Toward a Statistical Theory of Catenanes. 2. Calculation of Size and Chain–Ring–Catenane Distributions with DNA-Like Polymers

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ABSTRACT: Theory of the author is applied to polymer systems resulting from equilibria between DNA-like monomers with reactive ends with Kuhn segment (KS) length of 1000 Å. Computations are made for monomer sizes containing from 1.5 to 600 KS, corresponding to molecular weights from 3×10^5 to 1.2×10^8 daltons, at concentrations from undiluted polymer to mass dilution of $1{:}10^9$ and over the entire range of possible extents of reaction. Molecular size distributions, weight and number fractions and molecular weight for chains, rings, and simplest catenanes, and overall MW's are computed. Maximum catenane yields are shown to occur, for given fraction of reaction, at intermediate concentration of monomers, growing larger with extent of reaction. For less general systems, current theory gives higher catenane predictions than previous Monte Carlo and overlap calculations. Gelation due to catenane linking is estimated not to occur for the general system, but in the special case of ring-catenane equilibrium alone may (and does, in topoisomerase experiments).

This paper presents the results of some extended calculations based on the first paper of this series,[1] using DNA-like prototype molecules, whose monomers are capable of reversible end-to-end reaction and of thereby producing distributions of chains, rings, and catenanes in equilibrium. Starting from an early chain–ring equilibrium theory of the author and Stockmayer,[2] paper 1 uses a simplistic model of equilibrium between ring pairs and two-ring catenanes to estimate the likelihood of occurrence and molecular size distribution of such catenanes, as well as of chains and rings. The model postulates a free "ring passage", from which catenanes can be formed or broken by entry or departure of one ring into or out of another, and uses the kinetics of the collision rates of both processes to predict the equilibrium between rings and catenanes. This distribution is then essentially tacked onto the known chain and ring distribution to give a complete set of predicted species, given the parameters and conditions of the polymer system.

The results of 1 were applied to a hydrocarbon-like monomer protoype, with a Kuhn segment (KS) length of 5 Å, containing 9 KS. The ring and catenane fractions are seriously overestimated for such polymer mixtures, primarily because steric hindrance stemming from finite chain radius is predominantly important in flexible polymers of low degree of polymerization. This causes a decreased fraction of ring formation and a more aggravated decrease in configurations leading to catenane formation. This kind of error parallels similar overestimates made for the special "chain-free" case of pure ring–catenane equilibrium.[3] Attempts in 1 to correct for chain radius by using the known chain radius to decrease the average distance between colliding segments proved inadequate. This decrease requires a more complex averaging, taking account of the skewed non-Gaussian distribution of segment distances, to correct the theory properly.

There are polymers made from reactable "monomers" [I use the quotation marks here to prevent confusion over the word monomer, which customarily references a DNA nucleotide. In the balance of this paper, however, the word will refer to the multinucleotide pair units which themselves cyclize or polymerize to even larger structures.] of double-strand DNA which are themselves polymers of nucleotides. These show very little problem with excluded volume, when estimates of entropy of ring formation[4] are made, provided that the monomers contain at least a few Kuhn segments. Yamakawa and Stockmayer[5] have shown that the shorter polymers are better treated as wormlike chains, on account of their stiffness. The YS theory becomes asymptotically equivalent to JS for a sufficiently large number of (the order of 10) Kuhn segments. It therefore seems that a system of polymerizable DNA monomers should be capable of providing a good test of predictions. To that end, I present calculations from the theory of 1 for DNA-like polymers. The equations, definitions and notation of 1 will be used below; equations from 1 will be prefaced with a arabic numeral 1, e.g., (1-18). The results will not be repeated; results not mentioned in 1, however, will follow.

DNA molecules can be joined together by processes which may[6] or may not[7] require a polarity of direction. If no such polarity is required, the ring–chain equilibria correspond to the "decamethylene glycol case" (case I) of JS, as used in the calculations of 1. When chains combine with a determined polarity, however, the equilibria predicted correspond to the "ω-hydroxyundecanoic acid case" (case II) of JS. The distributions in a case I or II polycondensing system differ by statistical factors, as discussed for rings and chains in JS. These appear in the definition of the ring normalization factor[8] B [This quantity, as defined for case I in 1–7, and (4) of JS, is actually a numerical chain-parameter-dependent factor which represents the concentration of one end of the chain in the neighborhood of the other. It, as well as its experimental realization, has been flatteringly called the J factor by Davidson, Baldwin, and others. However, I will continue to term it B here and use it in the strict meaning of the derived numerical.] in both 1 and JS, and of the catenane normalization factor C in 1 (and, of course, of their molar equivalents B' and C'). For case II polymers, the value of B must be doubled. The reasons can be seen from kinetic argument to be the twice-as-rapid breakup of a ring made from unpolarized monomers, compared to one made from polarized monomers; JS used a symmetry number argument. As the most promising experimental systems for testing this theory with DNA are polarized monomers, the calculations below will be made with the case II values, with B and B' twice as large as given by 1. The value of C defined for (1-9) can be seen to quadruple, as the expected number of rings doubles in case II over case I. Numerically then, the value of C' used below is fourfold that given by 1.

The number fractions of rings and catenanes (ρ_n and σ_n) parallel the weight fractions (ρ_w and σ_w) given by (1-17) and (1-16); however, they must contain the factor D, the overall number average molecular weight, so they can add up to unity

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$$\rho_n = D(B'/c)\phi'(x,5/2) \quad (1)$$

$$\sigma_n = D(C'/c)\phi'(x,5/2) \quad (2)$$

where x is the reacted fraction of the chain fraction alone and c the molar concentration of monomers, i.e., the ground molarity. Paper 1 did not give an equation for D. It is

$$D = 1/\{1 - p + (B'/c)\phi(x,5/2) + (C'/c)\phi'(x,5/2)\} \quad (3)$$

Computations

The summations leading to T2 functions were rearranged, for computational purposes, into a form referred to as "convolutional" in 1. Substituting the quantity k for $i + j$, corresponding to the monomer count in the entire catenane, the summation of $\phi'(x,5/2)$ following (1-11) becomes

$$\sum_{k=2}^{\infty}\{x^k\sum_{j=1}^{k^*}[(k + q\{j(k-j)\}^{1/2})^{3/2}\{j(k-j)\}^{-5/2}]\} \quad (4)$$

The outer summation is made over all values of possible catenane sizes, i.e. from $k = 2$ to ∞, while the inner sum limits are now from $j = 1$ to k^*, where, since j must be the smaller or equal of the two ring sizes, k^* is the largest possible value for the smaller (or equal) ring and is the truncated value of $k/2$. The other T2 functions were similarly rearranged for calculation.

