DNA loop domain organization in glioblastoma astrocytoma cell lines U373 and U251

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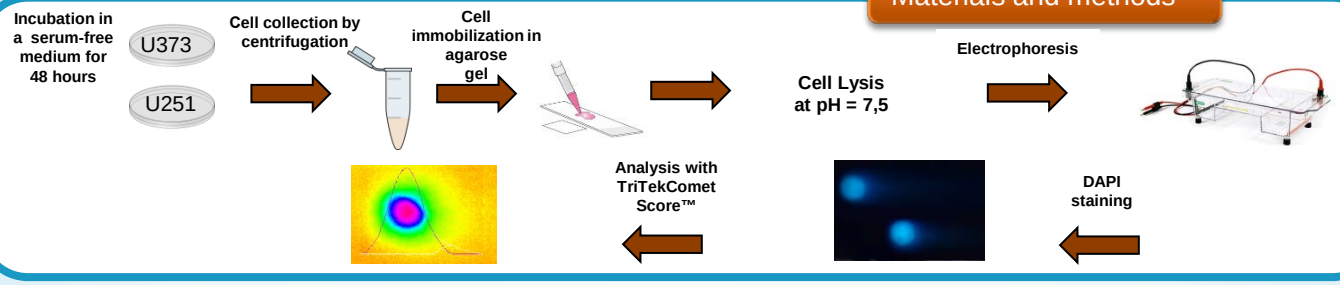
Introduction. Glioblastomas are the most common primary intracranial tumors in adults. Glioblastoma astrocytoma U373 cell line is frequently used as a model object for studying pathogenesis of this type of cancer. However, the identity of this cell line has often been questioned due to its cross-contamination by another glioblastoma astrocytoma cell line U251. Thus, the chromatin organization in these cells requires special attention. Therefore, the **aim** of the study was to investigate the DNA loop domain organization in two glioblastoma astrocytoma cell lines – U373 and U251.

Materials and methods



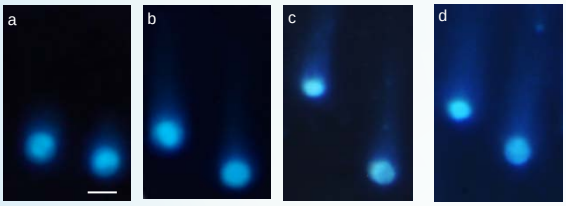


Figure 1. Typical images of comets after 20 (a, c) and 60 (b, d) minutes of electrophoresis for U373 (a, b) and U251 (c, d) cell lines. Bar – 10 nm.

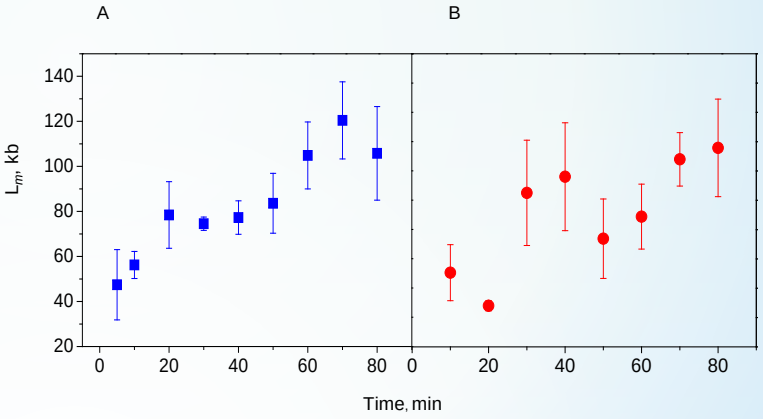


Figure 4. The contour length L_m of the longest loops as functions of electrophoresis time for nucleoids obtained from U373 (a) and U251 (b) cells.

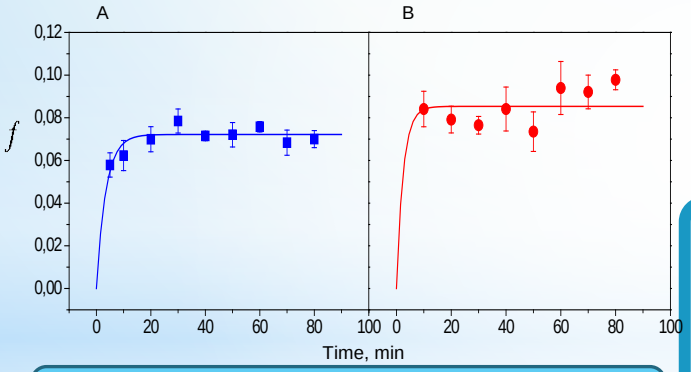


Figure 2. The average relative amount *f* of DNA in the comet tails as functions of electrophoresis time for nucleoids obtained from U373 (a) and U251 (b) cells. Each point is an average for 5 to 7 independent experiments, error bars are the standard errors.

Cell type	Kinetic parameters	
	k, min^{-1}	*A*
U373	0,29±0,05	0,07±0,001
U251	0,41±0,04	0,08±0,003

Figure 3. Kinetic parameters of DNA exit from nucleoids derived from U373 and U251 cells;
k is the rate constant; *A* is a maximum fraction of DNA that can exit into the tail.

Results

We used the kinetic approach to assess the DNA loop domain organization in the glioblastoma astrocytoma cell lines. It was shown that for both U373 and U251 cells the kinetic plots of DNA exit into the comet tails had one-step shape, which represents the migration of DNA loops from the nucleoid surface. Although the amount of DNA in the surface loops of both cell lines was almost the same and corresponded to ~0,07 and ~0,08 for U373 and U251 respectively, the rate constant *k* was higher for U251 cells ($p<0,05$).

The contour length of the surface loops was almost similar in both cell lines and varied from ~40 to ~120 kb. It should be noted that the migration of the inner DNA loops was not observed for U373 and U251 cells at all.

It was also shown that the loop densities, which represent the correlations between the length of the longest loops and the relative amount of DNA in the tails, were ~0,07 kb^{-1} in both cell lines.

In contrast to the cells described above, it was previously established by our research group that in another glioblastoma cell line T98G the kinetic plots of DNA exit had two distinct stages. The amount of DNA in the first stage was ~0,07. And there was an additional increase (~0,05) in the DNA fraction in the comet tails on the second stage reflecting the exit of the inner supercoiled loops.

Conclusion

The obtained results indicate similarity in both the DNA fraction in the surface loops and in their contour length for U373 and U251 cell lines.