

LOG-NORMAL DISTRIBUTION OF ELECTROSPUN NANOFIBRE DIAMETERS: DERIVATION FROM MULTIPLICATIVE JET SPLITTING AND EXPERIMENTAL VERIFICATION AS A PROCESS-STABILITY CRITERION

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Abstract:

The distribution law of electrospun nanofibre diameters is usually selected empirically, by fitting whichever function best matches the histogram, which prevents the result from being transferred between systems. This work derives the distribution from the physics of the process and then verifies it experimentally. Successive splitting of the electrospun jet is represented as a multiplicative random process, so that the logarithm of the fibre diameter is a sum of independent random contributions; by the central limit theorem the diameter is then log-normally distributed, in agreement with Kolmogorov's theory of stationary fragmentation. Two materials were examined by scanning electron microscopy at 1000× and 15000×: a composite web containing silk fibroin and a reference web spun from a 16% poly(vinyl alcohol) solution. The mean fibre diameters are 306.24 nm and 249.89 nm respectively, both within the range reported for regenerated silk fibroin. Pearson's goodness-of-fit test at a confidence level of 0.99 does not reject the log-normal hypothesis for either material ($\chi^2 = 4.31 < 20.1$ with $p = 0.82$, and $\chi^2 = 8.32 < 13.3$ with $p = 0.081$). Because conformity to the log-normal law follows from the stationarity of the splitting process, its experimental confirmation constitutes indirect evidence that the process is stable; conversely, departure from the law indicates the onset of instability. Use of the distribution test as an equipment-free criterion for statistical process control is therefore proposed, together with a discussion of the sampling and projection biases that affect diameter measurement from electron micrographs.



Keywords: Nanofibre; fibre diameter; log-normal distribution; Kolmogorov theory; Pearson criterion; scanning electron microscopy; process stability; statistical process control; electrospinning.

Introduction

Biomedical materials must satisfy strict requirements on composition, biodegradation time and biodegradation behaviour, and these requirements depend directly on the morphology and structure of the nanofibres that constitute the material [1, 2]. Because the nanofibre diameter is a random variable, knowledge of its distribution law is necessary for the correct statistical treatment of experimental data, for the calculation of means and confidence intervals, and for the analysis of the electrospinning process itself.

Numerous attempts have been made to describe the structure of nanofibres obtained from various polymers mathematically, but no consensus on the diameter distribution has yet been reached. Some authors state that the diameter distribution of electrospun nanofibres can often be described by a logarithmically normal law. Proponents of an alternative view hold that the diameter values obey a normal law, explaining this by the simultaneous formation of all the nanofibres in the material; they generally acknowledge that the shape of the experimental histograms is far from normal, and therefore propose combining several distributions so that their sum approaches the observed shape.

In most existing work the distribution law is selected purely empirically: the function that best fits the histogram is sought. Because such an approach is not connected to the physics of the process, the result cannot be transferred to other conditions, and no prediction follows from it. The novelty of the present work is that the log-normal distribution of nanofibre diameters is derived from the splitting mechanism of the electrospun jet rather than assumed, is verified statistically in two different polymer systems, and is then proposed as a criterion for monitoring the stability of the process.

The aim of the work is to justify theoretically and to verify experimentally the distribution law of the diameters of nanofibres obtained by electrospinning from a composite solution containing silk fibroin and from a reference 16% poly(vinyl alcohol) solution.

2. Theoretical basis

The diameters of electrospun fibres depend on the course of two principal processes: splitting of the jet and its stretching. If it is assumed that the stretching of the jets proceeds in the same manner at all stages of nanofibre formation, then the principal influence on the distribution law of the diameters is exerted by the splitting process.

Successive splitting of the jet is represented as a multiplicative random process. If the initial jet diameter is d_0 and the jet has undergone k splitting events, the diameter of the resulting fibre is

$$d_k = d_0 \cdot \xi_1 \cdot \xi_2 \cdot \dots \cdot \xi_k, \quad (1)$$


where ξ_i is the splitting ratio at the i -th stage, these being mutually independent random variables. Taking logarithms of Eq. (1) gives

$$\ln d_k = \ln d_0 + \sum \ln \xi_i. \quad (2)$$

By the central limit theorem, for sufficiently large k the sum of independent random variables tends to a normal distribution. Hence $\ln d$ is normally distributed and d itself follows a logarithmically normal (log-normal) distribution:

$$f(d) = 1 / (d \cdot \sigma \cdot \sqrt{2\pi}) \cdot \exp[-(\ln d - \mu)^2 / (2\sigma^2)], \quad d > 0, \quad (3)$$

where μ and σ are the expectation and standard deviation of $\ln d$. For the log-normal distribution the arithmetic mean of the diameter is related to these parameters by

$$E[d] = \exp(\mu + \sigma^2/2), \quad (4)$$

and is always greater than the median, $\exp \mu$; this property accounts for the right-skewed asymmetric shape of the observed histograms.