As an intermediate step, 9600 values of the internal sum over j were computed and put into a preliminary database, for use in performing the outer summation. Values of $x > 0.997$ required more than 9600 terms in the inner sum, due to slow convergence of the internal series. A set of corrections was prepared for the later values of the slowly converging T2 functions of large x, for $j = 9601$ to ∞, based upon the empirical fit of the internal sum, for large j, to $1.354/j$. Summation of the remaining terms of the external sum was replaced by integration. The correction values, obtained in closed form as exponential integral functions, were within about 1% of the true values. Appendix 1 gives the relations for the corrections.

The same kind of approximation was used to compute the T1 functions, which are based on a single sum which slowly converges for arguments near unity. These were similarly corrected for with approximation integrals, in this region, and they were obtainable as complementary error functions, as described in Appendix I. Although Truesdell[9] gave alternate series for getting the T1 functions at high values of x, the method given in Appendix 1 is simpler and better suited to machine computation parallel with the T2 summations.

As the computation of the T1, and especially the T2, sums is laborious, even for a computer, a database including all six functions was prepared, and electronically stored, for a set of 400 values of x, ranging from 0.0001 to 0.999 999 5. The maximum value of x, i.e., x^*, for which p, the overall fraction of end groups reacted, equals unity, was initially approximated by interpolations using these values of x. For $x > x^*$, the equations give a value of p above unity, and other spurious values, including a negative chain weight fraction. Although this is physically impossible, the values of x^* so calculated correspond to a system completely reacted and containing only rings and catenanes. Using Newton's method of successive approximations and starting with pairs of known values of x and p, in the region of $p = 1$, x^* was calculated. Hunt for the values of x at which $p = 0.9$ and $p = 0.5$ was made by using the same program, with slight alterations. The values of all variables in a system with specified p were calculated with the same algorithms, but with the hunted value of x as new input.

Numerical Results with DNA-Like Systems

Expected distributions and molecular weights were calculated from a prototype monomer similar to double-stranded DNA, with a KS length of 1000 Å. Monomer sizes considered ran from 1.5 to 600 KS, corresponding to 0.5–200 kilobase pairs (Kbp) or molecular weights of from 3×10^5 to 1.2×10^8 daltons. The values of the constants B' and C' for a polarized monomer were calculated from the case II numerical factors B and C, divided by 6×10^{20}. They vary with the square root of ν, the number of KS. The two sizes principally reported here are monomers with $\nu = 150$ and 15 KS. The former, corresponding approximately in size to a lambda phage genome, has values of 3.0×10^{-10} and 2.4×10^{-11} for B' and C', respectively. The latter, one tenth as long, resembles a DNA molecule with about 5 Kbp and represents a nearly optimum size monomer for experimental testing of 1. For this monomer, B' and C' are 9.43×10^{-9} and 7.6×10^{-10} mol/L, respectively.

For these DNA-like polymers with various monomers and concentrations, the following quantities were calculated: 1, the number average degrees of polymerization of the chain, ring, and catenane fractions, designated D_c, D_r, and D_{ca}, the weight average degrees, designated D_{cw}, D_{rw}, and D_{caw}, respectively, and also the number and weight ratios of catenanes to rings, I''/I' and N''/N'; 2, the molar concentrations of rings and catenanes; 3, the overall fraction of reaction, p; 4, the number and weight fractions of chains, rings, and catenanes (chain fractions follow as unity minus the ring and catenane fractions); 5, the overall number and weight degree of polymerization, D and D_w.

The first set of these quantities depends only on x. The remainder of the quantities depend on the monomer size, which fixes the system constants B' and C', and on the monomer molar concentration, c. Computation is most efficient when all desired quantities are computed with a single value of x, using varying concentrations. Tabulation, however, is best made for single concentrations, over the possible range of x. The following quantities are presented: 1, the system-independent values of the T1 and T2 sums and the number and weight average molecular weights of the chains, rings, and catenanes, as functions of x, from 0.0001 to 0.999 999 5 (the T1 and T2 sums are given in Table I here; Table II gives the average molecular weights of the three molecule types, along with I''/I' and N''/N'; these are also independent of B', C', and c; it should be noted that the catenane–ring ratios in 1 were calculated for a case I polymer system; these ratios are only half of those for a case II polymer, on account of statistical factors multiplying B and C; the number averages given here were not discussed or presented in 1; 2, system values for the DNA-like polymers (the quantities p, D, D_w, ρ_n, ρ_w, σ_n, and σ_w, are given in Tables III–V for the larger monomer type, at three concentrations: 6.8×10^{-5}, 1.6×10^{-10}, and 3.2×10^{-14} mol/L; the first of these concentrations represents an approximate solvent-free monomer mix, which I designate as c_{max}, when expressed as a molar concentration; the second corresponds approximately to the concentration used by Wang and Davidson,[4] and the third is an ultra-dilute solution; Table VI represents the smaller monomer, at concentration of 1.6×10^{-9} M, one corresponding in mass dilution to that of Table IV); 3, the value of x^*, at which $p = 1$ (in each of Tables III–VI, the last value of x is x^*, calculated with the approximation method mentioned above; the relation between x^* and c is given in Figure 1; the concentration, as fraction of maximum, c/c_{max}, is

