25) becomes

$$K_p'' = \frac{fA(K_p')^2 V}{K_o'} \qquad (5-1)$$

$K_p''$ in turn may be evaluated from the values obtained for A using the following parameters:

$$K_o' = 2.3 \times 10^{10} \text{ dyne/cm}^2$$

$$K_p' = 4.0 \times 10^{10} \text{ dyne/cm}^2 \quad (\text{Kono, 1960})$$

$$V = 1.5 \times 10^5 \text{ cm/sec}$$

The computed value of $K_p''$ for all the aqueous lattices throughout the frequency range observed falls in the range $0.1 \times 10^9$ to $0.3 \times 10^9$ dyne/cm$^2$. While these values are not precluded by measurements of $K_p''$ conducted in bulk polystyrene (Kono, 1960), a strict comparison is not possible since the error associated with the methods employed for determining $K_p''$ in bulk materials is of the same order of magnitude.

Attributing the excess attenuation to the volume viscosity, however, is associated with two major difficulties. 1) It does not explain the relatively large absorption increase that is observed when $H_2O-D_2O$ is employed as the

suspending liquid. Velocity and density measurements in

$D_2O$-$H_2O$, reveal that $K_0^1$ in the mixture is about 3% less

than that of water so that on the basis of (5-1) only

a slight increase is anticipated. 2) Polystyrene exhibits

a primary glass transition temperature between the glassy

and rubbery states at about $100^\circ C$ (Ferry, 1961). This

transition is not associated with an obvious phase or

structural transition but rather with the time constants

which govern flow or diffusion (Ferry, 1961). Thus, as

a material changes from a glass-like to a rubber-like or

liquid state, the relaxation times involved in viscous flow

decrease rapidly from the order of minutes to times that

may be important in the ultrasonic frequency range. If the

ultrasonic absorption in this type of material is measured

as a function of increasing temperature, the observed

losses will begin to increase in the region of the transition

temperature as the spectrum of relaxation times associated

with viscous processes become comparable to the period of

the wave, then pass through a maximum, and subsequently de-

crease as the material approaches the liquid state. Longitud-

inal and shear wave absorption measurements in polystyrene

indicate that this behavior is observed with absorption

maxima for volume and shear modes occuring at about $150^\circ C$

and $130^\circ C$, respectively (Kono, 1960). The present investi-

gations are conducted for the most part at $20.0^\circ C$, well

below the glass transition temperature for both shear and

volume viscosities. Thus, it would be expected normally

that increasing the temperature of observation should be accompanied by increases in the excess absorption. The experimentally observed behavior between about $17^{\circ}$ and $50^{\circ}$ does not show this trend, but, rather, A decreases with increasing temperature in a manner similar to that found in associated liquids such as water (Figure 3-11).

Both of the above observations indicate that the excess absorption may well be associated with the suspending liquid rather than the particle. In order to examine this possibility more closely, let it be recalled that the attenuation in water is described by the relation

$$\frac{\alpha}{f^2} = \frac{2\pi^2}{\rho_0 V_0^3} \left[ \eta_v' + (4/3)\eta_s' \right] \qquad (5\text{-}2)$$

The temperature dependence of both shear and volume viscosities of water is described by an Arrhenius relation (see for example, Hirai and Eyring, 1958), i.e.,

$$\eta_s', \eta_v' = K_{s,v} \, e^{\Delta F_{s,v}^{\ddagger}/RT}$$

where $K_{s,v}$ are constants and $\Delta F_{s,v}^{\ddagger}$ are apparent activation energies associated with the viscous process concerned. The apparent activation energies are nearly the same (Herzfeld and Litovitz, 1958). Thus, assuming $\Delta F^{\ddagger'} = \Delta F_v^{\ddagger} = \Delta F_s^{\ddagger} = 4.23$ KCAL (5-2) becomes

$$\frac{\alpha}{f^2} = \frac{2\pi^2}{\rho_0 V_0^3} \left[ K_v + (4/3)K_s \right] e^{\Delta F^{\ddagger}/RT}$$

so that the apparent energy F ' is the slope of the line
generated by plotting $\ln\left[\rho_o V_o^3 \alpha / f^2\right]$ vs 1/T (Figure
5-2). If the excess absorption is the result of an
interaction with the solvent such as the production of a
layer of liquid surrounding the particle which has a some-
what higher viscosity than in pure water, one might expect
that the contribution to the macroscopic viscosity occurs
additively as it does in suspensions of viscoelastic
particles. The salient Arrhenius presentation of the data
utilizing $\ln\left[\rho_o V_o^3 A\right]$ as the ordinate parameter (Figure
5-2) reveals that the apparent activation energies at
9 MHz, 15 MHz, and 25 MHz are all about 40% of that observed
in water (Table 5-1).

