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Short Spiral Macromolecules in an External Electromagnetic Field

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Abstract

The analysis of the results of irradiation of DNA isolated from lymphocytes by an ultrahigh frequency electromagnetic field was carried out. The natural frequency of torsional vibrations of the spirals of collagen molecules was found. It is proved theoretically that single-strand breaks in the DNA of lymphocytes by microwaves are caused by torsional vibrations of the helices of DNA molecules whose natural frequencies coincide with the frequencies of microwaves, and the resonant frequencies have been found. For research, instead of millimeter waves, multiple frequencies of centimeter waves were used. The theoretical analysis has been confirmed experimentally: a multiple frequency in the centimeter range causes a maximum increase in single-strand DNA breaks of arthropod hemolymph amoebocytes, while close frequencies do not lead to an increase in the number of single-strand breaks.

Subject Areas

Biophysics, Microbiology

Keywords

Resonance, Multiplicity, Replication, Breaks, Protein, Power, Lymphocytes

1. Introduction

In [1], it was proved that *in vitro* DNA molecules of bacteria *E. coli* ATCC 25922, *Mycobacterium avium* 104 and *Mycobacterium tuberculosis* H37RV (Pasteur) ATCC 25618 are capable of absorbing centimeter electromagnetic field waves (EMF). This happens when the frequency of the microwave EMF is resonant to the natural frequency of torsional vibrations of the free helix of the DNA molecule. In the elastic rod model, using Lagrange formalism and experimental data, general formula for these oscillations was obtained: $f \sim sqrt(G/J)$, *G*—stiffness, *J*—moment of inertia, $J = s\left(m_1 + m_2 + m_3 + \cdots + m_N\right)R^2$, *R* is the radius of the spiral coil, *m* is the mass of the coil, and *s* is the number of coils [2]. The stiffness is determined from the experiment. As a result, the formula is obtained:

$$f = 21.75/(bp)^{-1/2} \quad (1)$$

where *bp*—the number of nucleotide pairs, frequency—in THz [2]. The numerical coefficient integrally takes into account the heterogeneity of the DNA chain, its compactification and its environment.

As a result of excitation by an external field of torsional vibrations in DNA molecules, the number of single-strand breaks in DNA increases dramatically, and the bacterium dies.

The phenomenon of excitation of torsional vibrations in DNA is essentially classical, not quantum in nature.

In experiments on exposure to resonant frequencies, a sharp decrease in the survival rate of *E. coli* up to 20%, the complete destruction of *M. avium* and a thousandfold decrease in the survival rate of *Mycobacterium tuberculosis* were obtained [1].

2. Collagen and DNA of Lymphocytes

Spiral molecules of various types of collagen are a left-twisted spiral of three *α*-chains with a length of 270 - 300 nm, with a molecular weight of 270 - 300 kDa, and fibrils formed by them using intermolecular crosslinking with a length of up to 3200 nm. One turn of the a-chain helix contains three amino acid residues. Assume that the mass of one amino acid residue is approximately 112 Da. Then there are a maximum of 297 turns in one collagen molecule. In one turn of the DNA molecule—10 bp, one can express the formula (1) in terms of the number of turns *n* of spiral of DNA:

$$f = q/\sqrt{10n}$$

This formula is common to all spiral molecules. Using (2), we obtain the frequency of torsional vibrations of the collagen molecule—approximately 452 GHz.

It was shown in [3] that the effect of centimeter EMF on the DNA of lymphocytes leads to single-strand breaks (SSB) of the DNA helix. The effect of centimeter EMF on DNA in DNA isolated from lymphocyte cells was studied. Microwave generators were used: GKCH 53, R2-68, R2-69 with a capacity of 3 mW. The samples were exposed to radiation for 10 minutes at three different frequencies. With an increase in the frequency of microwave radiation, the amount of SSB of DNA in lymphocytes increases compared to control samples: when exposed to radiation with a frequency of 3.5 GHz by (32.3 ± 0.9)% (the first point of the curve), with a frequency of 50 GHz by (40.1 ± 1.1)% (the second point of the curve), with a frequency of 70 GHz by (49.8 ± 0.7)% (the third point of the curve). Note that the frequencies of 50 and 70 GHz cannot excite longitudinal oscillations in the DNA molecules of lymphocytes. The oscillation frequency of both DNA chains (simultaneous compression-stretching of both chains) is proportional to $(2k/M)^{1/2}$, where k is the stiffness coefficient, M is the mass of DNA. Estimates show that the frequencies of longitudinal oscillations are two orders of magnitude lower. It is not difficult to obtain an approximate stiffness coefficient N/m and an approximate resonant frequency for a model with 35 pairs of nucleotides—4 GHz. It is possible to calculate the stiffness for DNA of any length by the formula $k_i = kl/l_i$. Denote the number of nucleotide pairs in the model N_0, stiffness—k_0. General formula