Table I
T1 and T2 Functions

x	$\phi(x,^5/_2)$	$\phi(x,^3/_2)$	$\phi(x,^1/_2)$	$\phi'(x,^5/_2)$	$\phi'(x,^3/_2)$	$\phi'(x,^1/_2)$
0.0001	1.00×10^{-4}	1.00×10^{-4}	1.00×10^{-4}	5.48×10^{-8}	1.10×10^{-7}	2.19×10^{-7}
0.0002	2.00×10^{-4}	2.00×10^{-4}	2.00×10^{-4}	2.19×10^{-7}	4.38×10^{-7}	8.77×10^{-7}
0.0005	5.00×10^{-4}	5.00×10^{-4}	5.00×10^{-4}	1.37×10^{-6}	2.74×10^{-6}	5.48×10^{-6}
0.001	1.00×10^{-3}	1.00×10^{-3}	1.00×10^{-3}	5.48×10^{-6}	1.10×10^{-5}	2.19×10^{-5}
0.002	2.00×10^{-3}	2.00×10^{-3}	2.00×10^{-3}	2.19×10^{-5}	4.39×10^{-5}	8.78×10^{-5}
0.005	5.00×10^{-3}	5.01×10^{-3}	5.02×10^{-3}	1.37×10^{-4}	2.74×10^{-4}	5.50×10^{-4}
0.01	1.00×10^{-2}	1.00×10^{-2}	1.01×10^{-2}	5.50×10^{-4}	1.10×10^{-3}	2.21×10^{-3}
0.02	2.01×10^{-2}	2.01×10^{-2}	2.03×10^{-2}	2.21×10^{-3}	4.42×10^{-3}	8.89×10^{-3}
0.05	5.05×10^{-2}	5.09×10^{-2}	5.18×10^{-2}	1.39×10^{-2}	2.81×10^{-2}	5.69×10^{-2}
0.10	1.02×10^{-1}	1.04×10^{-1}	1.08×10^{-1}	5.67×10^{-2}	1.15×10^{-1}	2.37×10^{-1}
0.20	2.08×10^{-1}	2.16×10^{-1}	2.34×10^{-1}	2.35×10^{-1}	4.90×10^{-1}	1.05
0.30	3.18×10^{-1}	3.38×10^{-1}	3.85×10^{-1}	5.54×10^{-1}	1.19	2.65
0.50	5.55×10^{-1}	6.25×10^{-1}	8.06×10^{-1}	1.72	3.99	1.03×10
0.70	8.22×10^{-1}	1.00	1.58	3.95	1.06×10	3.64×10
0.80	9.72×10^{-1}	1.26	2.34	5.78	1.78×10	7.98×10
0.90	1.14	1.61	4.02	8.69	3.49×10	2.70×10^2
0.95	1.23	1.88	6.38	1.11×10	6.09×10	8.92×10^2
0.97	1.27	2.03	8.70	1.27×10	8.99×10	2.20×10^3
0.99	1.32	2.27	1.62×10	1.53×10	2.10×10^2	1.66×10^4
0.995	1.33	2.37	2.36×10	1.67×10	3.71×10^2	6.20×10^4
0.999	1.34	2.50	5.46×10	1.94×10	1.54×10^3	1.43×10^6
0.9995	1.34	2.53	7.78×10	2.04×10	2.95×10^3	5.60×10^6
0.9999	1.34	2.57	1.76×10^2	2.28×10	1.40×10^4	1.37×10^8
0.99995	1.34	2.59	2.49×10^2	2.37×10	2.76×10^4	5.44×10^8
0.99999	1.34	2.60	5.59×10^2	2.59×10	1.36×10^5	1.35×10^{10}
0.999995	1.34	2.60	7.91×10^2	2.69×10	2.71×10^5	5.42×10^{10}
0.999999	1.34	2.61	1.77×10^3	2.90×10	1.36×10^6	1.35×10^{12}
0.9999995	1.34	2.61	2.51×10^3	3.00×10	2.71×10^6	5.42×10^{12}
1.0	1.3415	2.6006	∞	∞	∞	∞

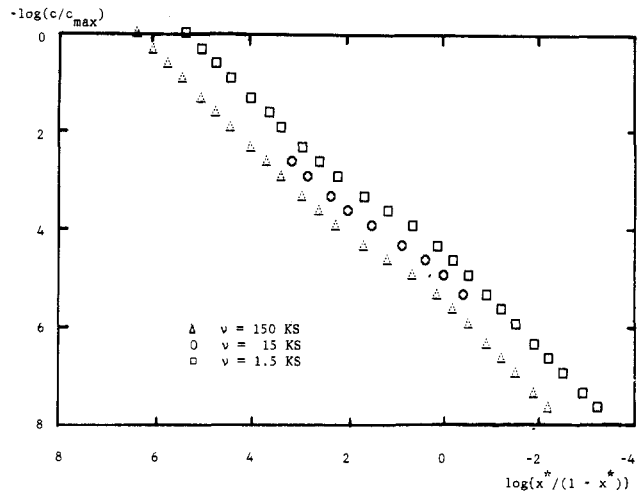

Figure 1. Relation between concentration and maximum x (x^*) for various values of ν, for DNA-like polymer ($b = 10^{-5}$ cm).

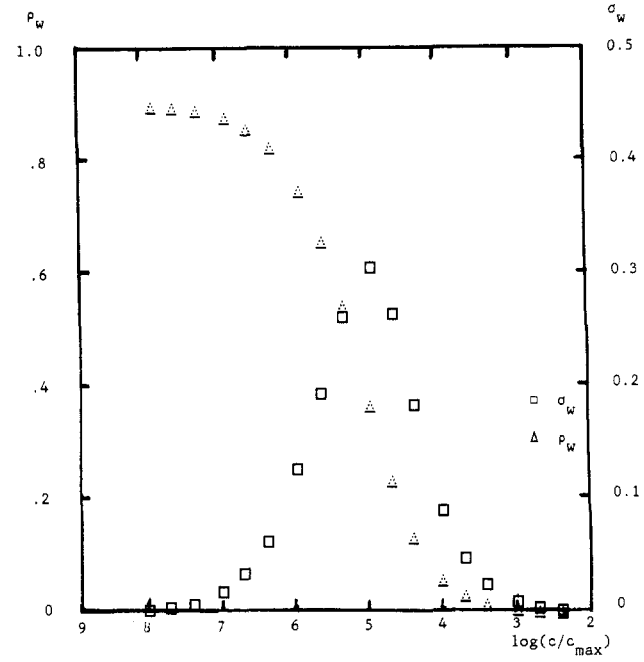

Figure 2. Weight fractions of rings and catenanes of DNA-like polymer of 150 KS at 90% reaction, as function of concentration.

plotted for three different sized monomers, 150, 15, and 1.5 KS, from the maximum value to high dilution; the function $x^*/(1 - x^*)$, is plotted against the concentration, in a log–log relation); 4, the weight fractions of rings and catenanes plotted directly against the logarithm of the concentration in Figures 2 and 3 (Figure 2 represents values of x for which $p = 0.9$ and Figure 3 for $p = 0.5$).