This conclusion is an application to electrospinning of the result proved by Kolmogorov [3] and Filippov [4] for stationary processes of particle fragmentation. In electrospinning the splitting of the spinning-solution jet occurs not through the interaction of individual jets with one another but as a result of the action of the electric field upon them. On that basis, provided that the splitting rate is constant along the whole path from emitter to collector, the process may be regarded as stationary. The converse also holds: if the diameter distribution obtained experimentally conforms to the log-normal law, this indirectly confirms the stationarity of the splitting process and hence the stability of the electrospinning process as a whole.

3. Materials and methods

Two materials were examined. The first was a nanofibrous web obtained from a composite spinning solution consisting of 40% silk fibroin solution (0.2 g of fibroin per 5.5 g of solvent) and 60% of a 16% poly(vinyl alcohol) solution. The second was a web obtained from the 16% solution of poly(vinyl alcohol), Arkofil PPL grade, alone. Because electrospinning from poly(vinyl alcohol) solutions is distinguished by its stability, the second material was adopted as a reference.

The structure of the nanofibres was visualised and their diameters measured using a LEO 1420 scanning electron microscope (Carl Zeiss, Germany) at magnifications of 1000 $\times$ and 15000 $\times$. Fibre diameters were determined from the resulting micrographs using image-analysis software. The structure of the fibroin-containing material is shown in Fig. 1 and that of the reference material in Fig. 2.



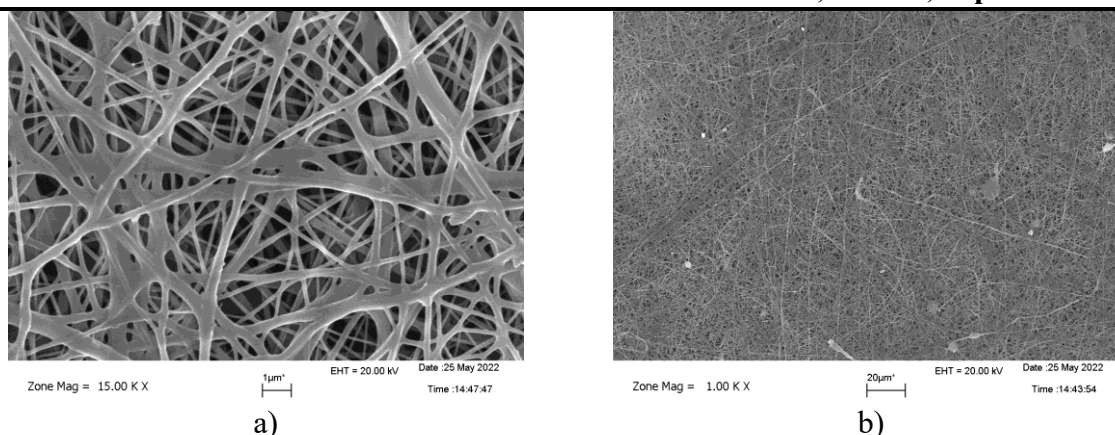


Fig. 1. Structure of the nanofibrous material containing silk fibroin at a) 15000× and b) 1000× magnification.

