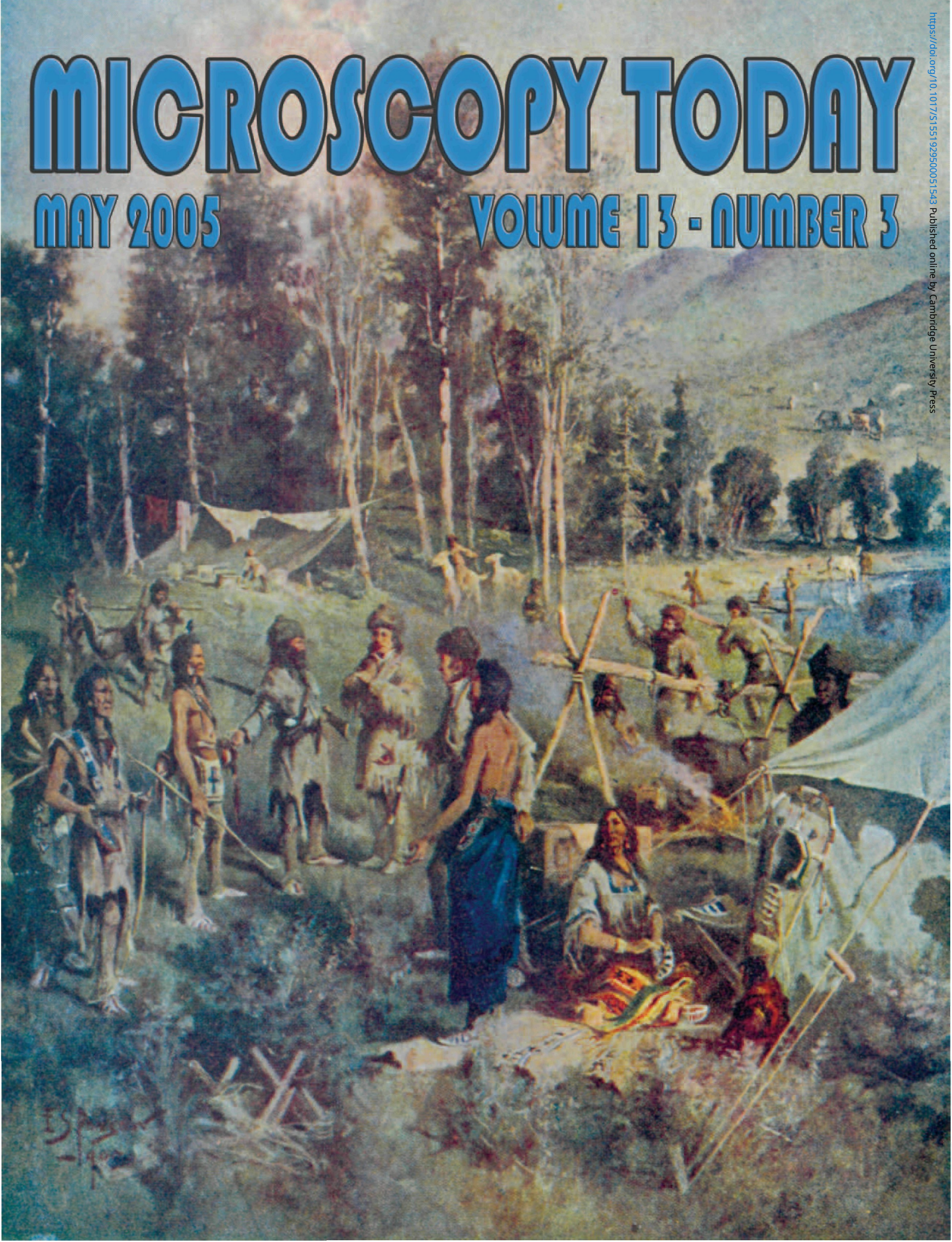
MICROSCOPY TODAY

MAY 2005 VOLUME 13 - NUMBER 3



Don't Take Charge

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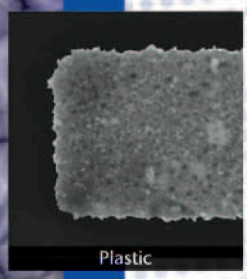
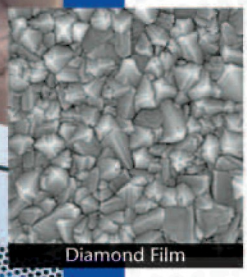
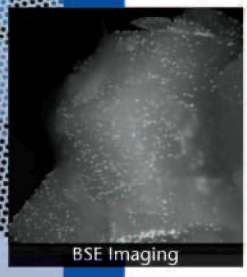
TOOLS FOR NANOTECH


Plastic


5 nm Platinum Particles


Diamond Film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Visualizing Gene Expression in Real-Time

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Gene expression has been visualized for a few decades, but in static forms such as blots and gene chips. Susan Janicki, Toshiro Tsukamoto, Simone Salghetti, William Tansey, Ravi Sachidanandam, Kannanganattu Prasanth, Thomas Ried, Yaron Shav-Tal, Edouard Bertrand, Robert Singer, and David Spector have recently designed a cell line in which gene expression can be observed with stunningly accurate spatial and temporal resolution.[2] Gene expression is a cascade of events beginning with transcription of RNA from the DNA template and ending with translation into a protein sequence. Janicki *et al.* were able to visualize the entire process at the levels of DNA, RNA, and proteins in living cells!

As pointed out by the authors, several specific steps in the gene expression paradigm have been discovered and extensively studied *in vitro*. However, the dynamics of mRNA synthesis at a single specific transcription site has not been revealed. They developed a cell line that allows the investigation of how events of gene expression are coordinated spatially and temporally *in vivo*. They began by constructing a plasmid, containing a transcription unit, and stably integrating it into a human cancer cell line. With the addition of other plasmids, they had a cell line that produced a series of detectable markers at specific events during gene expression. Transfected cells were maintained in a physiologic chamber on an inverted microscope and time-lapse imaging was acquired. Stacks of several images 0.5 µm apart were taken to assure that at least one image was focused on the structure of interest for each time point. Different wavelengths were used to excite the different reporter fluorochromes.