Discussion

The results of the model of 1 can be further amplified here, from the extensive calculations performed by using computer programs based upon the equations of 1. The existing databases of inner sums and T2 sums depend on the assumptions made. The T1 and T2 functions, as I emphasized in paper 1, are generic; change of assumptions, size corrections, or mathematically altered equations will give a new database set. Nevertheless, the programs used, apart from the subroutines needed to calculate the sums, are perfectly general. Given some other and better numerical coefficients or distribution functions, they could be easily modified to compute all the needed quanties by replacing subroutines or databases used. For reasonably long DNA, there is little correction for excluded volume or chain stiffness. However, short DNA gives large effects in both of these directions. Very large DNA also shows considerable excluded volume and will fail to follow the equations of 1 accurately, particularly showing a different dependence of B and C on the chain length from that of the simple Kuhn relation used in 1 and in JS.

Some comments on the functional nature of the T2 database quantities are in order. The sum over j of $[k + q\{j(k-j)\}^{1/2}]^{3/2}\{j(k-j)\}^{-5/2}$ equals approximately $1.354/k$, for high k. Computation of the T2 functions confirmed the divergence of all three of these sums at $x = 1$. Near

Table II
System-Independent Values for a Case II Chain–Ring–Catenane Equilibrium

x	I''/I'	N''/N'	D_c	D_{cw}	D_r	D_{rw}	D_{ca}	D_{caw}
0.0001	4.42×10^{-5}	8.84×10^{-5}	1.00	1.00	1.00	1.00	2.00	2.00
0.0002	8.84×10^{-5}	1.77×10^{-4}	1.00	1.00	1.00	1.00	2.00	2.00
0.0005	2.21×10^{-4}	4.41×10^{-4}	1.00	1.00	1.00	1.00	2.00	2.00
0.001	4.42×10^{-4}	8.84×10^{-4}	1.00	1.00	1.00	1.00	2.00	2.00
0.002	8.84×10^{-4}	1.76×10^{-3}	1.00	1.00	1.00	1.00	2.00	2.00
0.005	2.21×10^{-3}	4.42×10^{-3}	1.00	1.01	1.00	1.00	2.00	2.00
0.01	4.43×10^{-3}	8.85×10^{-3}	1.01	1.02	1.00	1.00	2.00	2.00
0.02	8.87×10^{-3}	1.77×10^{-2}	1.02	1.04	1.00	1.01	2.01	2.01
0.05	2.23×10^{-2}	4.45×10^{-2}	1.05	1.11	1.01	1.02	2.02	2.03
0.07	3.13×10^{-2}	6.25×10^{-2}	1.07	1.15	1.01	1.03	2.02	2.04
0.1	4.49×10^{-5}	8.97×10^{-2}	1.11	1.22	1.02	1.04	2.04	2.06
0.2	9.15×10^{-2}	1.83×10^{-1}	1.25	1.50	1.04	1.08	2.08	2.13
0.3	1.40×10^{-1}	2.83×10^{-1}	1.43	1.86	1.06	1.14	2.14	2.24
0.5	2.50×10^{-1}	5.16×10^{-1}	2.00	3.00	1.13	1.29	2.32	2.59
0.7	3.89×10^{-4}	8.55×10^{-1}	3.33	5.67	1.22	1.57	2.69	3.42
0.8	4.80×10^{-1}	1.14	5.00	9.00	1.30	1.86	3.08	4.48
0.9	6.15×10^{-1}	1.74	1.00×10	1.90×10	1.42	2.49	4.01	7.73
0.95	7.28×10^{-1}	2.61	2.00×10	3.90×10	1.53	3.38	5.47	1.46×10
0.97	8.02×10^{-1}	3.56	3.33×10	6.57×10	1.60	4.27	7.10	2.45×10
0.99	9.39×10^{-1}	7.46	1.00×10^2	1.99×10^2	1.72	7.14	1.37×10	7.90×10
0.995	1.01	1.26×10	2.00×10^2	3.99×10^2	1.78	9.95	2.22×10	1.67×10^2
0.997	1.07	1.91×10	3.33×10^2	6.66×10^2	1.82	1.27×10	3.26×10	2.90×10^2
0.999	1.17	4.98×10	1.00×10^3	2.00×10^3	1.87	2.18×10	7.95×10	9.24×10^2
0.9995	1.23	9.41×10	2.00×10^3	4.00×10^3	1.89	3.07×10	1.44×10^2	1.89×10^3
0.9997	1.27	1.52×10^2	3.33×10^3	6.67×10^3	1.90	3.95×10	2.27×10^2	3.20×10^3
0.9999	1.37	4.37×10^2	1.00×10^4	2.00×10^4	1.92	6.82×10	6.14×10^2	9.78×10^3
0.99995	1.42	8.60×10^2	2.00×10^4	4.00×10^4	1.93	9.63×10	1.16×10^3	1.97×10^4
0.99997	1.46	1.42×10^3	3.33×10^4	6.67×10^4	1.93	1.24×10^2	1.87×10^3	3.30×10^4
0.99999	1.56	4.22×10^3	1.00×10^5	2.00×10^5	1.94	2.15×10^2	5.25×10^3	9.96×10^4
0.999995	1.62	8.41×10^3	2.00×10^5	4.00×10^5	1.94	3.04×10^2	1.01×10^4	2.00×10^5
0.999997	1.66	1.40×10^4	3.33×10^5	6.67×10^5	1.94	3.92×10^2	1.64×10^4	3.33×10^5
0.999999	1.75	4.19×10^4	1.00×10^6	2.00×10^6	1.94	6.79×10^2	4.67×10^4	1.00×10^6
0.9999995	1.80	8.37×10^4	2.00×10^6	4.00×10^6	1.95	9.60×10^2	9.04×10^4	2.00×10^6

