Modulation of the cooperativity in the assembly of multistranded supramolecular polymers

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Supplementary information

Modelling the influence of salt concentration on polymerization.

The general equation for the formation of oligomers and polymers formed by **C** and **B** has been described elsewhere.^[31] Briefly, the expression for stepwise formation constants for complexes **CB**, **CB**₂ and **C**₂**B**, K_1 , K_2 and K_3 is

$$K_{1} = \frac{[CB]}{[C][B]}$$
(S1)

$$K_{2} = \frac{[CB_{2}]}{[CB][B]}$$
(S2)

$$K_{3} = \frac{[C_{2}B]}{[CB][C]}$$
(S3)

In the current paper, the complex CB is re-named monomer M. The oligomerization constant of M is

$$K_{o} = \frac{[M_{n}]}{[M_{n-1}][M]}$$
(S4)

Which can be written as a function of the stepwise binding constants:

$$K_o = \frac{4K_2K_3}{K_1}$$
 (S5)

The dimerization of the polymer \mathbf{M}_n leads to the double stranded polymer $(\mathbf{M}_2)_n$, which in the current paper is termed \mathbf{D}_n . The dimerization depends on the lateral association constant per unit repeat, K_l , as follows:

$$K_l^n E M^{n-1} = \frac{[D_n]}{[M_n]^2}$$
 (S6)

The total concentration of \mathbf{C} and \mathbf{B} , $[\mathbf{C}]_0$ and $[\mathbf{B}]_0$ can be written as

$$[C]_{0} = [C] + [CB_{2}] + 2[C_{2}B] + \sum_{n=1}^{\infty} n[M_{n}] + \sum_{n=1}^{\infty} 2n[D_{n}]$$

$$[B]_{0} = [B] + 2[CB_{2}] + [C_{2}B] + \sum_{n=1}^{\infty} n[M_{n}] + \sum_{n=1}^{\infty} 2n[D_{n}]$$
(S8)

In this work we are interested in relating the concentration of salt with the formation of the polymer. For simplicity, we have carried out the polymerization experiments in conditions where the total concentration of C equals that of B. With K_1 much larger than K_2 and K_3 , we can assume that the concentration of CB_2 and C_2B is unimportant in these conditions, thus:

$$[C]_{0} = [C] + \sum_{n=1}^{\infty} n[M_{n}] + \sum_{n=1}^{\infty} 2n[D_{n}]$$

$$[B]_{0} = [B] + \sum_{n=1}^{\infty} n[M_{n}] + \sum_{n=1}^{\infty} 2n[D_{n}]$$
(S9)
(S10)

Since $[C]_0 = [B]_0$, we have that [C] = [B], and equation (S9) and (S10) are identical.

The concentration **C** and **B** building blocks within \mathbf{M}_n and \mathbf{D}_n can be written as a function of the free concentration of **C** and **B** as follows:

(S12)

$$\sum_{n=1}^{\infty} n[M_n] = \frac{K_1[C][B]}{\left(1 - K_o K_1[C][B]\right)^2}$$
(S11)
$$\sum_{n=1}^{\infty} 2n[D_n] = \frac{2K_l K_1^2[C]^2[B]^2}{\left(1 - K_l EM(K_o K_1[C][B])^2\right)^2}$$
(S12)

Substituting in equation S9 we have that

$$[C]_{0} = [C] + \frac{K_{1}[C][B]}{\left(1 - K_{o}K_{1}[C][B]\right)^{2}} + \frac{2K_{l}K_{1}^{2}[C]^{2}[B]^{2}}{\left(1 - K_{l}EM(K_{o}K_{1}[C][B])^{2}\right)^{2}}$$
(S13)

and since [C] = [B] we have that

$$[C]_{0} = [C] + \frac{K_{1}[C]^{2}}{\left(1 - K_{o}K_{1}[C]^{2}\right)^{2}} + \frac{2K_{l}K_{1}^{2}[C]^{4}}{\left(1 - K_{l}EM(K_{o}K_{1}[C]^{2})^{2}\right)^{2}}$$
(S14)

We note the concentration of dimer repeats within the double stranded polymer as [D], so:

$$[D] = \sum_{n=1}^{\infty} n[D_n]$$
(S15)

Therefore, substituting in equation S12, we have that [D] can be written as

$$[D] = \frac{K_l K_1^2 [C]^4}{\left(1 - K_l E M (K_o K_1 [C]^2)^2\right)^2}$$
(S16)