$$f_i = \left(k_0 N_0 / 2m_0\right)^{1/2} \Big/ \pi N_i$$

where m_0 is the average mass of a DNA base pair, N_i is the number of base pairs for the i-th type of DNA.

If for *E. coli* with a chain length of 5 million base pairs, the resonant frequency according to (1) will be about 104 Hz, then for lymphocyte DNA it corresponds to the megahertz range.

The dependence of the number of SSB on the frequency of the three points in [3] above is approximately laid on the curve

$$y = 0.003x^2 + 0.5x + 0.15$$

Thus, 100% is achieved at a frequency of approximately 160 - 200 GHz.

The molecular weight of lymphocyte DNA is 3.5 (10^5 - 10^6) Da [4]. Then the frequency of torsional oscillations of the DNA helix of a lymphocyte with a molecular weight of 3.5×10^6 Da, calculated by the formula (1), is about 300 GHz.

For DNA with a molecular weight of 3.5×10^5 and, accordingly, the number of base pairs 507, the frequency of longitudinal oscillations is about 1 GHz, the frequency of torsional oscillations is about 1 THz, Thus, even taking into account the error in determining the molecular weight of DNA lymphocytes, the frequency of 200 GHz calculated according to the model curve from the data [3] does not quite satisfactorily correspond to the direct calculation of the frequency of torsional oscillations of DNA spirals 300 GHz according to (1).

Indeed, the considered curve cannot be a parabola, from almost zero point to the first point it grows rapidly, then it grows extremely slowly, at the second point the growth accelerates noticeably, finally the growth of the number of SSB according to (1) grows indefinitely with frequency. In addition, it is obvious that at zero frequency, the number of OP added to the usual level is zero, *i.e.* the coefficient c in the model equation must also be zero. Mathematical modeling shows a significant difference with a parabola. This means that the data pair of the first point [3] is incorrect. The frequency of 3.5 GHz cannot correspond to any kind of natural oscillations of the DNA of lymphocytes. In fact, these 3 points belong not to one, but to 3 different curves.

For data analysis [3], it is important that the absorption spectrum of centimeter waves by DNA molecules, since the length of the DNA helix is much larger than its diameter, has the form of an inverted parabola, the maximum of which is the peak of absorption. Therefore, it would be correct to approximate the growth curve of the number of SSB from the frequency by an inverted parabola of the form

$$y = 100 - a(x-b)^2$$

It means that at resonance, the number of SSB reaches 100%. Then, using the third point (70 GHz), obtained by the formula (4) that the resonant frequency of the DNA helix lymphocyte (*b*) is approximately equal to 310 GHz, which is close to the calculation made according to the formula of torsional vibrations (1).

For the first point the resonant frequency of 6.4 GHz, this frequency is no longitudinal or torsional vibrations from such a small DNA cannot be that again shows that the first point should be recognized correctly.

For the second point, 50 GHz, the resonant frequency is different, *b* = 188.5 GHz, since the DNA of different lymphocytes has different molecular weights and, accordingly, lengths. Approximately 13,320 base pairs correspond to this natural frequency.

Thus, it is proved that the SSB in DNA isolated from lymphocytes arise as a result of the excitation by microwaves in DNA molecules of exclusively torsional vibrations of the helices of molecules. And vice versa: it has been established that microwaves with a frequency equal to the natural frequency of torsional vibrations of any DNA helices cause SSB in them. That is, the data [3] confirm the results [1] [2].