Table III
Characteristics of a DNA-Like Chain–Ring–Catenane System at Equilibrium[a]

x	p	ρ_n	ρ_w	σ_n	σ_w	D	D_w
0.10	0.1000	0.0000	0.0000	0.0000	0.0000	1.11	1.22
0.20	0.2000	0.0000	0.0000	0.0000	0.0000	1.25	1.50
0.50	0.5000	0.0000	0.0000	0.0000	0.0000	2.00	3.00
0.70	0.7000	0.0000	0.0000	0.0000	0.0000	3.33	5.67
0.90	0.9000	0.0000	0.0000	0.0000	0.0000	1.00×10	1.90×10
0.95	0.9500	0.0001	0.0000	0.0001	0.0000	2.00×10	3.90×10
0.99	0.9900	0.0006	0.0000	0.0005	0.0001	9.99×10	1.99×10^2
0.995	0.9950	0.0012	0.0000	0.0012	0.0001	2.00×10^2	3.99×10^2
0.999	0.9990	0.0058	0.0000	0.0068	0.0005	9.88×10^2	2.00×10^3
0.9995	0.999 50	0.0115	0.0000	0.0141	0.0010	1.95×10^3	4.00×10^3
0.9999	0.999 90	0.0518	0.0000	0.0710	0.0049	8.82×10^3	1.99×10^4
0.999 95	0.999 950 5	0.0922	0.0000	0.1315	0.0098	1.57×10^4	3.98×10^4
0.999 99	0.999 990 5	0.2394	0.0000	0.3732	0.0481	4.07×10^4	1.95×10^5
0.999 995	0.999 995 5	0.2955	0.0000	0.4774	0.0960	5.02×10^4	3.81×10^5
0.999 999	0.999 999 5	0.3527	0.0000	0.6161	0.4793	6.00×10^4	1.52×10^6
0.999 999 52	1.0	0.3564	0.0000	0.6436	1.0	6.06×10^4	2.09×10^6

[a] ν = 150 KS/monomer; $c = 6.8 \times 10^{-5}$ M.

Table IV
Characteristics of a DNA-Like Chain–Ring–Catenane System at Equilibrium[a]

x	p	ρ_n	ρ_w	σ_n	σ_w	D	D_w
0.005	0.0143	0.0094	0.0093	0.0000	0.0000	1.00	1.01
0.01	0.0286	0.0189	0.0187	0.0001	0.0002	1.01	1.02
0.02	0.0574	0.0382	0.0375	0.0003	0.0007	1.02	1.04
0.05	0.1441	0.0988	0.0949	0.0022	0.0042	1.05	1.10
0.07	0.2022	0.1414	0.1388	0.0044	0.0084	1.07	1.14
0.1	0.2896	0.2088	0.1933	0.0094	0.0173	1.10	1.20
0.15	0.4355	0.3298	0.2957	0.0224	0.0402	1.15	1.30
0.2	0.5809	0.4598	0.4024	0.0421	0.0737	1.19	1.38
0.25	0.7248	0.5956	0.5138	0.0689	0.1192	1.22	1.44
0.3	0.8661	0.7318	0.6305	0.1028	0.1783	1.24	1.47
0.32	0.9217	0.7849	0.6787	0.1183	0.2062	1.24	1.47
0.34	0.9766	0.8365	0.7280	0.1347	0.2366	1.23	1.46
0.3486	1.0	0.8580	0.7494	0.1420	0.2506	1.23	1.45

[a] ν= 150 KS/monomer; $c = 1.6 \times 10^{-10}$ M.

Table V
Characteristics of a DNA-Like Chain–Ring–Catenane System at Equilibrium[a]

x	p	ρ_n	ρ_w	σ_n	σ_w	D	D_w
0.000 01	0.0932	0.0932	0.0932	0.0000	0.0000	1.00	1.00
0.000 02	0.1864	0.1864	0.1864	0.0000	0.0000	1.00	1.00
0.000 03	0.2796	0.2796	0.2796	0.0000	0.0000	1.00	1.00
0.000 05	0.4659	0.4659	0.4659	0.0000	0.0000	1.00	1.00
0.000 07	0.6523	0.6523	0.6523	0.0000	0.0000	1.00	1.00
0.0001	0.9319	0.9319	0.9318	0.0000	0.0001	1.00	1.00
0.000 107 3	1.0	1.0000	0.9999	0.0000	0.0001	1.00	1.00

[a] ν = 150 KS/monomer; $c = 3.2 \times 10^{-14}$ M.

that point, $\phi'(x,{}^5/_2)$ is a very slowly divergent function, increasing with $1/(1-x)$ at approximately a log–log rate; $\phi'(x,{}^3/_2)$ increases linearly, and $\phi'(x,{}^1/_2)$ increases quadratically. $\phi(x,{}^1/_2)$, the only diverging T1 function, increases

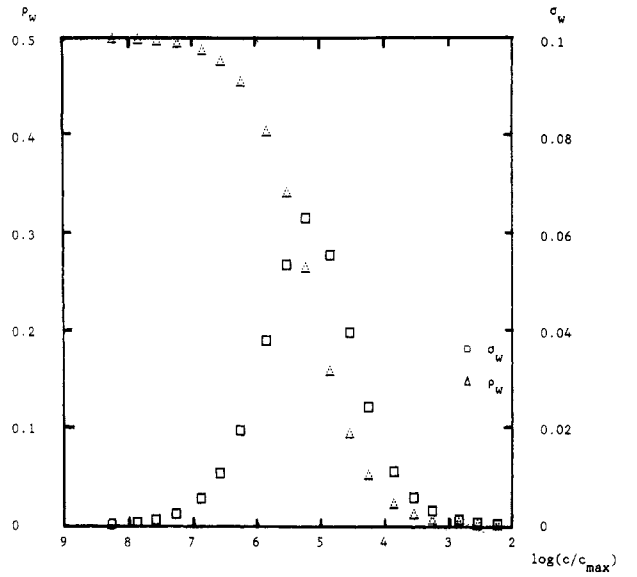

Figure 3. Weight fractions of rings and catenanes of DNA-like polymer of 150 KS at 50% reaction, as function of concentration.