And substituting in equation S14 we have that

$$[C]_{0} = [C] + \frac{K_{1}[C]^{2}}{\left(1 - K_{o}K_{1}[C]^{2}\right)^{2}} + 2[D]$$
(S17)

The increase of dimerization constant can be attributed to the binding of countercations to the double stranded polymers \mathbf{D}_n . Analysis of crystallographic data of the dimer repeating unit allows identifying up to 4 binding sites of Na⁺ (Figure 2C). Thus, Na⁺ binds to each of the repeating units \mathbf{D} to generate complexes of the form Na_j \mathbf{D} with $j \leq 4$. We can assume that the microscopic binding affinity of Na⁺ to each binding site is the same. The statistical correction factor *s* that has to be multiplied to the microscopic constant for the binding of a ligand to multivalent receptors is

$$s = \frac{n+1-i}{i} \tag{S18}$$

Where *n* is the total number of binding sites and *i* the number of binding sites occupied after the binding event. Since in our system n = 4, the overall formation constant for each of the possible complexes can be written as

$$\left(\prod_{i=1}^{i=j} \frac{5-i}{i}\right) K_{Na}{}^{j} = \frac{[Na_{j}D]}{[D][Na]^{j}}$$
(S19)

With $j \le 4$

The lateral association constant per unit repeat in the absence of Na⁺ is K_{lo} , and can be written as a function of the concentration of repeats in the double stranded polymer, [**D**], and the concentration of repeats in the corresponding single stranded polymer, [**S**] :

$$K_{l0}EM^{\frac{n-1}{n}} = \frac{[D]}{[S]^2}$$
(S20)

That for EM close to 1 and for n values larger than 10 can be written as

$$K_{l0} = \frac{[D]}{[S]^2}$$
(S21)

In the presence of Na⁺, some of the binding sites in the double stranded polymer will be occupied. On the other hand, we assume that the binding of the single stranded oligomer to the cation is much weaker and can be disregarded. This assumption is based on the fact that at least 2 and up to 4 sulphonate groups may interact simultaneously with Na⁺ in the double stranded polymer while in the single stranded polymer only one sulphonate can interact with sodium. The apparent lateral association constant per repeat units K_1 can be thus written as

$$K_{l} = \frac{[D] + \sum_{j=1}^{4} [Na_{j}D]}{[S]^{2}}$$
(S22)

Combining equations (S19) and equation (S22) we have:

$$K_{l} = \frac{[D] + 4K_{Na}[D][Na] + (3/2)K_{Na}{}^{2}[D][Na]^{2} + (2/3)K_{Na}{}^{3}[D][Na]^{3} + (1/4)K_{Na}{}^{4}[D][Na]^{4}}{[S]^{2}}$$
(S23)

combining with equation (S21) we have:

$$K_{l} = K_{l0}(1 + \sum_{i=1}^{i=4} (\frac{5-i}{i}) K_{Na}^{i} [Na]^{i})$$
(S24)

And substituting equation (S24) in equations (S16) we have that

$$[D] = \frac{K_{l0}(1 + \sum_{i=1}^{i=4} (\frac{5-i}{i})K_{Na}{}^{i}[Na]^{i})K_{1}{}^{2}[C]^{4}}{\left(1 - K_{l0}EM(1 + \sum_{i=1}^{i=4} (\frac{5-i}{i})K_{Na}{}^{i}[Na]^{i})(K_{o}K_{1}[C]^{2})^{2}\right)^{2}}$$
(S25)

To calculate K_{Na} , we need to relate the UV data to this constant. Typically, changes in the absorbance, related to changes in the relative amount of species in equilibrium, are used to determine the constant. The absorbance, however, depends on the absolute concentration of the species present. In our experimental design the changes in absorbance are overwhelmingly due to the increase in total concentration, rather than the binding events. The changes in the shape of the spectrum, on the other hand, are almost exclusively due to those binding events. It is therefore more convenient to use a measure of the shape of the spectrum, for example, the ratio of absorbance between two fixed wavelengths, to estimate the relative concentrations of species in equilibrium and, from it, evaluate the binding parameters. For convenience, we choose the ratio between the absorbance at 460 over the sum of absorbances at 430 and 460 nm, that we call x_A , that is

$$x_A = \frac{A_{460}}{A_{460} + A_{430}} \tag{S26}$$

At 460 nm, the only specie responsible for the signal is the double stranded polymer.^[31] All the species contribute to the signal at 430 nm, including the double stranded polymer, single stranded oligomer and free **C**. 430 nm is also the isosbestic point in the formation of single stranded polymers. This means that the value of absorbance at this wavelength does not change if the relative amounts of free **C** and single stranded polymer change, and we can therefore combine them in a single specie for the purpose of the analysis of the UV data. We call the total concentration of **C** building block within this combined specie [**N**], that is:

$$[N] = [C] + \frac{K_1[C]^2}{\left(1 - K_o K_1[C]^2\right)^2}$$
(S27)

