METHODS IN BRIEF

STEM CELLS

Abnormalities during reprogramming

Somatic cells can be rendered pluripotent by transfer into an enucleated oocyte or by the forced expression of reprogramming factors. Which of these processes is more faithful remains unknown. With recent successful embryonic stem (ES) cell derivation from zygotes generated by somatic cell nuclear transfer (SCNT), it has become possible to compare otherwise isogenic pluripotent cells derived by these different processes. Ma *et al.* compared human SCNT-ES cells and induced pluripotent stem cells (iPSCs) generated using the same source of dermal fibroblasts as donor cells. These were further compared to ES cells derived from *in vitro* fertilization (IVF) of oocytes from the same donor used for SCNT. The researchers found that copy-number variations are comparable between iPSCs and SCNT-ES cells but that SCNT yields more complete epigenetic reprogramming than IVF controls.

Ma, H. *et al. Nature* **511**, 177–183 (2014).

GENE EXPRESSION

Following transcription in real time

Transcription of highly expressed genes has been found to occur in stochastic bursts, a scheme thought to play an evolutionary role in maintaining population diversity. In order to understand the mechanism behind this phenomenon in bacteria, Chong *et al.* applied single-molecule methods. To monitor transcription elongation in real time, they used a fluorogenic RNA stain and, using total-internal-reflection fluorescence microscopy, recorded time-lapse movies of nascent mRNAs being produced on hundreds of single, immobilized DNA templates. This *in vitro* approach, along with a live-cell mRNA fluorescence *in situ* hybridization assay, allowed them to observe that a buildup of positive DNA supercoiling slows down and eventually halts transcription elongation, but gyrase binding to supercoiled DNA restarts transcription. Chong *et al.* concluded that supercoiling dynamics caused by gyrase association and dissociation are responsible for transcriptional bursting in bacteria.

Chong, S. *et al. Cell* **158**, 314–326 (2014).

CELL BIOLOGY

Intracellular topology

To capture the dynamic structure of chromatin in living cells, Baum *et al.* used an approach established for diffusion nuclear magnetic resonance spectroscopy in which the mobility of a nanosensor is used to gather information about a structure. They traced the mobility of GPF monomers, trimers and pentamers in the cytoplasm and the nucleus by fluorescence correlation spectroscopy with a line-illuminating multifocus fluorescence microscope. The topology of the intracellular space determines accessibility and dwell time of proteins; the researchers' measurements allowed them to generate a model of the intracellular architecture at nanometer scale. Their findings point to random obstacle structures in the nucleus rather than fractal or corral structures with fixed sizes.

Baum, M. *et al. Nat. Commun.* **5**, 4494 (2014).

GENETICS

Identifying genetic barriers to reprogramming

The ability to reprogram somatic cells to induced pluripotency has revolutionized many areas of biological research. For the most part, however, this process is quite inefficient. In recent work, Qin *et al.* carried out pooled short hairpin RNA (shRNA) screens to systematically identify genes that function as barriers to reprogramming. They screened almost 20,000 human genes with 30 shRNAs per gene in human BJ fibroblasts undergoing reprogramming with lentivirally delivered factors. Using sequencing, they identified genes that are more likely to be knocked down in cells expressing the pluripotency marker TRA-1-81. Statistical analysis and further validation led to the identification of 956 genes as barriers to reprogramming, including genes involved in processes such as cell adhesion, endocytosis and ubiquitination. The researchers make their data available as an online resource.

Qin, H. *et al. Cell* **158**, 449–461 (2014).