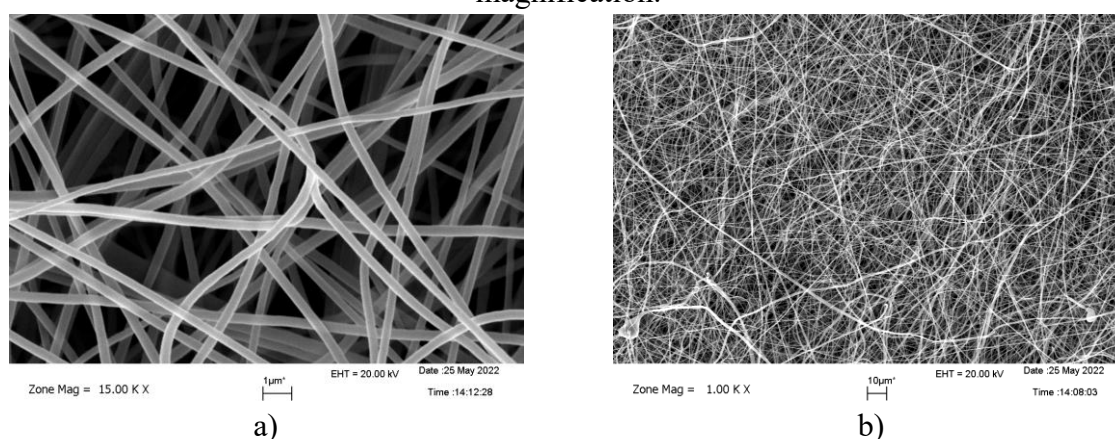


Fig. 2. Structure of the nanofibrous material obtained from the 16% poly(vinyl alcohol) solution at a) 15000× and b) 1000× magnification.

Conformity of the empirical distribution to the theoretical log-normal law was tested by Pearson's goodness-of-fit criterion:

$$\chi^2 = \sum (m_i - n \cdot p_i)^2 / (n \cdot p_i), \quad (5)$$

where m_i is the number of measurements falling in the i -th interval, p_i the probability of falling in that interval according to the theoretical distribution, and n the total number of measurements. The hypothesis is accepted if $\chi^2 < \chi^2_{cr}$, the critical value being determined from the adopted confidence level and the corrected number of degrees of freedom. The computations were performed with statistical software at a confidence level of 0.99.



4. Results

The statistical characteristics of the electrospun fibres are given in Table 1.

Table 1. Statistical characteristics of the electrospun nanofibres.

Material	Mean diameter, nm	χ^2	χ^2_{cr}	f corrected	p value
Composite web containing silk fibroin	306.24	4.31	20.1	8	0.82
16% poly(vinyl alcohol) (reference)	249.89	8.32	13.3	4	0.081

For the fibroin-containing material the computed value $\chi^2 = 4.31$ is 4.7 times smaller than the critical value ($\chi^2_{cr} = 20.1$), and for the reference material $\chi^2 = 8.32$ is 1.6 times smaller than its critical value ($\chi^2_{cr} = 13.3$). In both cases, therefore, the log-normal hypothesis is not rejected at a confidence level of 0.99.

The diameter histograms of the two materials are shown in Figs. 3 and 4. Both exhibit the right-skewed asymmetric shape that follows from Eq. (3).

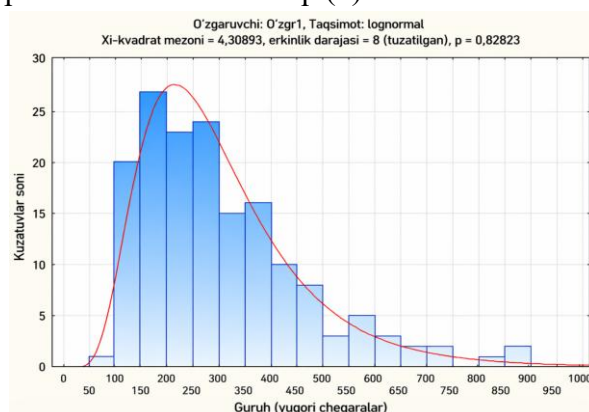


Fig. 3. Histogram of the nanofibre diameter distribution for the material containing silk fibroin.

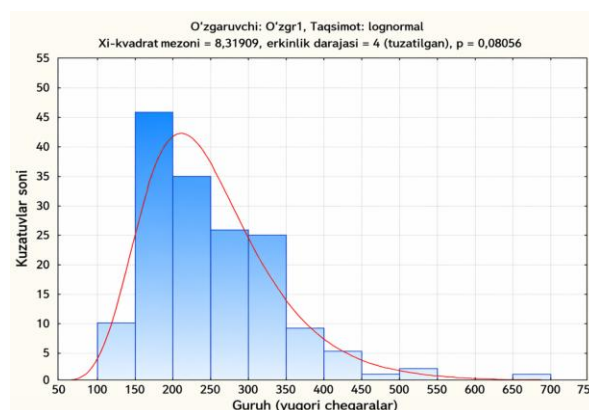


Fig. 4. Histogram of the nanofibre diameter distribution for the reference poly(vinyl alcohol) material

The mean fibre diameter of the fibroin-containing material, 306.24 nm, exceeds that of the reference material (249.89 nm) by 22.6%. Both values lie within the range reported for regenerated silk fibroin, approximately 100–800 nm [5–7].