Therefore, the total concentration of **C** can be written as:

$$[C]_0 = [N] + [D]$$
(S28)

 x_A can be then written as a function of [**D**] and [**N**] as follows:

$$x_{A} = \frac{2[D]\varepsilon_{D460}}{2[D]\varepsilon_{D430} + 2[D]\varepsilon_{D460} + [N]\varepsilon_{M430}}$$
(S29)

Where ε_{D430} and ε_{D460} are the molar extinction coefficients of building block **C** within the double stranded polymer at 430 and 460 nm, and ε_{N430} the extinction coefficient of the combined specie **N** at 430 nm.

We define x_D , that is the fraction **C** within the double stranded polymer over the total concentration of **C**, as:

$$x_D = \frac{2[D]}{[C]_0} \tag{S30}$$

Combining equations (S29) and (S30) we have that:

$$x_A = \frac{\varepsilon_{D460}}{\varepsilon_{D430} + \varepsilon_{D460} + \frac{1 - x_D}{x_D} \varepsilon_{M430}}$$
(S31)

It is possible therefore to determine x_D from x_A using the appropriately re-arranged version of equation (S31):

$$x_D = \frac{\varepsilon_{R1} x_A}{1 - x_A (1 + \varepsilon_{R2} - \varepsilon_{R3})}$$
(S32)

Where the values \mathcal{E}_{R1} , \mathcal{E}_{R2} and \mathcal{E}_{R3} are:

$$\varepsilon_{R1} = \frac{\varepsilon_{M430}}{\varepsilon_{D460}}$$
(S33)
$$\varepsilon_{R2} = \frac{\varepsilon_{D430}}{\varepsilon_{D460}}$$
(S34)

$$\varepsilon_{D460}$$
 (S34)
 ε_{M430}

$$\varepsilon_{R3} = \frac{1}{\varepsilon_{D460}}$$
(S35)

We used the experimental absorbance data (a representative example of which is shown in Supplementary Figure 1A and B) to determine x_D (Supplementary Chart 1). The values of extinction coefficient used for this calculation where obtained as the average of the values of spectra in the absence of oligomers (low concentration of **C**, **B** and salt), for \mathcal{E}_{N430} , and with quantitative formation of oligomer (high **C** and **B** and high salt concentration) for \mathcal{E}_{D430} and \mathcal{E}_{D460} (Supplementary Table 1).

	Concentration of Phosphate Buffer (pH 7.2) mM								
	1.95	3.91	7.81	15.6	31.3	62.5	125	250	500
	[Na ⁺] mM								
	2.93	5.86	11.72	23.4	46.9	93.8	188	375	750
[C] μM	X _A								
0.53	-0.002	0.001	0.000	-0.003	0.002	0.001	0.008	0.004	0.159
1.06	0.002	0.005	0.003	0.004	0.003	0.000	0.004	0.045	0.400
2.13	0.006	0.005	0.006	0.006	0.005	0.006	0.239	0.539	0.625
4.26	0.007	0.008	0.007	0.009	0.009	0.015	0.707	0.643	0.665
8.51	0.009	0.009	0.009	0.011	0.010	0.700	0.649	0.709	0.684
17.0	0.014	0.014	0.015	0.016	0.016	0.698	0.658	0.739	0.712
34.0	0.014	0.014	0.013	0.024	0.225	0.616	0.654	0.728	0.722
68.1	0.015	0.015	0.015	0.026	0.604	0.672	0.728	0.761	0.749
136	0.011	0.011	0.011	0.016	0.661	0.681	0.745	0.764	0.760

Supplementary Chart 1. Values of x_A (top panel) for all the samples analysed, and their corresponding values of x_D (bottom panel) calculated using the parameters in Supplementary Table 1.