TABLE 5-1

Apparent Activation Energies Associated With Excess Absorption
In DC 586 Latex.

| Frequency | $\Delta F^{\ddagger}{}'$ |
|---|---|
| 9 MHz | 1.75 K cal/mole |
| 15 MHz | 1.77 " |
| 27 MHz | 1.82 " |
| Water (All Freq.) | 4.23 " |

If the observed absorption character is, in fact,
a manifestation of a process associated with the liquid
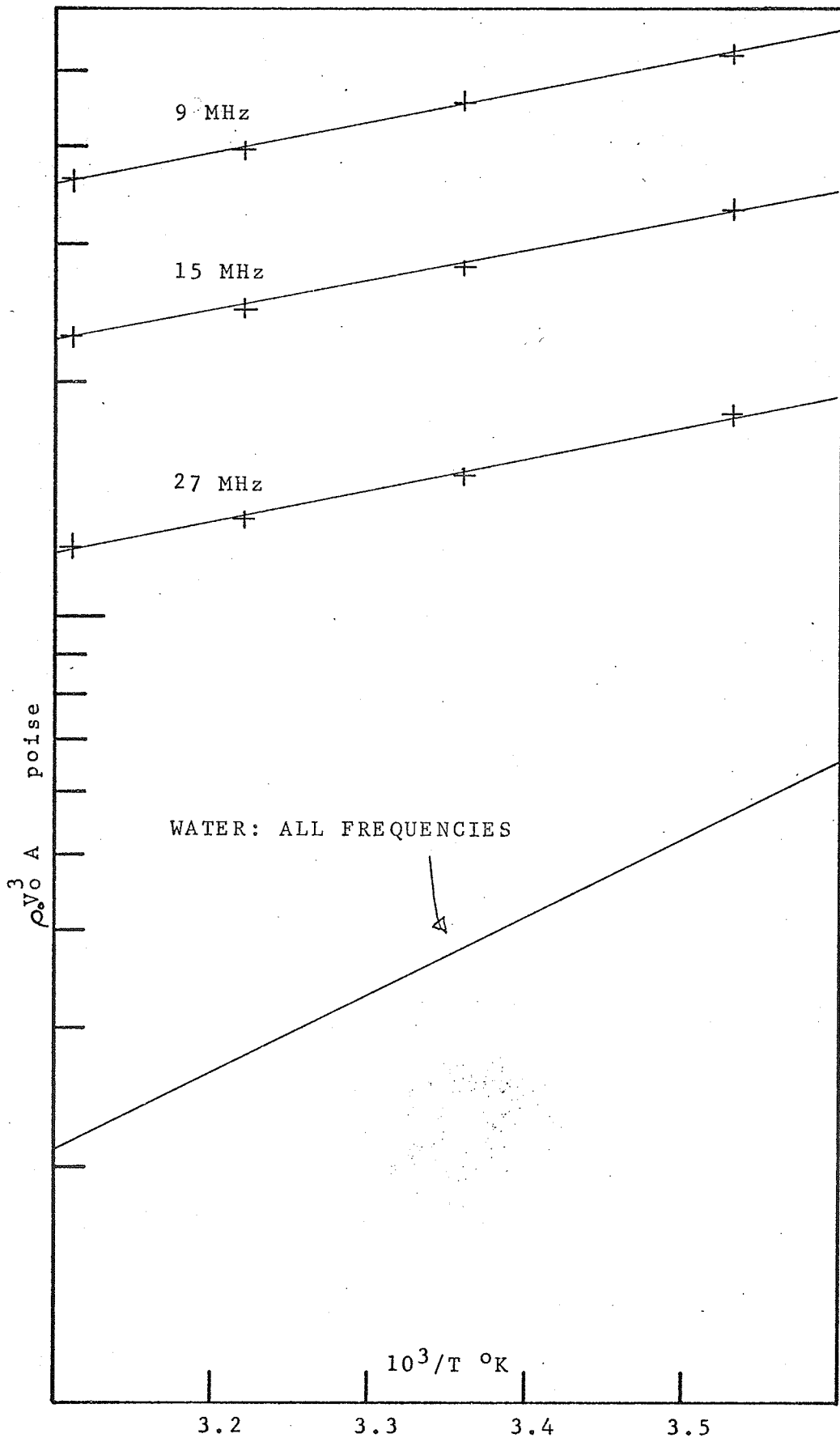independent of the internal viscosity of the particle,