3. Single RNAs

In most single RNA, there are short complementary sequences that pair and form loops, so short intramolecular double-stranded sections are constantly formed in single chains. Base pairing leads to the formation of nucleotide sections in the form of loops, spiral sections form U-shaped fragments in which antiparallel sequences of nucleotides connected by hydrogen bonds are connected by at least three nucleotides forming a bend. Due to the interaction of RNA nucleotides and their combination into pairs according to the principle of complementarity, in addition to loops, hairpins, hairpins with an inner loop, duplexes with two protrusions, pseudo-nodes (two hairpins combined in a special way), and molotoid ribozymes appear in the molecule.

Thus, a single RNA molecule has the form of a randomly folded chain. Local conformations of nucleotides are close to the optimal canonical for 85% of nucleotides, 15% are in one of the more intense canonical conformations, ensuring consistency of local and long-chain interactions [5] [6].

Since the length of the spiralized sections in single-stranded RNA is small, and the spirals themselves are imperfect, they are quite easily destroyed.

The moment of inertia of the double helix of DNA *J* is twice as large as that of the single helix of RNA, half as much as BP. Accordingly, the natural frequency for RNA is $2^{1/2}$ times greater than the frequency for DNA. It may seem that the rigidity of a single RNA helix is half as much, so the formula for RNA should not differ from the formula for DNA. However, in DNA, two linked helices rotate relative to each other. The probability that the deviation of the spiral rotation angle from the equilibrium one for these sections separated by a distance L will be $\Delta\varphi$, is equal $P(\Delta\varphi) = Z\exp\left[-q(\Delta\varphi)^2/2LkT\right]$, Z is the normalization factor, k is the Boltzmann constant, q is the constant that determines the magnitude of the axial twist, T is the absolute temperature, L is the distance between adjacent base pairs. The amplitude of fluctuations in the angle of spiral rotation in a double helix: $\sqrt{\langle\Delta\varphi^2\rangle} = \sqrt{LkT/q}$. The torsion stiffness constant q of a double helix for the rotation of one DNA chain relative to another corresponds to the doubled work that must be done on a rod of unit length by turning one end relative to the other around the axis of the rod by one radian. Estimates of the magnitude of q lie in the interval (1.5 - 3) × 10^{-19} erg·cm, the most reliable estimates based on the use of the properties of short ring DNA give a value of q equal to 3 × 10^{-19} erg·cm, it corresponds to the root-mean-square amplitude of fluctuations in the angle of spiral rotation of 4 degrees. *i.e.*, two DNA helices react to microwave EMF separately, the rigidity in (1) during the transition from DNA to RNA relative to torsional vibrations can be considered unchanged, therefore, the multiplier $\sqrt{2}$ in the formula for RNA is preserved.

The nitrogenous base in RNA, complementary to adenine, is not thymine with a molar mass of 126.11334 g/mol, as in DNA, but uracil with a molar mass of 112.08676 g/mol (an unmethylated form of thymine). That is, the mass and, accordingly, the moment of inertia of the spiral coil is less. As a result, we have:

$$f_{\text{SINGLE-RNA}} = 31.196N^{-1/2}$$

where N is the number of bases (not pairs). For double helix RNA

$$f_{\text{DOUBLE-RNA}} = 22.0589N^{-1/2}$$

where N is the number of base pairs. The natural frequencies of torsional vibrations of bacterial DNA lie in the centimeter range. Since the length of the RNA chain is significantly smaller, the natural frequencies of torsional vibrations of RNA chains lie in the millimeter range.

4. Multiple Frequencies

Due to the fact that millimeter waves are shielded by a solution containing DNA or RNA, it is difficult to use EMF in the millimeter range. Consider the forced torsional vibrations of a spring:

$$\ddot{\varphi} + 2\beta\dot{\varphi} + \omega_0^2\varphi = f\cos\omega t$$

where φ is the angle of twisting of the spring, ω_0 is the natural frequency, ω is the frequency of the driving force, f is the amplitude of the driving force, β is the coefficient of friction.