	Concentration of Phosphate Buffer (pH 7.2) mM								
	1.95	3.91	7.81	15.6	31.3	62.5	125	250	500
	[Na⁺] mM								
	2.93	5.86	11.72	23.4	46.9	93.8	188	375	750
[C] μM		•			X D				
0.53	-0.005	0.001	0.000	-0.007	0.004	0.002	0.020	0.011	0.326
1.06	0.004	0.011	0.008	0.010	0.006	0.001	0.011	0.102	0.671
2.13	0.014	0.011	0.014	0.014	0.011	0.015	0.455	0.817	0.895
4.26	0.018	0.019	0.016	0.022	0.020	0.035	0.961	0.910	0.928
8.51	0.022	0.022	0.022	0.026	0.025	0.956	0.915	0.963	0.944
17.0	0.033	0.033	0.036	0.037	0.039	0.955	0.923	0.986	0.965
34.0	0.032	0.033	0.032	0.056	0.434	0.888	0.920	0.978	0.973
68.1	0.037	0.035	0.036	0.061	0.877	0.934	0.978	1.002	0.993
136	0.027	0.026	0.026	0.037	0.925	0.941	0.991	1.004	1.001

Supplementary Table 1. Extinction coefficient values for **N** and **D** at 430 and 460 nm, and their corresponding ratios. The units are $\mu M^{-1} cm^{-1}$

ε _{N430}	ε _{N460}	Е _{D430}	Е _{D460}
0.25	0	0.033	0.10



Supplementary Figure 1 A. Top panel: Spectra of solutions 1:1 of **C** and **B** in buffer phosphate 125 mM, pH 7.2. The concentration of **C** and **B** increases exponentially from the darkest trace (0.53 μ M) to lightest trace (136 μ M). Bottom panel: Spectra of solutions 1:1 of **C** and **B** at a concentration 8.51 μ M in in buffer phosphate, pH 7.2. The concentration of phosphate increases exponentially from the darkest trace (1.95 mM) to lightest trace (500 mM). B. Changes of the mole fraction of monomer within the double stranded polymer, x_D , with the concentration of Na⁺ and the total concentration of **C** (which is equal to that of **B**). The red spheres are the experimental values of x_D calculated from the UV spectra. The blue surface is the best fit to the model described by equations (S14), (S16), (S25) and (S30).

 x_D was then fitted to the model defined by equations (S14), (S16), (S25) and (S30), using the program Micromath Scientist 3.0 (Supplementary Figure 1). We used the values of K_1 , K_0 and K_{l0} previously obtained as fixed parameters (i.e., 1.16×10^6 , 8.40×10^3 and $6.1 \, \text{M}^{-1}$ respectively) with EM = 1, in the fitting. The fit to the experimental data is for the most part good, returning a value for K_{Na} of 60 M⁻¹. However, it is clear that the formation of the double stranded polymer as the salt concentration increases is more cooperative than allowed from our model (Supplementary Figure 1B). To improve the fitting, we assume that the binding of Na⁺ is subjected to strong allosteric cooperativity, in which only the states with no Na⁺ or with full binding site occupancy are populated. K_{Na} can thus be written as a function of the concentration of Na⁺ as follows:

$$K_{Na}{}^{4} = \frac{[Na_{4}D]}{[D][Na]^{4}}$$
(2)

Modifying equation (S22) accordingly we have that

$$K_l = \frac{[D] + [Na_4D]}{[S]^2}$$
 (S36)

That written as a function of the concentration of Na⁺ is:

$$K_{l} = \frac{[D] + K_{Na}^{4}[D][Na]^{4}}{[S]^{2}}$$
(S37)

And substituting in equation (S21) we have:

 $K_{l} = K_{l0}(1 + K_{Na}^{4}[Na]^{4})$ (3)

Substituting in equation (S14) we have that:

$$[C]_{0} = [C] + \frac{K_{1}[C]^{2}}{\left(1 - K_{o}K_{1}[C]^{2}\right)^{2}} + \frac{2K_{l0}(1 + K_{Na}^{4}[Na]^{4})K_{1}^{2}[C]^{4}}{\left(1 - K_{l0}(1 + K_{Na}^{4}[Na]^{4})EM(K_{o}K_{1}[C]^{2})^{2}\right)^{2}}$$
(4)

With:

$$[D] = \frac{K_{l0}(1 + K_{Na}{}^{4}[Na]^{4})K_{1}{}^{2}[C]^{4}}{\left(1 - K_{l0}(1 + K_{Na}{}^{4}[Na]^{4})EM(K_{o}K_{1}[C]^{2})^{2}\right)^{2}}$$
(5)

We fitted the x_D data to the model defined by equations (4) and (5), together with equation (6), using the program Micromath Scientist 3.0. This model provides a better fit that is consistent with the rapid increase in polymerization extent with the concentration of salt (Figure 3A).

Surface fit representations.

