Computer methods for visualization chromosome-specific DNA sequences in FISH images

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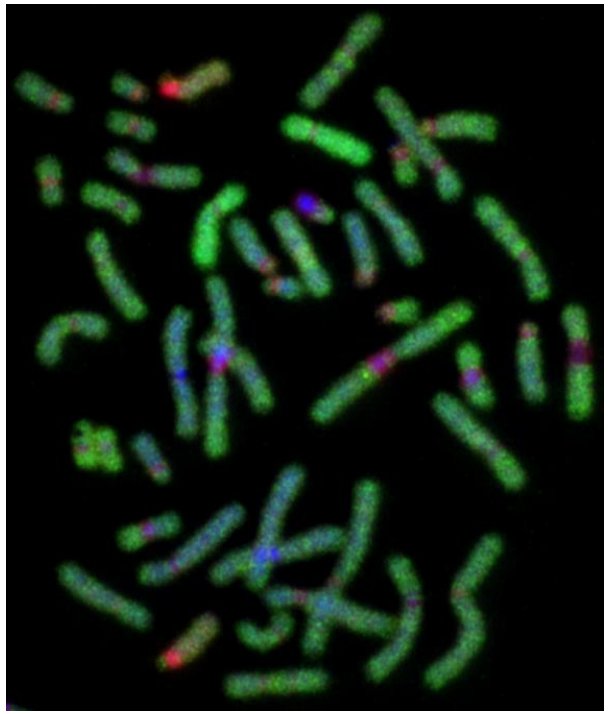
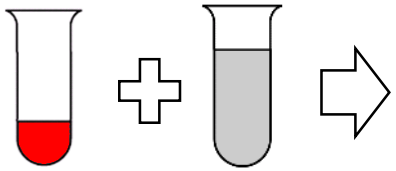
Problem



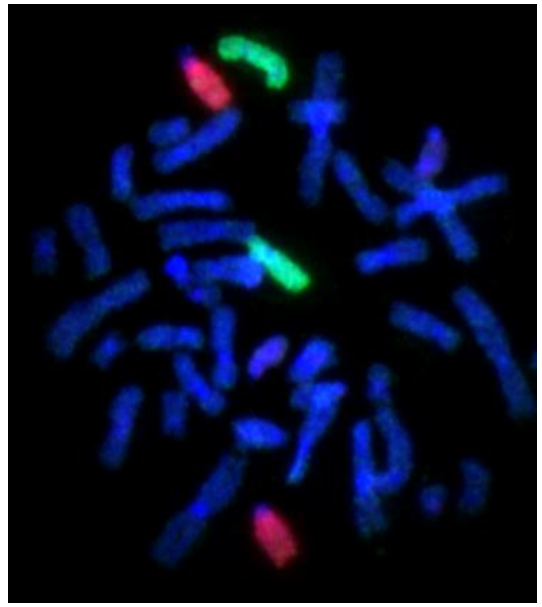
The direct use of DNA-probes appears in a high background level of hybridization.

Both figure is fluorescence in situ hybridization (FISH) DNA-probes, obtained from 15 (red) and 10 (green) human chromosomes to metaphase chromosomes (left – usual FISH, right – CISS-hybridization)

Traditional solution: CISS-hybridization



Pre-hybridization of the DNA-probe with an unlabelled repetitive DNA fraction



Problem of availability of DNA material in necessary volume to do CISS-hybridization

CISS-hybridization not effective in cases, when genome contain too high numbers of repetitive sequences

