

# Experimental report

18/06/2017

**Proposal:** 8-03-887

**Council:** 4/2016

**Title:** Investigation of the fractal chromatin arrangement in the biological cell nuclei by means of the small-angle neutron scattering

**Research area:** Biology

**This proposal is a new proposal**

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**Samples:** the samples of chicken erythrocyte nuclei and samples of Hela nuclei

Instrument	Requested days	Allocated days	From	To
D11	4	3	02/12/2016	05/12/2016
D22	4	0		

**Abstract:**

The most mysterious feature of DNA in the nuclei is the way it is packed so that any part of it could be accessed, read, transcribed and translated at any time in the process of replication. We propose a continuation of SANS experiment to further investigate chromatin organization in the systems such as chicken erythrocyte cells and Hela cells.

# Investigation of the fractal chromatin arrangement in the biological cell nuclei by means of the small-angle neutron scattering

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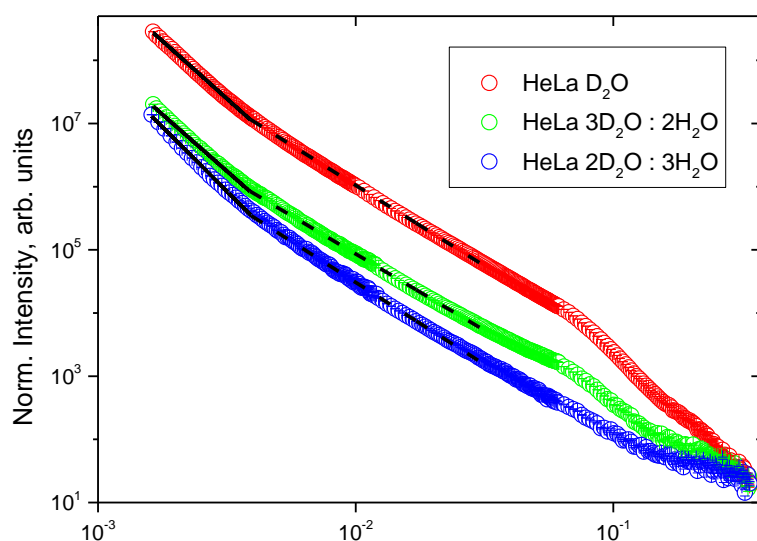
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## Background

Nowadays multiscale fractal model of DNA arrangement inside the nuclei [1,2] is the most promising to explain this mystery. Experimental proof of this theory is complicated firstly because of necessity to investigate nuclei under conditions as close to those inside the organism as possible and secondly by simultaneous coexistence of DNA together with proteins inside the nuclei. Neutron scattering seems to be the most suitable instrument for such investigation due to such advantages of neutrons as high penetration depth, undamaging and possibility to use contrast variation method. The last is very important as it allows one to eliminate separate structure of DNA and proteins in the nuclei.

Previously, arrangement of chromatin in interphase chicken erythrocyte nuclei was investigated by small (and ultra-small) angle neutron scattering [1,2]. The scattering spectra have revealed that on scales between 15 nm and 1.5  $\mu\text{m}$  the interior of the nucleus exhibited properties of a mass fractal. The fractal dimension of the protein component of the cell nucleus was found to be approximately 2.5, while the DNA organization was found to consist of two phases with fractal dimension slightly higher than 2 on scales smaller than 300 nm and approaching 3 on larger scales. The preliminary measurements have shown ability of SANS to reveal the features of the chromatin arrangement in the biological cell nuclei.

## Experiments



*Fig.1 Small-angle neutron scattering from chicken erythrocyte nuclei in 40, 70 and 100% D2O buffers.*

In the present study, we report the experimental results of small-angle neutron scattering (SANS) from HeLa nuclei. The SANS measurements of small-angle neutron scattering curves were performed in the scattering-vector range from  $10^{-3}$  to  $10^0 \text{ \AA}^{-1}$  with the use of the contrast-variation technique at the instrument D11 at the ILL, Grenoble, France. In Fig. 1 we show the SANS spectra measured for HeLa. To study protein structure, samples of cell

nuclei were prepared in a buffer solution with scattering density close to that of DNA (64% D<sub>2</sub>O and 36% H<sub>2</sub>O). The nucleic acid component of chromatin was investigated with the use of a mixture of 40% D<sub>2</sub>O and 70% H<sub>2</sub>O, whose scattering density was close to that of most of proteins. In the experiment a neutron beam with a wavelengths  $\lambda = 6$  nm, and a bandwidth  $\Delta\lambda/\lambda = 0.1$  was used. The SANS measurements were performed at room temperature for three different detector positions: 1.5, 8, 39 m.

## Conclusion

The data processing resulted in the linearization of these data on a double-logarithmic scale. There are linear region all over the measured  $Q$  range in the double-logarithmic plot of the scattering intensity versus momentum transfer ( $I(Q) \sim Q^{-D}$ ). In the range of  $Q$  from  $4 \cdot 10^{-2}$  to  $4 \cdot 10^{-1} \text{ \AA}^{-1}$ , the relationship between the scattering intensity and the scattering vector follows the power law and thus can be linearized when plotted on a double-logarithmic scale with the slope of the linear fit slightly hair then  $D = 2.4$  for all contrast.

For the lower scattering angles, where the values of  $Q$  lie between  $1 \cdot 10^{-3}$  and  $4 \cdot 10^{-3} \text{ \AA}^{-1}$ , the scattering curve plotted on a double-logarithmic scale is also linear, but the power law exponent changes, so that the slope of the linear fit becomes  $D$  slightly higher than 3.5.

We thank the responsible of the D11 SANS facility Dirk Honecker for the excellent support during the experiment.

## References

- [1] D.V. Lebedev, M.V. Filatov, A.I. Kuklin, A.Kh. Islamov, E. Kentzinger, R. Pantina, B.P. Toperverg, V.V. Isaev-Ivanov // FEBS Letters. 2005. V. 579. P. 1465-1468.
- [2] D. V. Lebedev, M. V. Filatov, A. I. Kuklin, A. Kh. Islamov, J. Stellbrink, R. A. Pantina, Yu. Yu. Denisov, B. P. Toperverg, and V. V. Isaev-Ivanov, Crystallography Reports, 2008, Vol. 53, No. 1, pp. 110–115.