The program PovRay 3.6 was used to generate the surface fits shown in Figure 3A, 3B and Supplementary Figure 1B. For Figure 3A and Supplementary Figure 1B, the experimental data were represented as spheres. The data to generate the surface was obtained carrying out a simulation with the program Micromath Scientist 3.0, using the parameters obtained from the fitting. Values of the concertation of free [**C**] calculated during the simulation where substituted in equation (7) in order to obtain the corresponding values of $\langle N_2 \rangle$. These values where used to generate the surface in Figure 3B.

NMR experiments

The formation of large polymers following a nucleation-growth mechanism as the concentration of the building blocks increased has been detected by NMR, as described in our earlier work.^[31] Here, we show that the increase of the salt concentration has a similar effect on the NMR spectrum: the sharp signals assigned to discrete species decrease intensity as the salt concentration increases, consistent with a cooperative formation of large polymers that are not visible using typical solution NMR. Changes in the relative intensity of the signals assigned to the beta proton of **C** and that assigned to excess **B** is consistent with changes in the fraction of double stranded oligomer, x_D , as predicted by our model (Figure 2B).

Cooperativity plots

Multistranded polymerization model

For general applicability, the plots were generated for polymers formed by the assembly of a monomer **M**, with **M** a covalent-linked unit, unlike the **CB** monomer, which is formed by the association of **C** and **B**. The equations used are thus somewhat simplified. For the double stranded polymer the equations that relate K_o and K_l is obtained by replacing K_1 [**C**][**B**] for the concentration of free **M** in equation (S13):

$$[M]_{0} = \frac{[M]}{\left(1 - K_{o}[M]\right)^{2}} + \frac{2K_{l}[M]^{2}}{\left(1 - K_{l}EM(K_{o}[M])^{2}\right)^{2}}$$
(S38)

that for polymers with any number of strands *m* becomes:^[31]

$$[M]_{o} = \sum_{i=1}^{i=m} \frac{iK_{l}^{i-1}[M]^{i}}{(1 - K_{o}^{i}K_{l}^{i-1}EM^{i-1}[M]^{i})^{2}}$$
(S39)

With the concentration of **M** in the *m* stranded polymer, [**P**], being:

$$[P] = \frac{mK_l^{m-1}[M]^m}{(1 - K_o^m K_l^{m-1} E M^{m-1}[M]^m)^2}$$
(S40)

The fraction of *m*-stranded polymer, x_P , is:

$$x_P = \frac{[P]}{[M]_0} \tag{S41}$$

The maximum concentration, $[\mathbf{M}]_{max}$, for a *m*-stranded polymer is the concentration at which the concentration of polymer tends to the infinity, that is, the denominator in equation (S40) tends to zero:

$$(1 - K_o^m K_l^{m-1} E M^{m-1} [M]_{max}^m)^2 = 0$$
(S42)

Re-arranging equation (S42) we have that

$$[M]_{max} = \frac{\left(K_{l}EM\right)^{\frac{1-m}{m}}}{K_{o}}$$
(S43)

The concentration as multiples of $[\mathbf{M}]_{max}$, $n[\mathbf{M}]_{max}$, was then calculated by dividing the total monomer concentration by the maximum monomer concentration:

$$n[M]_{max} = \frac{[M]_0}{[M]_{max}}$$
(S44)

The growth of **P** with the total concentration of **M**, Δ [**P**]/ Δ [**M**]₀, was calculated graphically from the data of [**M**]₀ and [**P**] generated in the corresponding simulations. The corresponding plots are shown in Figure 5. The corresponding traditional cooperativity plots, that show the mole fraction of polymer as a function of the total concentration of monomer, are shown in Supplementary Figure 3.



Supplementary Figure 3. A. Changes in the mole fraction of polymer over total monomer (x_p) with the total concentration of monomer, expressed as multiples of the maximum monomer concentration, $n[\mathbf{M}]_{max}$, for double stranded polymers with different K_i values. The values of K_i are 50 (blue trace), 500 (red trace) and 50000 (green trace) M⁻¹, and infinity (dotted trace). The value of K_o is 8400 M⁻¹ in all cases. B. Changes in the fraction of polymer over total monomer (x_p) with the total concentration of monomer, expressed as multiples of the maximum monomer concentration, $n[\mathbf{M}]_{max}$, for polymers with different number of strands m. The number of strands m are 1 (dotted line), 2 (green trace), 3 (red trace) and 4 (blue trace). The green, red and blue traces are overlapping. K_o is 8400 M⁻¹ and K_i is 50000 M⁻¹ in all cases.