Table VI
Characteristics of a DNA-Like Chain-Ring-Catenane System at Equilibrium[a]

x	p	ρ_n	ρ_w	σ_n	σ_w	D	D_w
0.001	0.0059	0.0059	0.0059	0.0000	0.0000	1.00	1.00
0.002	0.0138	0.0118	0.0118	0.0000	0.0000	1.00	1.00
0.005	0.345	0.0296	0.0295	0.0001	0.0001	1.00	1.01
0.01	0.0691	0.0596	0.0591	0.0003	0.0005	1.01	1.02
0.02	0.1384	0.1206	0.1187	0.0011	0.0021	1.02	1.04
0.05	0.3477	0.3109	0.3000	0.0069	0.0133	1.05	1.09
0.08	0.5585	0.5105	0.4854	0.0183	0.0347	1.07	1.13
0.10	0.6996	0.6470	0.6114	0.0290	0.0548	1.07	1.16
0.12	0.8409	0.7848	0.7393	0.0424	0.0799	1.08	1.17
0.13	0.9115	0.8537	0.8040	0.0501	0.0943	1.08	1.17
0.14	0.9822	0.9222	0.9893	0.0584	0.1100	1.09	1.17
0.14252	1.0	0.9394	0.8858	0.0606	0.1142	1.09	1.17

[a] ν = 15 KS/monomer; $c = 1.6 \times 10^{-9}$ M.

approximately as $(1 - x)^{-1/2}$ near $x = 1$.

The predictive consequence of these divergences was discussed in 1, namely, that at sufficiently high values of x, one can get a 100% nonchain distribution at $p = 1$ *for any concentration.* While it may seem obvious that at $p = 1$ there can be no free polymer ends [One must disallow closing of the fast "infinite" chain ends into a macroscopic ring at $p = 1$, since this ring, as well as being extremely unlikely, does not rightly belong to the ring molecule distributions.], one might expect that the chain fraction would have an infinite molecular weight at that point. In JS it was shown that at concentrations weaker than a "critical dilution", corresponding to $(B'/c)\phi(x,^3/_2) = 1$, the system is composed of 100% rings at $p = 1$ (catenanes being assumed absent). The ring fraction alone can never equal unity above this concentration, even if catenanes are forbidden. The corresponding relation, (1-24), defines x^*, a maximum value of x, where $p = 1$, and all end groups (i.e. chains) have disappeared. And, since $\phi'(x,^3/_2)$ diverges at $x = 1$, there can be no concentration at which the reaction cannot be at least theoretically driven to completion to give only rings and catenanes. However, at high concentration (Table III), the mixture moves toward 100% catenanes, while at low ones (Table V), nearly 100% rings are produced.

The value of x^* is highly concentration-dependent, however, as shown in Figure 1. At low concentration, Figure 1 shows x^* to be a linear function of the concentration; for high concentrations, it is linear in $1/(1 - x)$. Using $x/(1 - x)$ as the abscissa, the graph shows both of these regions to have unity as logarithmic slope, with a hitch between the regions at intermediate concentrations, at values of x between 0.5 and 0.999 or so. This function is ruled by its numerator at small values of x and by its denominator near $x = 1$. For concentrated solutions, chains remain the predominant species, as is well-known. At very high values of x, however, the T2 functions become large, and the catenane fraction and its molecular weight become considerable. Curiously, the weight average molecular weight of the catenanes above $x = 0.999$ proves to be about half of the weight average molecular weight of the chains. The far more heterogeneous catenanes, however, have a much lower number average molecular weight than the chains. The rings remain a small portion of the total number of molecules, and their weight fraction becomes vanishingly small. When $x = x^*$ and $p = 1$, an equilibrium mixture of rings and catenanes consists almost exclusively of catenanes, at high x. For the 150 KS DNA-like prototype used, x^* for undiluted polymer becomes 0.999 999 52, a practically unreachable value corresponding to chains of nearly 2 million monomers; or about 500 billion daltons.

In dilute solutions, however, the chains never get very long, as x^* becomes quite low. At $p = 1$, the system portrayed in Table V, with a concentration of 3.2×10^{-14} M (a mass dilution of about 2×10^9:1), almost no catenanes can be discerned, largely because of the minute value of the T2 functions, compared to the T1. Here rings comprise nearly 100% of the mass at equilibrium; almost every ring and chain present is a monomer, and the very few catenanes are dimers. The calculated distributions indicate the maximum catenane size and yield occurs with a concentrated solution whose reaction is driven to completion. In reality, the maximum polymer size is limited by monomer imperfections and by the kinetic impossibility of reaching completion. Examination of the results of Figures 2 and 3, calculated at constant p of 0.9 or 0.5, shows the catenane fraction to reach a maximum at some intermediate concentration low enough to foster ring formation but high enough to produce reasonable values of the T2 functions. The higher the p, the larger the maximum fraction of catenanes.

Comparison with Previous Theory

The first calculations predicting appearance of catenanes in DNA were made by Wang and Davidson,[11] using the overlap concepts of Frisch and Wasserman,[3] combined with some JS equilibrium theory. They showed that an equilibrium expectation existed for some catenanes, whenever cyclic dimers were present, and that the ratio of the simplest catenane structure numbers to that of cyclic dimers was *independent of monomer size, concentration, or degree of reaction.* Verification of this relationship is given in Appendix II. The superposition of a ring passage equilibrium on the general system ensures that ring–catenane ratios are the same for both the chain-free and the general systems. However the numerical results of 1, based on Ca_{11} (1-9) and R_2 (1-7), give a catenane-to-dimer ring ratio of 2.5, while the Wang–Davidson estimate is 0.19. [A factor of $2^{5/2}$ was inadvertently omitted in their paper, which reported the ratio as 0.033.] Although there is some uncertainty in any overlap prediction, a factor this large is substantial. It is probably due to a fundamental difference between the theory of 1 and simple overlap. Equation 1-5 sets the mean square distance between the segments of the chain at a value which is, for 1,1-catenanes, over threefold greater than that from overlap as used by Wang and Davidson; use of the root-mean-square distance

to the third power in (1-9) accounts for most of the discrepancy. A band slightly above background was noted in their ultracentrifuge runs at the approximate point where ring dimers should have showed up. Such a band would have run at a slightly slower rate than a 1,1-catenane band, which may or may not have been present; Wang and Davidson did not report scrutiny of the band contents with electron microscopy.

