$p = p_c$ is approached at constant T. Also one could test the present scaling prediction for the static equation of state in random substitutional magnetic alloys, in particular the relation $\delta = 1 + \gamma/\beta$.

Thus a simple calculation led to complicated dynamical behavior, a percolation droplet picture consistent with Monte Carlo results was developed, and experiments were suggested as tests for parts of the present predictions. I thank K. Binder for suggesting parts of this work, and him, J. W. Essam, D. P. Landau, H. Müller-Krumbhaar, D. A. Smith, and G. Toulouse for discussions and information.

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Orientation of Nucleic Acids in High Magnetic Fields

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The magnetic orientation of native and synthetic nucleic acids has been observed for the first time by measuring the Cotton-Mouton effect in magnetic fields B up to 14 T. Our data give evidence that a partial alignment of the nucleic acids takes place in the plane perpendicular to B and that the diamagnetically anisotropic bases are responsible for this orientation. The method reported here represents a new way to determine the persistence length of flexible polymers.

The magnetically induced orientation of large biological systems like cells with diamagnetic anisotropy has been observed[1-3] in magnetic fields around 1 T, and the orientation of polymer aggregates having a size of a few microns has recently been reported[4] for polystyrene solutions subjected to 1.7 T. The availability of higher magnetic fields makes possible the measurable orientation also of single macromolecules in dilute solutions. We report here the first experiments on the orientation of high-molecular-weight native deoxyribonucleic acid (DNA) and of some synthetic nucleic acids in aqueous solution in high magnetic fields up to 14 T. Using these examples we show that the method described here represents a new and general way to determine the persistence length of flexible polymers.

Let us first consider the case of a *rigid rod* made up of N diamagnetically anisotropic subunits with $\chi_\parallel > \chi_\perp$, $\chi_\parallel$ and $\chi_\perp$ being the absolute susceptibility values parallel and perpendicular to the rod axis, respectively. The magnetic energy re-

quired to turn such a rod from a position parallel to B to a perpendicular orientation is $-N(\chi_{\parallel}-\chi_{\perp})B^2/2$. The probability w_x, w_y, w_z to find a rod oriented in x, y, z respectively in Cartesian coordinates is given for B parallel to z and $N(\chi_{\parallel}-\chi_{\perp})B^2 \ll kT$, by $w_x = w_y = w_z[1+N(\chi_{\parallel}-\chi_{\perp})B^2/2kT]$, and therefore the degree of orientation β is

$$\beta = \frac{w_x - w_z}{w_z} + \frac{w_y - w_z}{w_z} = N\frac{(\chi_{\parallel}-\chi_{\perp})B^2}{kT}. \quad (1)$$

In the case of a *flexible polymer* chain, one can calculate the mean apparent diamagnetic anisotropy $\bar{\chi}_{\parallel}-\bar{\chi}_{\perp}$ by summing the susceptibility contributions in the directions parallel and perpendicular, respectively, to the direction of a given nth monomer:

$$\bar{\chi}_{\parallel}-\bar{\chi}_{\perp} = (\chi_{\parallel}-\chi_{\perp})\sum_{k=-\infty}^{\infty}\cos\theta_{nk}, \quad (2)$$

where θ_{nk} is the angle between the directions of the nth and the kth monomer. The effective radius of curvature or the stiffness of a flexible polymer is usually expressed by its Kratky-Porod persistence length[5,6]

$$P = \tfrac{1}{2}l_0\sum_{k=-\infty}^{\infty}\cos\theta_{nk}, \quad (3)$$

where l_0 is the length of one monomer. A combination of Eqs. (1)–(3) gives

$$\beta = (2P/l_0)(\chi_{\parallel}-\chi_{\perp})B^2/kT. \quad (4)$$

The degree of orientation β was determined optically by measuring the magnetically induced birefringence $\Delta n_B = n_{\parallel}-n_{\perp}$, i.e., the Cotton-Mouton effect, where $n_{\parallel}$ and $n_{\perp}$ are the refractive indices parallel or perpendicular to the applied magnetic field B, respectively. For small degrees of orientation the birefringence is proportional to β, to the anisotropy of the molecular optical polarizability $(\alpha_{\parallel}-\alpha_{\perp})$, and to the polymer concentration c: $\Delta n_B \propto (\alpha_{\parallel}-\alpha_{\perp})\beta c$, or, by introducing the Cotton-Mouton constant[7-9] C_M,

$$\Delta n = C_M\lambda B^2, \quad (5)$$

where λ is the wavelength of light. Therefore the persistence length is determined by comparison of Δn_B with the birefringence Δn_{sat} of a fully aligned polymer sample

$$\Delta n_B/\Delta n_{sat} = \tfrac{1}{6}\beta, \quad (6)$$

or by the ratio of the monomeric and polymeric Cotton-Mouton constant, $C_M(\text{mono})/C_M(\text{poly}) = 2P/l_0$.