The general solution is sought in the form $\varphi_1 = a\cos\omega_0 t$, the particular one in the form $\varphi_2 = b\cos\omega t$. Substituting into the equation, in the absence of friction, we get:

$$\varphi = a\cos\omega_0 t + \left[f/\left(\omega_0^2 - \omega^2\right)\right]\cos\omega t$$

That is, the system makes an oscillation, which is a combination of two oscillations. In the presence of friction, the first term decreases exponentially with time. When the frequency of the driving force coincides with the natural frequency of the oscillator, the solution of the equation has the form:

$$\varphi = a\cos\omega t + (f/2\omega)t\sin\omega t$$

That is, with resonance, the oscillation amplitude increases linearly with time. In the presence of friction, the oscillator oscillates not with its own frequency, but with frequency $\omega = \sqrt{\omega_0^2 - \beta^2}$. This system can be considered as a system with a driving force $f = -2\beta\dot{x}$. If the friction is high, the oscillation frequency can be a multiple of the natural frequency at certain discrete coefficients of friction:

$$\beta = \sqrt{3}\omega_0/2; \sqrt{8}\omega_0/3; \sqrt{17}\omega_0/4; \cdots; \omega = \omega_0/2; \omega_0/3; \omega_0/4; \cdots$$

The usual resonance occurs at all frequencies of the driving force, which satisfy the relation, where *m*, *n* are integers. Thus, resonance occurs at subharmonics, at frequencies of the driving force that are multiples of the natural frequency, for example, $\omega = \omega_0/2, \omega = \omega_0/3, \omega = \omega_0/4, \cdots$ etc. With increasing multiplicity, the resonance intensity decreases rapidly, the greater the multiplicity, the slower the amplitude increase [7]. Therefore, the exposure time should be increased.

Subharmonic resonance is a special case of parametric resonance. If the oscillations are described by the Hill-Mathieu equation, the resonance occurs at a frequency *n* times less than twice the natural frequency:

$$\ddot{x} + \omega_0^2(1 + a\cos\omega t)x = 0; \omega_{рез} = 2\omega_0/n$$

see [8]. The "driving force" has the form $f = -a\omega_0^2[\cos\omega t]x$. Then, for example, for the 1st subharmonics:

$$f \sim [\cos\omega t]\cos\omega_0 t \sim \cos(\omega_0/2) + \cos(3\omega_0/2)$$

That is, multiple frequencies are special points of the equation. Thus, under the action of a driving force with a frequency n times less than its own, the amplitude of the oscillations increases to a maximum exceeding the amplitude of the free oscillator oscillations.

Subharmonic resonances for an inverted pendulum with excitation frequencies n times less than its own are also considered in [9]. General view of subharmonic vibrations at resonance:

$$x = a\cos(\omega_0 t/n) + g(n)(fn/2\omega_0)t\sin(\omega_0 t/n)$$

where *g*(*n*) decreases with the growth of *n* faster than *n* grows.

Thus, to influence short macromolecules, centimeter waves can be used, the frequencies of which are multiples of the natural frequencies of the millimeter range.

5. Results

To confirm the theoretically obtained results, the DNA of amoebocytes of the hemolymph of *Nauphoeta cinerea* and *Dermacentor silvarum* with a weight 3.7 × 10^6 Da (length is 5330 bp) was selected as the object of the study. The corresponding frequency of torsional vibrations of the DNA data helix is 298 GHz.

The method of liquid electrophoresis was used to determine the weight of DNA. An urea solution was used to isolate DNA from amoebocytes. A solution of ethidium bromide was used for DNA labeling. The method of fluorescence spectrophotometry was used to calculate the number of single-strand breaks. Since the quantum yield of fluorescence Φ is proportional to the number of intact DNA, the growth of F is inversely proportional to the number of single-strand breaks $\Phi \sim 1/N$, thus, the increase in the number of SSB in irradiated DNA:

$$\text{Gr} = \left[\left(N_{\text{irrad}} - N_{\text{control}}\right)/N_{\text{control}}\right] \times 100\%$$

An Agilent Technologies E82570 1 microwave generator with a non-thermal power flux density of 2.5 mW/cm² was used, the exposure time was increased to 24 hours, and the temperature was 23 degrees Celsius.

Frequencies 7 GHz, 7.5 GHz, 8.5 Ghz, 9 GHz, 9.5 GHz, 10 GHz and 10.5 GHz, irradiation of amoebocyte DNA did not cause an increase in the number of SSB. When irradiated frequency was 8 GHz, that is much closer to a multiple frequency of 8054 GHz, an increase in the number of SSB was observed by 37.2%, and at 8100 GHz an increase in the number of SSB was observed by 34.8%. The frequency of 298 GHz with a high degree of accuracy is a multiple of the frequency of 8.054 GHz. When irradiated with EMF of this frequency at the same power flux density, an increase in the number of SSB was observed by 98.7%.