5. Discussion

5.1. Distribution parameters and the number of splitting stages

The two parameters of Eq. (3) can be recovered from the reported mean and coefficient of variation, since for a log-normal variable

$$\sigma^2 = \ln(1 + V^2), \quad \mu = \ln \bar{d} - \sigma^2/2, \quad (6)$$

where V is the coefficient of variation expressed as a fraction. Applying Eq. (6) to three webs produced under identical solution conditions but at different collector speeds, for which the coefficients of variation are 30.6%, 23.1% and 28.7%, gives the values in Table 2.

Table 2. Log-normal parameters recovered from the measured mean and coefficient of variation.

Specimen	Mean $\bar{d}$, nm	V, %	σ of $\ln d$	μ	Median = exp μ , nm	Mean / median
200 min ⁻¹	180.11	30.6	0.299	5.149	172.2	1.046
1100 min ⁻¹	175.72	23.1	0.228	5.143	171.2	1.026
2000 min ⁻¹	182.23	28.7	0.281	5.166	175.2	1.040

Three observations follow. First, the logarithmic standard deviation is confined to a narrow band, 0.23–0.30, across regimes that differ by a factor of ten in collector speed. Since σ is a property of the splitting process and the collector acts only after solidification, this constancy is precisely what the multiplicative model predicts, and it constitutes an independent check on the model that does not rely on the goodness-of-fit test.

Second, the mean exceeds the median by only 2.6–4.6%. The distribution is therefore only weakly skewed, which explains why some authors have been able to fit a normal law to such histograms with apparent success: at $\sigma \lesssim 0.3$ the log-normal and normal densities are numerically close, and discriminating between them requires either a large sample or a formal goodness-of-fit test rather than visual inspection.

Third, and most usefully, σ can be linked to the number of splitting stages. From Eq. (2), if the individual splitting ratios are identically distributed then

$$\sigma^2 = k \cdot \text{Var}(\ln \xi), \quad (7)$$

so that $k = \sigma^2 / \text{Var}(\ln \xi)$. An independent estimate of k follows from the overall reduction in diameter: a jet of the order of the capillary diameter, say 100 μm , is reduced to 180 nm, a factor of 555, which requires $k \approx 9$ stages if each splitting event halves the diameter, or $k \approx 18$ if the



mean ratio is 0.7. Substituting $\sigma^2 = 0.079$ and $k = 9$ gives $\text{Var}(\ln \xi) \approx 0.009$, corresponding to a standard deviation of the splitting ratio of under 10%.

The implication is that jet splitting in a stable electrospinning process is remarkably reproducible from stage to stage: the observed spread of fibre diameters is generated not by erratic splitting but by the accumulation of many nearly identical, only slightly variable events. This is a stronger statement than the goodness-of-fit result alone supports, and it is testable: a process rendered deliberately unstable should show a marked increase in σ while the mean diameter may barely move, which suggests σ rather than $\bar{d}$ as the sensitive monitoring variable. Analysis of the structure of the nanofibrous materials shows that in both specimens the fibre-diameter distribution conforms to the log-normal law, which confirms the multiplicative splitting model set out in Section 2 and indicates that the electrospinning process is stable. The approach of assessing process stability through a statistical criterion has a practical advantage: it requires no additional measuring equipment and relies only on data obtained from the finished material itself.

The higher mean diameter of the fibroin-containing material compared with the reference has two explanations. First, although the dynamic viscosity of the composite solution (2139.6 mPa·s) is lower than that of the pure 16% poly (vinyl alcohol) solution (3992.5 mPa·s), the composite contains calcium chloride, which alters the ionic composition and electrical conductivity of the solution and hence the charge density carried by the jet. Second, thickenings formed by salt microgranules within the fibres increase the measured mean diameter. The second factor specifically lengthens the right tail of the log-normal distribution and, in accordance with Eq. (4), shifts the arithmetic mean further above the median.

The difference between the p values of the two materials is also meaningful. For the fibroin-containing material $p = 0.82$, appreciably higher than for the reference ($p = 0.081$), that is, the experimental distribution of the composite is closer to the theoretical log-normal one. The comparatively poorer agreement of the reference material is probably associated with the narrower diameter range obtained from a pure poly(vinyl alcohol) solution and the consequent approach of its distribution to normality: in a highly viscous single-component solution (3992.5 mPa·s) the number of jet-splitting stages is small, so that the sum in Eq. (2) satisfies the conditions of the central limit theorem less closely. On this interpretation, the degree of conformity to the log-normal law is itself a measure of how many splitting stages the jet has undergone.