The magnetic birefringence Δn_B was measured continuously using a photoelastic modulation technique with an automatically compensating Pockels cell as described previously.[10,11] We achieved a resolution of $\Delta n/n \simeq 2\times10^{-9}$. A Bitter solenoid with a 5-cm inner diameter was used to produce the high magnetic fields, and consequently our temperature-stabilized (± 0.05 C) sample thickness between the two deflecting mirrors was only 1 cm. For our measurements we used aqueous solutions of high-molecular-weight native calf thymus DNA,[12] of polyadenylic acid (Poly A)[12] and of polyadenylic-2-polyuridylic acid [Poly (A+2U)].[12] In the following we discuss in detail the results on DNA. Figure 1 shows the observed values of Δn_B plotted against B^2. Even at fields up to 13 T the magnetically induced birefringence is strictly linear in B^2, not reaching saturation, attaining, for example, at 12 T a value of $\Delta n = 2.31\times10^{-7}$. In order to determine from Δn_B the degree of orientation β [Eq. (6)], we measured for comparison the saturation birefringence Δn_{sat} of a completely oriented DNA solution: We produced the alignment by shearing the solution between two glass plates of 0.6 mm distance and measured Δn as a function of the relative shear velocity up to 10 mm/sec. Here, saturation of Δn was almost reached. The extrapolated value $\Delta n_{sat} = 1.57\times10^{-4}$ for $c = 10$ mg/ml leads to the *small degree* of orientation $\beta = 0.88\%$ in $B = 12$ T.

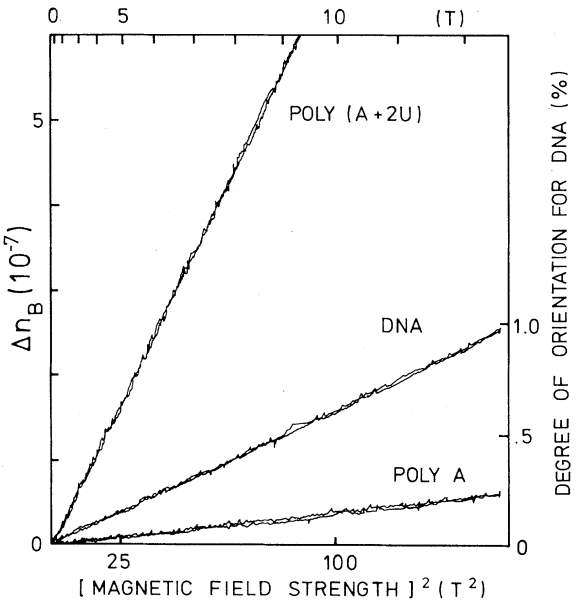

FIG. 1. Cotton-Mouton effect on aqueous solutions of calf thymus DNA (concentration $c \simeq 10$ mg/ml, $T = 20.6$ °C; the degree of orientation is determined as described below), of Poly A ($c \simeq 19$ mg/ml, $T = 19.7$°C, and of Poly(A+2U) ($c \simeq 20$ mg/ml, $T = 19.7$°C). Time of field sweep up and down, 2×3 min; $\lambda = 6328$ Å.

From signs of Δn_{sat} found by shearing parallel and perpendicular to the solenoid axis respectively, we conclude that DNA tends to orient magnetically with the filament axis *in the plane perpendicular to B.*

Figure 2 shows the concentration dependence of C_M. The linear increase of C_M with concentration up to around 5 mg/ml indicates, in agreement with Eq. (5), that in this range β does not depend on the DNA concentration; consequently, the orientation of one double helix is independent of the neighboring double helices. This lack of interaction leads one—following Eq. (4)—to expect a T^{-2} dependence of C_M, because P is proportional to T^{-1},[6] compatible with our observations below 60 C (see Fig. 3). The orientation behavior becomes of particular interest above 60 C, in the well-known[14,15] denaturation region of the DNA. Here the relatively rigid[6] DNA double helix with the base pairs[15] stacked coplanar to each other and perpendicular to the long filament axis splits into two statistical, flexible, random coils, where the bases are quasifree to rotate. In this phase-transition region (width 25°C around the melting temperature $T_M = 87$°C according to Marmur and Doty[15] for calf thymus DNA under our solvent conditions) the magnetic orientation or C_M *decreases dramatically to zero.* Following the argument above, that the magnetic orientability of a semirigid polymer decreases strongly if its monomers become free to rotate, our observed decrease of C_M leads us to assume that the base pairs are responsible for the magnetic alignment of DNA. These base pairs in part consist of aromatic, i.e. diamagnetically anisotropic,[16] rings and consequently tend to orient their planes parallel to B. The DNA filament axis therefore tends to orient itself perpendicular to B, which is also in agreement with our observation.