An experiment shortly afterward by Wang and Schwartz[12] confirmed the presence of some catenanes in cyclizable DNA systems, although not under conditions which the unaltered theory of 1 can directly treat. Two separate and mutually unreactive cohesive end DNA monomers, λ and 186, were used. λ, the longer one, was labeled with 5-bromouracil, while the 186 DNA was totally cyclized. A preparation of opened circles of λ, i.e. linear monomers, was then allowed to reach equilibrium in the presence of a relatively high concentration of the 186 rings. Density gradient equilibrium sedimentation of the reacted mixture then segregated the two species of DNA, lumping all individual molecules from each monomer into a single band. The appearance of a small but definite band of intermediate density showed unequivocal evidence of a compound molecule that could only have formed by threading and closing of the λ monomers within the 186 rings. Further small bands indicated catenane formation between λ monomers and cyclic dimers and trimers of the 186. More complex catenanes could have also been present, but only in small amounts, imperceptible here.

The importance of this experiment here is in the overlap calculations made by Wang and Schwartz, in which the expectation of catenanes was found to be about threefold higher than observed. Besides the imprecision of overlap predictions, uncyclizable monomers of either type, or steric hindrance, would lower the yield of properly threaded and closed catenanes. The difficulty of making comparisons with the data of Wang and Schwartz stems from several sources: 1. The theory of 1 does not make provision for more than one different type of monomer, although doing so presents no insuperable problems. However, their system, if at equilibrium, could well show different values of p for the two monomer species. Their presumption was stability for the 186 rings and equilibrium for the threading and cyclizing λ monomers. 2. They ignored the β factor of Frisch and Wasserman,[3] measuring the degree to which overlapping rings actually interlock. 3. They furthermore took a linear mean-square radius for the 186 DNA, and a cyclic one for the λ DNA, and a value of 700 Å for b. As the fraction of rings and catenanes is quite sensitive to these factors, any disagreement between their observations and calculations is quite understandable.

Further comparison can be made between the theory of 1 and Frank-Kamenetskii et al.[10] (FLV), who made Monte Carlo calculations of linkage probabilities of nearby closed chains, corresponding to the chain-free case of 1. Their eq 8, corresponding to (1-26), gives an estimate of the fraction of catenanes that can be directly measured against that of 1

$$C_c = c^2B$$

where C_c is the catenane concentration, c the concentration of ring monomers, and B here [Their symbol B should not be confused with the ring normalization factor.] the second virial coefficient given in terms of the volume b^3, where b is the KS length, as above. For monomers of 20, 40, 60, and 80 KS, they calculated second virial coefficients of 17, 60, 143, and 228, respectively. Multiplying this coefficient by 10^{-15}, the cube of the Kuhn segment length used here for DNA-like prototype, by 6.02×10^{20}, and then by the concentration of monomers in moles/liter, the fraction of catenanes is directly obtained. For $\nu = 20$ KS, the FLV catenane fraction is $1.0 \times 10^7 c$; for $\nu = 80$ KS, it is $1.4 \times 10^8 c$. A log–log extrapolation to $\nu = 150$ KS leads to a B of about 800 and to a catenane concentration of about $1.5 \times 10^9 c$. Equation 1-26 gives, for a 20 KS monomer, $7.2 \times 10^7 c$, for 80 KS, $5.7 \times 10^8 c$, and for 150 KS, $1.5 \times 10^9 c$. A noteworthy distinction between the two theories is the apparent dependence of the catenane fraction on ν. The FLV log–log graph indicates a dependence to the 1.9 power of ν, while paper 1 predicts a dependence to the 1.5 power. For monomers shorter than λ, 1 predicts more catenanes, while for larger ones, presumably FLV would predict more. However, in view both of the questionable validity of the log–log extrapolation and the complications of knots and steric hindrance in larger monomer systems, such speculations are not useful here.

Note that the FLV work, as does 1, predicts an eventual value of C_c/c at and above unity, at high enough concentrations. For the 20 and 80 KS polymers, FLV predicts the unity ratio at concentrations of 10^{-7} and 7×10^{-8} M, respectively; both treatments predict unity C_c/c for 150 KS at about 6×10^{-10} M. At about twice these values, one would expect a gelation-like phenomenon to occur in a chain-free system, due to multiple catenane formation, unpredicted by 1 or FLV, and requiring extension of theory. Such gels have been observed[13] after treatment of closed circular DNA with type II topoisomerases and suitable cofactors. In the presence of chains at equilibrium, however, these concentrations would produce very few rings and catenanes. Thus the phenomenon of catenane gelation with the general system should not occur, except possibly at extremely high values of p.

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Appendix I. Corrections to T1 and T2 Sums, for x Near Unity

Convergence of the series for the T2 functions $\phi'(x,{}^5/_2)$, $\phi'(x,{}^3/_2)$, and $\phi'(x,{}^1/_2)$ and for the $\phi(x,{}^1/_2)$ is slow for values of x near unity, as all of these diverge for $x = 1$. It is consequently expeditious to replace the latter terms in the infinite series for each of these by an integral rather than to try brute force computation of large numbers of terms. This is simple for the T1 functions, which are single summations over n from 1 to ∞, of $x^n n^{-5/2}$, $x^n n^{-3/2}$, and $x^n n^{-1/2}$. The terms from $n = 1$ to m are computed, and the value of the terms from $m + 1$ to ∞ is replaced by remainder integrals of the summed arguments. These lead to the following for $R(\phi)$, the integrals

$$R\{\phi(x,{}^1/_2)\} = \int_m^\infty x^n n^{-1/2}\, dn = (-\pi/\ln x)^{1/2} \operatorname{erfc}\{(-m \ln x)^{1/2}\} \quad (5)$$

$$R\{\phi(x,{}^3/_2)\} = \int_m^\infty x^n n^{-3/2} = 2x^m m^{-1/2} + 2(\ln x) R\{\phi(x,{}^1/_2)\} \quad (6)$$

$$R\{\phi(x,{}^5/_2)\} = \int_m^\infty x^n n^{-5/2} = {}^2/_3[x^m m^{-3/2} - (\ln x) R\{\phi(x,{}^3/_2)\}] \quad (7)$$

where $\operatorname{erfc}(t)$ is the complementary error integral

$$\int_t^\infty e^{-y^2}\, dy$$

For the T2 sums, simple integration is not reasonable. A good approximation for S_i, the inner sum, i.e. the sum of U from $j = 1$ to $k^*/2$, where $U = [k + \{q(k-j)j\}^{1/2}]^{3/2}\{(k-j)j\}^{-5/2}$, was found empirically, for high k, to approach $1.354/k$. Substituting this value into eq 19 of the main paper, we get

$$\phi'(x,5/2) \simeq \sum_{k=2}^{m} x^k(S_i) + 1.354 \int_m^\infty x^k/k \, dk$$