Figure 5-2: ARRHENIUS PLOT OF EXCESS ABSORPTION
IN POLYSTYRENE LATEX 586

it is difficult to justify a dependence of the excess

absorption restricted solely to the physical properties of

polystyrene. Similar behavior would be expected in

aqueous dispersion of other materials. Stakutis et al.,

(1955) have investigated the temperature dependence of the

absorption in aqueous suspensions of both kaolin and

lycopodium spores in frequency range from 15 MHz to 30 MHz

and find that in both suspensions the excess absorption is

proportional to $\left[\eta'_s\right]^{1/2}$. The authors have interpreted their

results in terms of an expression relating the excess

absorption to $\left[\eta'_s\right]^{1/2}$ which they obtain by simplification of

the Epstein relation. They have, however, evidently reduced

this expression incorrectly since it has been previously shown

that the relation in its complete form predicts that the

excess absorption due to relative motion has a negligible

dependence on the viscosity of the suspending liquid (Urick,

1948). The $\left[\eta'_s\right]^{1/2}$ dependence, however, is essentially

equivalent to the result that is observed in the polystyrene

latex. In view of the diverse physical properties of the

suspended material concerned, it seems reasonable to conclude

that the origin of the excess absorption is intimately

associated with the suspending liquid. Unfortunately, it is

not possible to determine precisely the mechanism or

interaction responsible for the absorption behavior on the

basis of the above observations.

Aqueous solutions of serum albumin (Carstensen et al.,

1953) and hemoglobin (1959b) also exhibit a negative

temperature coefficient of the excess absorption in the

frequency range from 1 MHz to 10 MHz. The dependence, how-
ever, is not as pronounced as it is in the latex and seems to
be related to frequency. In view of the similarities ot the
other acoustic parameters, i.e., magnitude and frequency
dependence of excess absorption and speed of sound, it seems
likely that at least a portion of the absorption in the pro-
teins may be attributable to the same mechanisms important
in determining the absorption character of aqueous polystyrene
lattices.

## C. Scattering

The absorption coefficient in suspensions of the
largest spheres (styrene-divinylbenzene copolymer) displays
a Rayleigh fourth-power frequency dependence (Figure 4-10).
The experimental result may be analyzed in terms of (2-23),
modified to describe a distribution of particle sizes, viz.,

$$\sum_i \alpha_i = \left(\frac{2\pi f}{V_0}\right)^4 \left[\frac{\left(\frac{\rho_P}{\rho_0}-1\right)^2}{\left(\frac{\rho_P}{\rho_0}+2\right)^2} + \frac{1}{3}\left(1-\frac{K_0'}{K_1'}\right)^2\right] \sum_i R_i^3 C_i \tag{5-3}$$

where $\sum_i \alpha_i = c_v f^2 A.$

All the physical parameters necessary to evaluate this
relation are known with the exception of $K_1'$ (Table 5-2).
By employing the experimentally determined absorption and
(5-3), one can compute $K_1' \approx 4.1 \times 10^{10}$ dyne/cm$^2$. This may be
compared to a value of $3.1 \times 10^{10}$ dyne/cm$^2$ for styrene-butadiene
copolymer (Wada, 1960) and $4.0 \times 10^{10}$ dyne/cm$^2$ for polystyrene.
Thus, the Epstein relation for scattering of sound by
elastic particles appears to be compatible with the

experimental data.

TABLE 5-2

SCATTERING PARAMETERS

$$\frac{\rho_p}{\rho_o} = 1.055$$

$K'_o = 2.3 \times 10^{10}$ dyne/cm$^2$ (Calculated from velocity meas.)