The method [2] was used for calculations.

A slight deviation of the generator frequency from the calculated one turns out to be significant, because it is multiplied by a multiplicity of 37, if a deviation of 0.5% is significant for DNA with a resonant frequency of about 10 GHz, then in this case the deviation is (0.054 × 37) GHz, *i.e.* about 0.7% of 298 GHz.

Thus, the peak on the resonance curve is very sharp, see **Figure 1**.

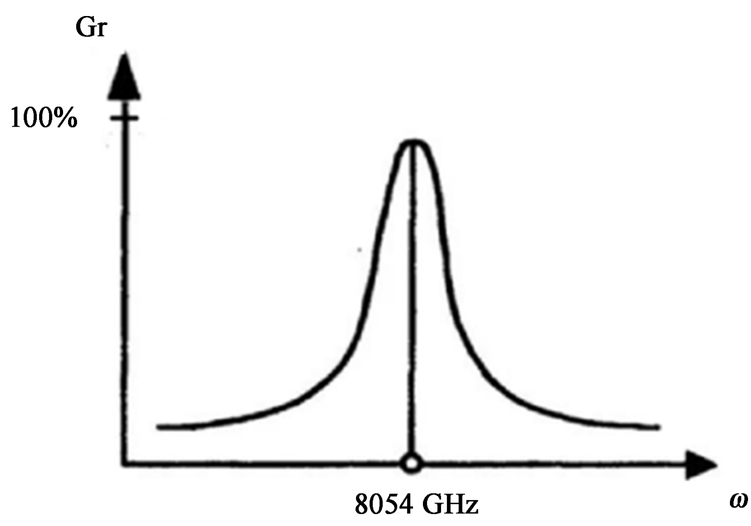

Figure 1. The dependence of the voltage at the output of the generator on time.

Positive COVID-19 samples were used for the case of single RNAs. In 14 experiments, it was shown that microwaves at estimated frequency of 10.022 GHz completely destruct COVID-19 RNA molecules. The control and experimental positive samples were selected, the exposure time for the experimental sample was 24 hours. The RT-LAMP test was used for verification.

6. Discussion

A slight deviation of the generator frequency from the calculated one turns out to be significant, because it is multiplied by a multiplicity of 37, if a deviation of 0.5% is significant for DNA with a resonant frequency of about 10 GHz, then in this case the deviation is (0.054 × 37) GHz, *i.e.* about 0.7% of 298 GHz.

In the case of DNA or RNA virus replication, the parameters of their spirals change over time, respectively, the natural frequency of their torsional oscillations changes. In this case, the frequency modulation of the generator signal can be used to destroy the molecules. The equation for the angular deviation of the turns of the spiral x is written as follows:

$$\ddot{x} + \omega_0^2 x = \omega_1^2 x_0 \sin \omega_1 t + \omega_2^2 x_1 \sin \omega_2 t$$

If $x_0 = x_1 = 1$ the equation is written more simply, a time offset occurs. For clarity, let the natural frequency be 10 GHz, the main frequency of the generator is 10.1 GHz, and the modulation is 150 MHz. Then

$$\ddot{x} + 100x = 102 \sin\left[2\pi\left(10.1t + 3t^2\right)\right]$$

As a result, the oscillation frequency is shifted from the driving frequency to its own:

$$\begin{aligned} x = {} & c_1 \cos 10t + c_2 \sin 10t - c_3 \left[0.205581(3.46322t + 4.90807)\sin 10t \right. \\ & -1.02054(3.46322t + 6.7463)\sin 10t - 1.4582(3.46332t + 4.90807)\cos 10t \\ & \left. -1.06165(3.46322t + 6.7463)\cos 10t\right] + c_4 \left[1.4582(3.46322t + 4.90807)\sin 10t \right. \\ & -1.06165(3.46322t + 6.7463)\sin 10t - 0.205581(3.46322t + 4.90807)\cos 10t \\ & \left. +1.02054(3.46322t + 6.7463)\cos 10t\right] \end{aligned}$$

Thus, with a frequency-modulated signal, the fundamental frequency of which is slightly different from its own, a resonance occurs with an increase in the amplitude of the oscillations, limited by energy losses.