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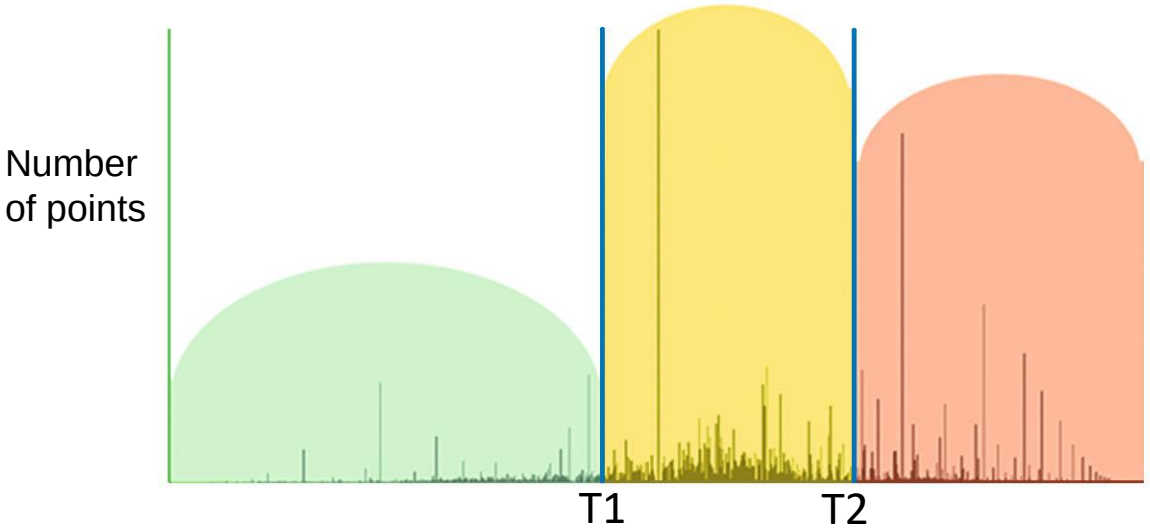
Both methods use the results of hybridization of two different labeled probes

The main idea of image analysis methods: DNA-probe intensity signals from interspersed DNA sequences are similar.

Method RENS (Rens et al., 2006)

Expert sets the limits of the intensity ratio range for the signals determined by repetitive sequences



Ratio between signal intensities from two fluorochromes in a point

Method VISSIS (Bogomolov et al., 2012)

Signals from the chromosome-specific sequences = The total signal at the point of grayscale image − Signal intensity of another DNA probe

Materials

FISH of DNA probes obtained from human chromosomes 3, 7, 8, 10, 13, 15, 18, 19 and X with human chromosomes was performed according to the standard CISS-hybridization protocol and without suppression of hybridization of the repetitive sequences

REFERENCES

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2) A.G. Bogomolov, K.S. Zadesenets, T.V. Karamysheva, N.L. Podkolodnyi, N.B. Rubtsov, “Visualization of chromosome-specific DNA sequences by fluorescence in situ hybridization of microdissection DNA probes with metaphase chromosomes,” Russian Journal of Genetics: Applied Reserch Vavilov J. Genet. Sel., 2012, vol. 2, pp. 413–420