$V_o = 1.48 \times 10^5$ cm/sec

$T = 20.0^{\circ}$C

Particle size distribution (Dow Chemical Co.), $c_v$ 0.1 cc/cc

| $R_i$ x $10^4$ cm | $C_i$ cc/cc |
|---|---|
| 2 | .006 |
| 3 | .047 |
| 4 | .035 |
| 5 | .010 |
| 6 | .002 |
| 7 | .0002 |

CHAPTER VI

CONCLUDING REMARKS

It has been shown that the origin of the excess absorption in aqueous suspensions of both dextran and polystyrene is intimately associated with the suspending liquid. This observation is particularly significant in regard to the polystyrene lattices. In previous investigations, generally, it has been accepted that the absorption behavior in suspensions of small rigid particles is accounted for by the Lamb-Urick-Epstein relations. In situations where departure between theory and experiment have been observed, nonextenuating explanations have been offered. Mechanisms are usually devised which are consistent with the theory. Some of these, such as the introduction of an additive to account for an additional volume viscosity (Wada et al., 1960), appear to be well founded, while others such as that of Stakutis et al., (1955) are clearly erroneous. None of these explanations, however, account for the observed absorption behavior in the polystyrene lattices. While the results of this investigation considered alone do not constitute a substantial disagreement with the theory, the similarity of the dependence of the excess absorption on the viscosity of the solvent in the polystyrene lattices and in dispersions of kaolin and lycopodium suggest that the Lamb-Urick-Epstein theory may not be an entirely realistic description of the absorption mechanisms in particulate

suspensions. Because of the fundamental nature of the problem and the importance which is attached to the Lamb-Urick-Epstein relations, it is felt that a reevaluation of the absorption properties of particulate suspensions is necessary. In addition, it seems likely that such an investigation would contribute a substantial insight into the understanding of the absorption mechanisms of solutions of macromolecules.

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APPENDICES

APPENDIX I

## COMPUTATION OF DYNAMIC PROPERTIES OF DEXTRAN

A completely flexible linear polymer whose shape is
determined solely by Brownian movement of its molecular
segments will assume the tertiary configuration of a
statistical sphere. The mean-squared end-to-end distance
$L^2$ can be deduced from random-walk arguments and has the
form (Tanford, 1961)

$$L^2 = b^2 N$$

where N is the number of identical flexible segments of
length b. The segment length b usually has a minimum
value corresponding to the length of a monomer segment $b_o$.
Any interaction which interferes with free rotation of
the monomer segments will tend to increase the end-to-end
distance over that predicted on the basis of $b_o$ as the
segment length. The description of the configuration
is conveniently handled by introducing an effective segment
$b_e$ so that

$$L^2 = N_e b_e^2$$

where $$N_e b_e = N_o b_o$$

and $$b_e \geq b_o$$

$N_o$ being the number of monomer segments. $b_e$ is also referred
to as the Kuhn statistical segment length or the hydrodynamic
segment length.

Because the dextran fractions used in this investigation

have about 5% of the glucose residues appearing as branching elements, a coefficient is necessary to account for additional excluded volume effects (Stockmayer and Fixman, 1953) so that

$$L^2 = N_e b_e^2 g$$

where g is molecular weight dependent. Granath (1958) has determined the molecular weight dependence of both $L^2$ and g for the type of dextran of interest in the present investigation. On the basis of his observations one can write

$$b_e = 2.6 \; \overline{M}_W^{0.095} \; \overset{\circ}{A}$$

for $M_W < 5 \times 10^5$. The corresponding values for $N_e$ may be evaluated on the basis of monomer molecular weight of 180 (glucose) and $b_o = 5.15A$.

The frequency range in which the Rouse theory approximation for $G'' - \omega \eta_o$ is applicable is determined by

$$2 < \omega \tau_1 < \frac{N_e^2}{250}$$

or more conveniently

$$f_{min} = \frac{1}{\pi \tau_1} < f < \frac{N_e^2}{500} = f_{max}.$$

Using equation (2-14) to determine $\tau_1$, $f_{min}$ and $f_{max}$ may be calculated. These limits as well as the values for $b_e$, $N_e$, $f_1$, $\tau_1$, are listed in Table A-1.