However, a much more effective way to increase the number of single-strand breaks of short spiral macromolecules is forced oscillations under periodic pulsed action. Mathematical modeling using Matlab with the ratio of the frequencies of the driving force and its own 1/10, shows that already at the 4th pulse the amplitude of the oscillations doubles, what can be seen on the virtual oscilloscope, see **Figure 2**.

Resonance occurs at all frequencies of the driving force that satisfy the ratio $mf - nf_0 = 0$, where m, n are integers In our case, it is necessary that the frequency ratio be a half-integer: $f_0 = 3nf/2$, where $n = 1, 2, 3, \cdots$.

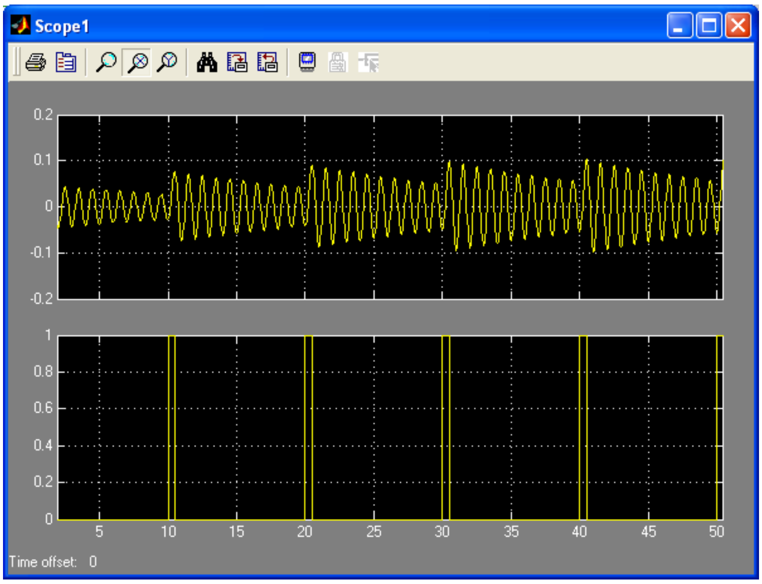

Figure 2. Dependence of the oscillation amplitude of the virtual oscillator on time with a pulsed driving force.

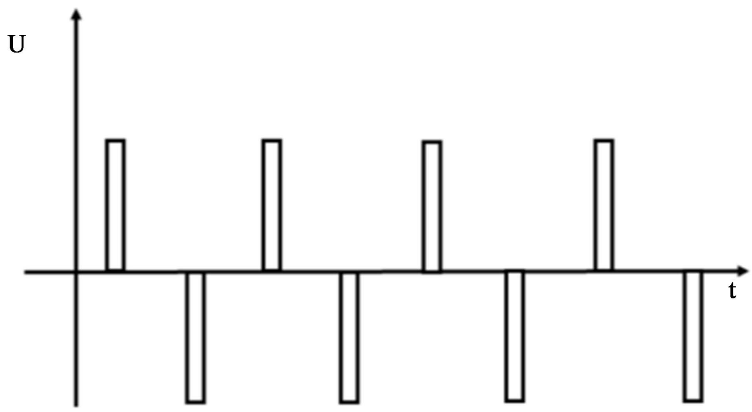

Figure 3. The dependence of the voltage at the output of the generator on time.

The most effective signal form of the generator is an alternating meander with a high borehole of the form, see Figure 3, it is new type of meandr.

7. Results

In 6 experiments with control with positive COVID-19 samples, it was shown that the use of pulse technology can reduce the irradiation time to 1 hour. The radiation source was an Agilent Technologies E82570 1 microwave generator, an Agilent Technologies E82570 power amplifier was used to amplify the signal to 1 W. The density of the microwave power flux was approximately 0.9 mW/cm^2, borehole $S = 8$. The RT-LAMP test was used for verification.

8. Conclusions

Since the effect of microwave EMF on the DNA of cells leads to a sharp increase in the number of single-strand DNA breaks, cell death under the influence of resonant microwave EMF occurs both due to the stopping of the DNA replication process and due to single-strand breaks.

The data obtained can be used in various fields, including agriculture and medicine [10] [11].

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Conflicts of Interest

The author declares no conflicts of interest.

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