General polymerization model

In the polymerization model commonly used for nucleation growth, two binding constants are defined, the nucleation constant K_n and the growth constant K_g , with the cooperativity factor α defined as the ratio between the constants, i.e.



Supplementary Figure 4. Schematic representation of the nucleation-growth assembly of a hexastranded polymer that follows an helical assembly path, as an example of a polymer whose polymerization requires the assembly of a nucleus containing 6 building blocks. The total concentration of monomer, $[\mathbf{M}]_0$, can be written as the sum of species smaller than the nucleus (which we call $[\mathbf{M}]_i$, with i < m), and polymers equal or larger than the nucleus (i.e., polymers of the form $[\mathbf{M}]_{m+n}$)(Supplementary Figure 4).

$$[M]_{0} = \sum_{i=1}^{i=m-1} i[M]_{i} + \sum_{n=0}^{n=\infty} (n+m)[M]_{n+m}$$
(S45)

The concentration of polymer with m+n building blocks can be written as:

$$[M]_{m+n} = K_n^{m-1} K_g^{n} [M]^{m+n}$$
(S46)

And the total concentration of monomer within the polymer is

$$\sum_{n=1}^{n=\infty} (n+m)[M]_{n+m} = \sum_{n=0}^{n=\infty} (n+m)K_n^{m-1}K_g^{n}[M]^{m+n}$$
(S47)

On the other hand the concentration of small assemblies can be written as

$$[M]_{i} = K_{n}^{i-1}[M]^{i}$$
(S48)

With *i* < m

Equation (S47) can be re-arranged as:

$$\sum_{n=1}^{n=\infty} (n+m)[M]_{n+m} = mK_n^{m-1} \sum_{n=0}^{n=\infty} K_g^{n} [M]^{m+n} + K_n^{m-1} \sum_{n=0}^{n=\infty} nK_g^{n} [M]^{m+n}$$
(S49)

and furthermore as

$$\sum_{n=1}^{n=\infty} (n+m)[M]_{n+m} = mK_n^{m-1}[M]^m \sum_{n=1}^{n=\infty} K_g^{n-1}[M]^{n-1} + K_n^{m-1}[M]^m \sum_{n=1}^{n=\infty} nK_g^{n-1}[M]^{n-1}$$

(S50)

Taylor formulae for the relevant convergent series state that, for *x* < 1:

$$\sum_{n=1}^{\infty} nx^{n-1} = \frac{1}{(1-x)^2}$$
(S51)
$$\sum_{n=1}^{\infty} x^{n-1} = \frac{1}{1-x}$$
(S52)

Therefore, we have that

$$\sum_{n=1}^{n=\infty} (n+m)[M]_{n+m} = \frac{mK_n^{m-1}[M]^m}{1-K_g[M]} + \frac{K_n^{m-1}[M]^m}{\left(1-K_g[M]\right)^2}$$
(S53)

Substituting equation (S48) and (S53) on equation (S45) results in:

$$[M]_{0} = \sum_{i=1}^{i=m-1} iK_{n}^{i-1}[M]^{i} + \frac{mK_{n}^{m-1}[M]^{m}}{1-K_{g}[M]} + \frac{K_{n}^{m-1}[M]^{m}}{\left(1-K_{g}[M]\right)^{2}}$$
(S54)

We have defined [**P**] as the concentration of monomer within the polymers. Substituting in equation (S53) we have that for this model, [**P**] can be written as a function of the free monomer as:

$$[P] = \frac{mK_n^{m-1}[M]^m}{1 - K_g[M]} + \frac{K_n^{m-1}[M]^m}{(1 - K_g[M])^2}$$
(S55)

In this model, $[\mathbf{M}]_{max}$ is the concentration at which the concentration at which the denominator in the last member of equation (S54) tends to zero. Therefore, we have that

$$[M]_{max} = \frac{1}{K_g} \tag{S56}$$

The concentration as multiples of $[\mathbf{M}]_{max}$, $n[\mathbf{M}]_{max}$, was then calculated by dividing the total monomer concentration by the maximum monomer concentration:

$$n[M]_{max} = \frac{[M]_0}{[M]_{max}} \tag{S44}$$

The growth of **P** with the total concentration of **M**, Δ [**P**]/ Δ [**M**]₀, was calculated graphically from the data of [**M**]₀ and [**P**] generated in the corresponding simulations. The corresponding plots are shown in Figure 6, for *m* = 2 and *m* = 6.