The practical significance of the result lies in monitoring. If under production conditions the fibre-diameter distribution departs from the log-normal law, this indicates that instability has arisen in the process — for example through structuring of the solution, partial blockage of the capillary, or a change in field strength. Periodic testing of conformity to the distribution law can therefore serve as an inexpensive instrument of statistical process control, complementary to the direct measurement of the mean diameter, which by itself does not reveal a change in the shape of the distribution.



Several limitations affect the measurement. First, the diameters were obtained from two-dimensional projections of scanning electron micrographs; the inclination of a fibre relative to the image plane introduces a systematic error into the measured diameter, which is a projection of the true diameter only for fibres lying in the plane. Second, only fibres fully contained within the micrograph were included in the analysis, which may bias the sample in favour of thinner fibres, since coarse fibres are more likely to extend beyond the frame. Third, the distribution parameters μ and σ are not reported here; determining them would permit quantitative comparison of materials obtained under different regimes and is a direction for further work. Fourth, the number of size classes used in the χ^2 test, and hence the corrected degrees of freedom, differ between the two materials (8 and 4), which limits the strict comparability of their p values.

5.2. Consequences for the measurement protocol

The recovered value of σ also determines how many fibres must be measured for a stated precision, a question that is almost never addressed explicitly in the electrospinning literature although diameters are routinely quoted to four significant figures. For a mean estimated from n independent measurements, the half-width of the 95% confidence interval expressed as a fraction of the mean is $1.96 \cdot V/\sqrt{n}$, whence

$$n = (1.96 \cdot V / \delta)^2, \quad (8)$$

where δ is the required relative precision. Table 3 evaluates Eq. (8) for the coefficients of variation observed in these materials.

Table 3. Number of fibre measurements required for a stated relative precision of the mean diameter.

Coefficient of variation V , %	n for $\pm 5\%$ precision	n for $\pm 3\%$ precision
23.0	81	226
28.7	127	352
30.6	144	400

With the $n = 100$ measurements per specimen used here and a coefficient of variation of 30.6%, the achieved half-width is 6.0% of the mean, that is approximately ± 11 nm on a mean of 180 nm. Reporting such a mean as 180.11 nm therefore conveys a precision that the sample size does not support, and the practice of quoting electrospun fibre diameters to two decimal places without a confidence interval should be regarded as misleading.

The requirement is more severe for the shape parameter than for the mean. Estimating σ itself to within $\pm 20\%$ at 95% confidence requires approximately 100 measurements, and to within $\pm 10\%$ approximately 380. Since Section 5.1 argued that σ , rather than the mean, is the variable sensitive to process instability, a monitoring protocol based on the distribution shape requires



sample sizes at the upper end of what is practical from electron micrographs — which is an argument for automated image analysis rather than manual measurement.

A further consideration concerns independence. Fibres measured within a single micrograph are not strictly independent observations, since they were deposited within a short interval and from the same segment of jet; the effective sample size is therefore smaller than the nominal count, and Table 3 should be read as a lower bound. Distributing the required measurements over several micrographs taken at different points of the web, as was done here, mitigates but does not eliminate this effect.

6. Conclusions

Successive splitting of the spinning-solution jet in electrospinning was represented as a multiplicative random process, and the log-normal distribution of nanofibre diameters was justified theoretically on the basis of the central limit theorem, in agreement with the theory of stationary fragmentation processes.

The mean fibre diameter determined from scanning electron micrographs is 306.24 nm for the material containing silk fibroin and 249.89 nm for the reference 16% poly(vinyl alcohol) material; both values correspond to the range reported in the literature for regenerated silk fibroin.

Pearson's criterion at a confidence level of 0.99 confirms conformity of the diameter distributions to the log-normal law in both materials ($\chi^2 = 4.31 < 20.1$ and $\chi^2 = 8.32 < 13.3$), which indicates that the jet-splitting process is stationary and the electrospinning process stable. It was shown that testing conformity to the distribution law can be used as a criterion for monitoring the stability of electrospinning without additional equipment, and that the degree of conformity may itself reflect the number of jet-splitting stages undergone.

Funding. This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Declaration of competing interest. The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability. The data supporting the findings of this study are available from the corresponding author upon reasonable request.

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