HIGHLIGHTS

HIGHLIGHT ADVISORS

WENDY BICKMORE

MRC HUMAN GENETICS UNIT,
UK

SEAN B. CARROLL

UNIVERSITY OF WISCONSIN,
USA

ADAM EYRE-WALKER

UNIVERSITY OF SUSSEX, UK

JANE GITSCHIER

UNIVERSITY OF CALIFORNIA,
SAN FRANCISCO, USA

RALPH J. GREENSPAN

THE NEUROSCIENCES
INSTITUTE, CALIFORNIA, USA

YOSHIHIDE HAYASHIZAKI

RIKEN GENOMIC SCIENCES
CENTER, JAPAN

PETER KOOPMAN

UNIVERSITY OF QUEENSLAND,
AUSTRALIA

LEONID KRUGLYAK

FRED HUTCHINSON CANCER
RESEARCH CENTER, USA

BARBARA MEYER

UNIVERSITY OF CALIFORNIA,
BERKELEY, USA

LEE NISWANDER

SLOAN-KETTERING INSTITUTE,
NEW YORK, USA

CHRISTOS OUZOUNIS

THE EUROPEAN
BIOINFORMATICS INSTITUTE,
UK

NORIYUKI SATOH

KYOTO UNIVERSITY, JAPAN

MARC VIDAL

DANA-FARBER CANCER
INSTITUTE, BOSTON, USA

VIRGINIA WALBOT

STANFORD UNIVERSITY, USA

DETLEF WEIGEL

MAX PLANCK INSTITUTE FOR
DEVELOPMENTAL BIOLOGY,
GERMANY

LEONARD I. ZON

CHILDREN'S HOSPITAL,
BOSTON, USA

TECHNOLOGY

All the king's horses and all the king's men...

Humpty Dumpty had a famous problem — he sat on a wall, had a great fall and...couldn't be put back together again. Genome biologists have had a similar problem until now — they could break genomes up and sequence them but not synthesize them *de novo*, or at least not in any efficient way. A group led by Craig Venter has now devised a method for assembling the complete genome of a bacteriophage from a pool of chemically synthesized oligonucleotides. Although still in need of a correcting mechanism, the method might in future be used to make synthetic chromosomes.

Driven by the desire to improve on previous attempts to assemble active viral genomes, Smith *et al.* chose the ϕ X174 bacteriophage — the first living organism to have its 5,386 bp genome sequenced — to test their three-step approach to synthetic genome assembly. The first step involves oligonucleotide synthesis, based on both strands of the phage genome. In the second step, the oligonucleotides are phosphorylated, mixed and ligated to give, on average, 700 bp double-stranded fragments. Two rounds of polymerase cycle assembly are used in the third step to assemble these fragments into full-length genome molecules, which are PCR-amplified, linearized and tested for size by electrophoresis.

As the ultimate test of successful

in vitro synthesis is that of molecular function, recircularized synthetic phages were electroporated into *E. coli* to test for their infectivity. Careful comparison of infection efficiency between the wild-type and the synthetic phages indicated that some 9–10 inactivating mutations were present per synthetic genome, although at least one synthetic isolate was 100% identical to the original ϕ X174 sequence.

Smith *et al.* suggest that these point mutations probably result either from replication errors or are already present in the oligonucleotides.

Given the mutation rate, the authors point out that without an ability to select for the wild-type sequence, synthetic forms will carry, on average, a point mutation per 2 kb. Undeterred by this, they propose that, in combination with DNA sequencing and site-directed repair, their method can be used to assemble larger genomes.

Synthetic genomics could solve the problems of dealing with degraded biological material or extinct species. Genome sequences could also be modified during syn-

Illustration courtesy of Smudge

thesis to provide an alternative to genetic engineering. Although this application of synthetic genomics might be around the corner for microbial genomes, much larger, metazoan genomes might remain in the 'Humpty Dumpty' class for a while yet.

Magdalena Skipper

References and links

ORIGINAL RESEARCH PAPER Smith, H. O. *et al.* Generating a synthetic genome by whole genome assembly: ϕ X174 bacteriophage from synthetic oligonucleotides. *Proc. Natl Acad. Sci. USA* 2 Dec 2003 (doi:10.1073/pnas.2237126100)