Whence, the remainder integral is

$$R\{\phi'(x,5/2)\} = 1.354 \, \mathrm{Ei}(-m \ln x) \quad (8)$$

where $\mathrm{Ei}(t)$ is the exponential integral

$$\int_t^\infty (e^{-y}/y) \, dy$$

Similarly

$$\phi'(x,3/2) \simeq \sum_{k=2}^{m} kx^k(S_i) + 1.354 \int_m^\infty x^k \, dk$$

Whence

$$R\{\phi'(x,3/2) = 1.354x^m/(-\ln x) \quad (9)$$

and

$$\phi'(x1/2) \simeq \sum_{k=2}^{m} k^2x^k(S_i) + 1.354 \int_m^\infty kx^k \, dk$$

Whence

$$R\{\phi'(x,1/2)\} = 1.354\{x^m(-\ln x)\}\{m + 1/(-\ln x)\} \quad (10)$$

For values of x near unity, with which the correction terms are used, $-\ln x$ is closely equal to $1 - x$, or $1/D_c$, the chain number-average molecular weight. Algorithm for computing the complementary error function is built into the PL/I compiler used for the program; the exponential integrals were found by using the IMSL Fortran Mathematical Library, as function MMDEI.

Appendix II. Comparison of Numerical Errors in the Ratio of Ring-Catenane Isomers

Examination of (1-7) and (1-9) allows computation of the number ratio of any size rings (n) to the isomeric ($i + j = n$) catenanes. This ratio is independent of x, ν, and b, i.e., invariant with respect to both the chain parameters and the degree of polymerization. The ratio is given by

$$Ca_{ij}/R_{i+j} = (C/B)[i + j + q(ij)^{1/2}]^{3/2}[(i + j)/ij]^{3/2} \quad (11)$$

For the case $i = j$, this reduces to

$$Ca_{ii}/R_{2i} = (C/B)(2 + q)^{3/2}2^{5/2}/i \quad (12)$$

and for the case where $i = j = 1$, i.e., the ratio of 1,1-catenanes to dimer rings

$$Ca_{11}/R_2 = (C/B)(2 + q)^{3/2}2^{5/2} = 2.6 \quad (13)$$

Dependence of x, ν, and b are absent from all three of these relations, all quantities therein except the size of the isomers being numerical. The degree of reaction (represented by x) cancels out while $i + j = n$, for any group of catenane isomers, compared to isomeric rings. The chain parameters ν and b cancel out for any group of rings and catenanes whatever, compared together, appearing in the same relation in both C and B.

Requirements that there be no sensible steric hindrance, and that the polymer combinations follow Gaussian statistics, however, remain. Any change from these conditions would cause different ratios for catenanes to rings. These relations hold for both the general and the chain-free systems, as they pertain only to the ring–catenane ratios.

References and Notes

(1) Jacobson, H. *Macromolecules* **1984**, *17*, 705. Hereinafter referred to as 1 or paper 1.
(2) Jacobson, H.; Stockmayer, W. H. *J. Chem. Phys.* **1950**, *18*, 1600. Hereinafter referred to as JS.
(3) Frisch, H.; Wasserman, E. *J. Am. Chem. Soc.* **1961**, *83*, 3789.
(4) Wang, J. C.; Davidson, N. *J. Mol. Biol.* **1966**, *15*, 111.
(5) Yamakawa, H.; Stockmayer, W. H. *J. Chem. Phys.* **1972**, *57*, 2843. Hereinafter referred to as YS.
(6) Hershey, A. D.; Burgi, E.; Ingraham, L. *Proc. Natl. Acad. Sci. U.S.A.* **1963**, *49*, 748.
(7) Sgaramella, V.; van der Sande, J. H.; Khorana, H. G. *Proc. Natl. Acad. Sci. U.S.A.* **1970**, *67*, 1468.
(8) Flory, P. *Chem. Rev.* **1946**, *39*, 137.
(9) Truesdell, C. A. *Ann. Math.* **1945**, *46*, 144.
(10) Frank-Kamenetskii, M. D.; Lukashin, A. V.; Vologodskii, A. V. *Nature (London)* **1975**, *258*, 398. Hereinafter referred to as FLV.
(11) Wang, J. C.; Davidson, N. *J. Mol. Biol.* **1966**, *19*, 469.
(12) Wang, J. C.; Schwartz, H. *Biopolymers* **1967**, *5*, 953.
(13) Krasnow, M. A.; Stasiak, A.; Spengler, S. J.; Dean, F.; Koller, T.; Cozzarelli, N. R. *Nature (London)* **1983**, *304*, 559.

Induced Chain Rigidity, Splay Modulus, and Other Properties of Nematic Polymer Liquid Crystals

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ABSTRACT: We present a numerical analysis of the induced chain rigidity or global persistence length, the order parameter, the splay modulus, and other properties of a polymer nematic. The macromolecules are viewed as long slender wormlike cylinders interacting via hard-core repulsions in the second virial approximation. We calculate the orientational distribution function from the nonlinear integrodifferential equation first formulated by Khokhlov and Semenov. A bifurcation analysis of this equation is also given. Exact expressions for the susceptibility and the global persistence length are derived in terms of the distribution function. Analytical estimates of these quantities based on the usual methods are extremely poor approximations to those determined numerically. We also discuss the splay modulus which is directly related to the susceptibility and the global persistence length.

I. Introduction

The average dimension of an *isolated* wormlike chain is a well-known function of the persistence length P which in turn equals the chain bending constant divided by the temperature.[1,2] It is not widely appreciated that this relation is not universally valid since it is statistical in nature.

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