With use of the obtained value for β it is possible—following Eq. (4)—to determine the persistence length of DNA, since the diamagnetic anisotropy of the base pairs is known: $\chi_{\parallel} - \chi_{\perp}$ has been calculated[16] to exceed that of benzene (9.48×10^{-21} erg/T^2)[17] by a factor of 1.098 and 0.827 for adenine-thymine (A-T) and guanine-cytosine (G-C), respectively. With the known base composition of calf thymus DNA (42% G-C and 58% A-T)[15] and taking $l_0 = 3.4$ Å,[16] we find that $N = 262$ and $P = 445$ Å. The accuracy of this *absolute* value is only limited by the uncertainty of Δn_{sat} [of Eq. (6)] corresponding to full alignment and therefore is of the order of 10%. By comparison, indirect measurements of the persistence length by light-scattering[18,19] and hydrodynamic[18-21] techniques give values ranging between 250 and 1700 Å. Light-scattering results depend on the knowledge of molecular weight and its distribution[19] and also on theoretical assumptions[22] about the excluded-volume effect on the chain-folding statistics.[23] For evaluation from viscosity and sedimentation measurements, on the other hand, assumptions about the effective hydrodynamic diameter are required (ranging for DNA between 18 and 80 Å).[20] The method of magnetic orientation described here ($\Delta n/n \geq 10^{-9}$, $B \leq 15$ T) enables us to measure persistence lengths for all polymer samples fulfilling the condition $c(\alpha_{\parallel}$

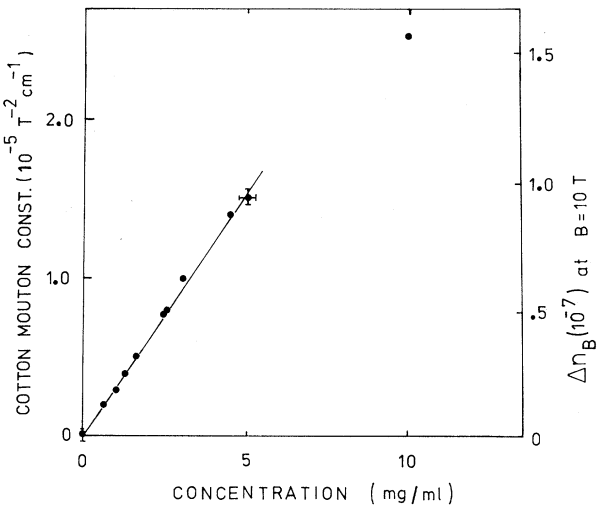

FIG. 2. Concentration dependence of the Cotton-Mouton constant C_M for calf thymus DNA. $T = 21.7$°C, other parameters as in Fig. 1.

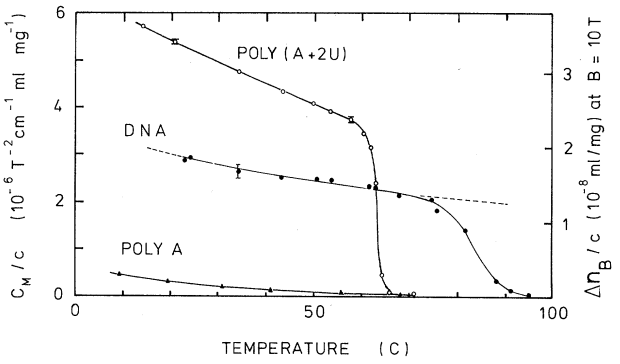

FIG. 3. Temperature dependence of the specific Cotton-Mouton constant C_M/c for (closed circles) calf thymus DNA [$c \simeq 4$ mg/ml; the dotted line is proportional to T^{-2} (K); $T_M = 87$°C according to Ref. 14], for Poly(A+2U) (open circles) ($c \simeq 20$ mg/ml, $T_M = 62$°C according to Ref. 13), and for Poly A (triangles) ($c \simeq 19$ mg/ml).

$-\alpha_\perp)(\chi_\parallel - \chi_\perp)P/l_0 \gtrsim 1\times10^{-25}$ erg/T^2. The main advantage of this method is the possibility to detect *small changes* of the persistence length with external parameters like ionic strength, pH, and temperature independently from evaluation of Δn_{sat}. We illustrate this high resolution showing the Cotton-Mouton signals and their variation with temperature in Fig. 1 and Fig. 3, respectively, for triple-helical[14,24] Poly(A+2U) and single-helical[25,24] Poly A. A more detailed presentation of the results on various polynucleotides and their interpretation is in preparation. Furthermore, corresponding studies on DNA-protein complexes and on polymer melts will be published elsewhere.

We have shown that *in magnetic fields* the small but steady orientation of macromolecules like DNA depends only on the relative position of well-defined diamagnetic parts inside the macromolecule. Furthermore, the macromolecule is always in thermal equilibrium with its ionic solvent. In contrast, the orientation behavior of polydisperse polyelectrolytic solutions *in electric fields* normally depends on contributions of constant and induced electric dipole moments[26-29] and of the hyperpolarizability[29] of the macromolecule. Therefore, in order to avoid heating effects caused by electrolytic currents, transient, nonequilibrium orientation techniques are in use and a spectrum of relaxation times has to be interpreted.

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