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Results

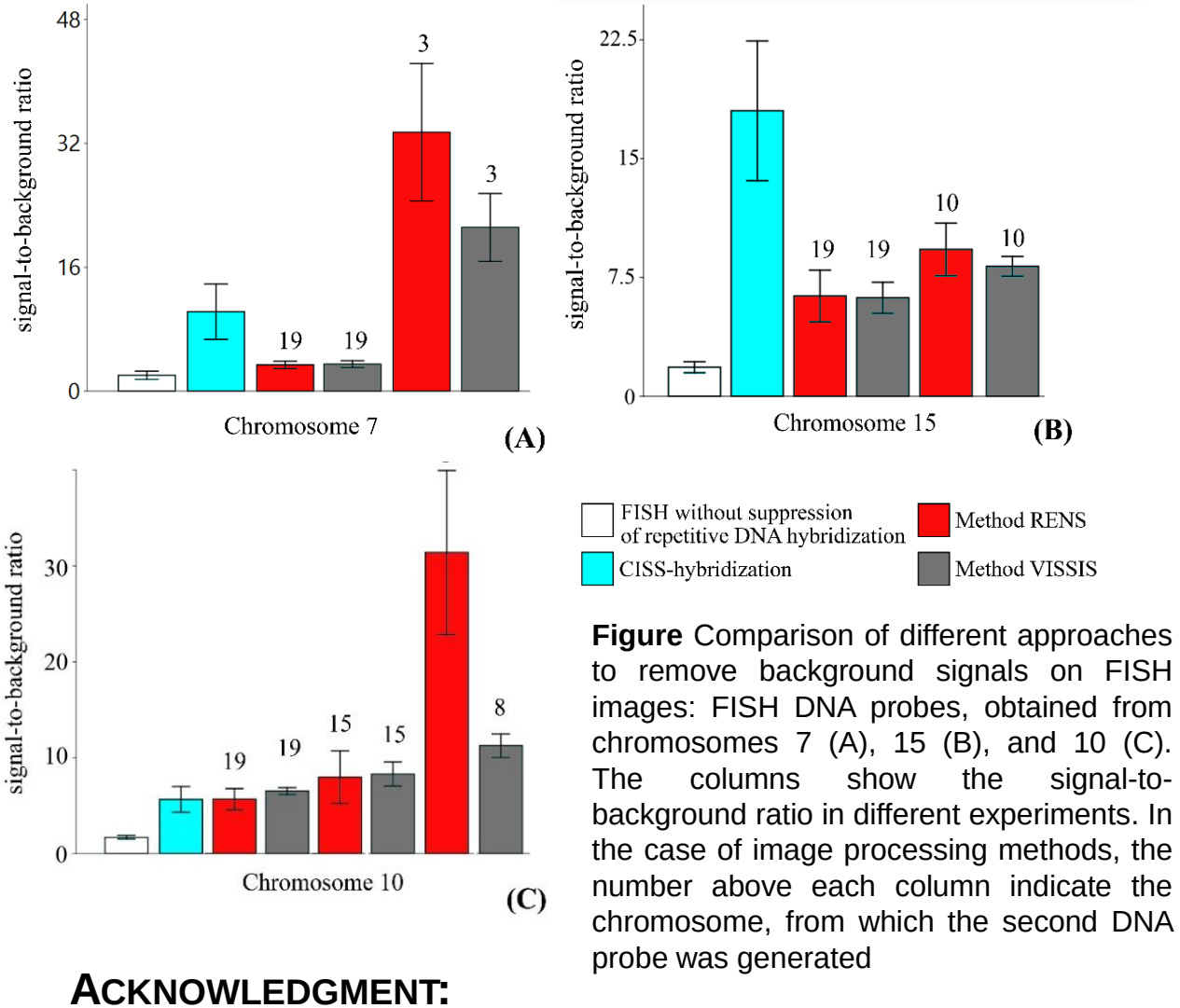

Figure Comparison of different approaches to remove background signals on FISH images: FISH DNA probes, obtained from chromosomes 7 (A), 15 (B), and 10 (C). The columns show the signal-to-background ratio in different experiments. In the case of image processing methods, the number above each column indicate the chromosome, from which the second DNA probe was generated

$$DChr(a,b) = \sqrt[2]{(s_a - s_b)^2 + (l_a - l_b)^2}$$

where a and b – names of source chromosomes;
s_a and s_b – ratio of short interspersed nuclear elements number of sequences in chromosomes a and b;
l_a and l_b – ratio of long interspersed nuclear elements number of sequences in chromosomes a and b

We found **negative correlation between signal-to-background ratio of processed images and *Dchr*** of chromosomes, from which DNA probes were obtained (Pearson's coefficients, Spearman's rank correlation, Kendall's rank correlation, $p<0.01$).

Conclusion

Computer methods can replace the step of preannealing with repetitive DNA in FISH experiments.

But their effectiveness for background signal removing depends on the pairs of DNA probes that derived from different chromosomes.

ACKNOWLEDGMENT:

The reported study was funded by RFBR according to the research project № 19-015-00084a and by project 0324-2019-0042 of IC&G SB RAS.