After extensive analysis, the interconnectedness of chromatin remodeling, transcription, mRNA processing, and messenger ribonucleoprotein (mRNP) export became apparent. Janicki *et al.* developed a cell system that allows the visualization of an inducible array of transcription units and their RNA and protein products in living cells. This allows the evaluation of dynamic changes in chromatin structure, RNA synthesis, and factor dissociation/association during the induction of transcription. The movie can be seen at http://spectorlab.cshl.edu. This study provided significant insight into the dynamic spatial and temporal changes that occur as chromatin transitions from a heterochromatic to a euchromatic state. This system has considerable potential to address a broad range of questions relating to gene expression, DNA replication, and chromatin stability! ■

References

1. The author gratefully acknowledges Dr. David Spector for reviewing this article.
2. Janicki, S.M., T. Tsukamoto, S.E. Salghetti, W.P. Tansey, R. Sachidanandam, K.V. Prasanth, T. Ried, Y. Sharon Shav-Tal, E. Bertrand, R.H. Singer, and D.L. Spector, From silencing to gene expression: Real-time analysis in single genes, Cell 116:683-698, 2004.

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ABOUT THE COVER

Lewis and Clark in Camp on Traveler's Rest, (Lolo) Creek, Montana
September 10, 1805

by Edgar Paxson

This watercolor (1903, 5-1/2" by 7"), the frontispiece for Volume 1 of Wheeler's /The Trail of Lewis and Clark/ (1904), became the prototype for one of the murals Paxson painted for the Missoula County Courthouse in 1914. From Discovering Lewis & Clark®, http://www.lewis-clark.org © 1998 VIAs Inc. See the article by Alden, *et al.* on page 8.