By combining the relation between intrinsic viscosity

TABLE A-1

| $\tilde{M}_w$ x $10^{-3}$ | $\tau_1$ x $10^6$ sec | $f_1$ (MHz) | $b_e$ Å | $N_e$ | $f_{min}$ (MHz) | $f_{max}$ (MHz) |
|---|---|---|---|---|---|---|
| 11.2 | .0274 | 5.81 | 10.7 | 30 | 12 | 21 |
| 21.8 | .0800 | 1.99 | 10.0 | 62 | 4 | 31 |
| 39.8 | .197 | .81 | 9.5 | 120 | 1.6 | 46 |
| 72.0 | .461 | .34 | 8.9 | 231 | 0.68 | 68 |
| 167.0 | 1.59 | .10 | 8.3 | 576 | 0.20 | 133 |
| 370.0 | 4.62 | .034 | 7.5 | 1411 | 0.068 | 275 |

$\tau_1$, $f_1$:  Primary relaxation time and frequency, respectively.

$b_e$:  Hydrodynamic segment length

$N_e$:  Number of statistical segments in molecule, rounded to nearest integer.

and molecular weight,

$$[\eta]_0 = .243\overline{M}_w^{0.243} \, cc/g$$

with equation (2-20), one has the result

$$\frac{\Delta\alpha}{\omega^2} = \frac{C}{2\rho_0 V_0^3}\left[\frac{0.729\, RT\eta_s}{\omega\, \overline{M}_w^{0.57}}\right]^{1/2}$$

In water at 20°C, this takes the form

$$\frac{\Delta\alpha}{C f^2} = 1.4\times10^{-11} f^{-\frac{1}{2}}\overline{M}_w^{-0.28} \, cm^{-1}sec^2 ccg^{-1}$$

which may be used to predict the shear viscosity contribution to the absorption.

## APPENDIX II

## VOLUME REQUIREMENTS FOR ULTRASONIC MEASUREMENTS

The requirements for free field diffraction make it possible to calculate the lowest frequency at which the absorption coefficient may be determined with a specified accuracy as well as the minimum sample volume required at any frequency. The requirements[1] imposed are

(i) that the maximum acoustic path length is $R^2/2\lambda$ (Pinkerton, 1949),

(ii) that $R \gtrsim 25\lambda$ (Nozdrev, 1965), and

(iii) that the minimum container radius be at least twice that of the source, i.e., $R' \gtrsim 2R$ (Del Grosso, 1965)

(where $R'$ is the radius of the container).

In addition, we assume the minimum detectable absorption increment that may be accurately measured to be approximately $0.5 dB \approx 0.06 cm^{-1}$ (Nozdrev, 1965) and that the intrinsic absorption property of the sample is separable from, and noninteracting with diffraction losses (Del Grosso, 1964).

Combining (i) and (ii) we find that

$$Z\ max \leq 315\lambda \qquad (II-A)$$

(where Z max is the maximum path length). The minimum attenuation increment requires that

$$Z\alpha \geq \delta\alpha = 0.06 cm^{-1} \qquad (II-B)$$

(where $\alpha$ is the amplitude absorption coefficient per unit path length and $\delta\alpha$ the minimum increment). Combining (II-A) and (II-B) we find that

$$\alpha\lambda \geq 2 \times 10^{-4}.$$

For water (20°C) $\alpha\lambda \approx 4 \times 10^{-11}f$ (where f is frequency) so that the minimum frequency limit becomes about 5000 KHz. The minimum volume requirement at a given frequency may be estimated as follows: for a cylindrical geometry of radius R' the volume is given by

$$V = \frac{\pi R'^2 z}{n}$$

where n is the number of transits by the pulse through the medium. From (II-B), (ii), and (iii) we find

$$V \geq \frac{\pi (50\lambda)^2 \delta\alpha}{n\alpha} \approx \frac{470 \, c^2}{n\alpha f^2}$$

For aqueous materials the velocity is usually about 1.5 x 10$^5$ cm/sec. This means that for such materials

$$V \geq \frac{10^{13}}{n\alpha f^2}$$

For water (20°C) and n = 2, the requirement would be at least four liters at 1 MHz. The necessary volume may be reduced by iterative pulse reflection, but values of n greater than 2 are generally avoided because of uncertainties that are introduced when the quartz disc is used as a reflecting surface.

1.   These conditions are stated in the original literature
so as to include appropriate "safety" margins.   Relaxation
of any or all of the requirements without serious compli-
cations is undoubtedly possible.   The requirements are
rigorously observed here, however, so that maximum assurance
of the simplest acoustic geometry is achieved.

VITA

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