and release. It could also sustain a compressive stress about ten times higher than that of an ultralight metallic microlattice with a similar density[9], and can be elongated by 16.5% before fracturing. In addition, using only reduced GO sheets, Sun *et al.* prepared the lightest known aerogel: with a density of 0.16 mg cm^{-3}, the material is more than 7 times lighter than air.

As well as producing aerogels that are ultralight and super-elastic, the synergistic effect between graphene and CNTs also yields aerogels that have excellent thermal stability, high absorption capacity for liquids, and good electrical conductivity[1]. For example, the resulting aerogels have an ultrahigh oil-absorption capacity of 215–913 times their own weight, depending on the oil density, which is up to two orders of magnitude higher than those of commercial oil absorbents. What's more, the aerogel's superelasticity means that it can be used repeatedly following mechanical extrusion to release the absorbed oil. Taken together with several other studies[3,4,7,8], Sun and colleagues' results expand the range of applications for carbon aerogels.

"Sun and colleagues' work demonstrates how the synergistic assembly of two types of carbon nanomaterial can be used to create multifunctional materials."

However, two major challenges remain: preparing aerogels that have a more ordered network of thin, solid walls than that achieved here; and fully translating the excellent intrinsic properties of graphene and CNTs, such as their electrical and thermal conductivity, into the aerogels. The latter aspect is plagued by the electrical and thermal resistance between CNTs and/or reduced GO sheets, and by the poor electrical and thermal conductivity of reduced GO. The technique of template-directed chemical-vapour deposition may provide a solution, because graphene foams prepared by this method[10], with nickel or other metal foams serving as a template, have an electrical conductivity two to three orders of magnitude higher than that of a graphene–CNT aerogel with a similar density.

The hybrid aerogels may also prove beneficial in composite materials, in which the pores of solid foams are filled with another material to improve their properties. Graphene-foam-based composites have already found several diverse uses, such as elastic conductors[10], flexible lithium-ion batteries[11], supercapacitors[12] and materials for lightweight electromagnetic interference shielding[13]. Basing composite structures on the hybrid aerogels may further widen the range of applications.

Sun and colleagues' work demonstrates how the synergistic assembly of two types of carbon nanomaterial can be used to create multifunctional materials. Besides graphene and CNTs, a vast number of nanomaterials have been developed since the 1980s, some of which have complementary properties to carbon-based nanomaterials. Extending the authors' synergistic approach to other nanomaterials may produce systems with various novel properties. Indeed, new electronic systems have been created by combining graphene with boron nitride or molybdenum disulphide sheets in a particular stacking order[14]. Finally, it is the nanostructured nature of these building blocks that gives them mechanical flexibility and other useful properties, and this makes them an appealing choice for building flexible devices. ■

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1. Sun, H., Xu, Z. & Gao, C. *Adv. Mater.* **25,** 2554–2560 (2013).
2. Kistler, S. S. *Nature* **127,** 741 (1931).
3. Qiu, L., Liu, J. Z., Chang, S. L. Y., Wu, Y. & Li, D. *Nature Commun.* **3,** 1241 (2012).
4. Hu, H., Zhao, Z., Wan, W., Gogotsi, Y. & Qiu, J. *Adv. Mater.* **25,** 2219–2223 (2013).
5. Geim, A. K. *Science* **324,** 1530–1534 (2009).
6. Saito, R., Dresselhaus, G. & Dresselhaus, M. S. *Physical Properties of Carbon Nanotubes* (Imperial College Press, 1998).
7. Nardecchia, S., Carriazo, D., Ferrer, M. L., Gutiérrez, M. C. & del Monte, F. *Chem. Soc. Rev.* **42,** 794–830 (2013).
8. Kim, K. H., Oh, Y. & Islam, M. F. *Nature Nanotechnol.* **7,** 562–566 (2012).
9. Schaedler, T. A. *et al. Science* **334,** 962–965 (2011).
10. Chen, Z. *et al. Nature Mater.* **10,** 424–428 (2011).
11. Li, N., Chen, Z., Ren, W., Li, F. & Cheng, H.-M. *Proc. Natl Acad. Sci. USA* **109,** 17360–17365 (2012).
12. Cao, X. *et al. Small* **7,** 3163–3168 (2011).
13. Chen, Z., Xu, C., Ma, C., Ren, W. & Cheng, H.-M. *Adv. Mater.* **25,** 1296–1300 (2013).
14. Britnell, L. *et al. Science* **335,** 947–950 (2012).

DNA REPLICATION

Driving past four-stranded snags

Unusual DNA structures, such as G-quadruplexes, can stall DNA replication with drastic consequences for the cell. The Pif1 helicase family of enzymes has evolved to disentangle these structures efficiently. SEE ARTICLE P.458

SERGEI M. MIRKIN

Genomic DNA is long, shows profound sequence redundancy and is heavily tangled. These properties create formidable problems during DNA replication — the process at the heart of heredity. A particular obstacle is the presence of repetitive runs of DNA that can form unusual structures such as hairpins, cruciforms, three-stranded triplexes and four-stranded G-quadruplexes. In this issue, Paeschke *et al.*[1] (page 458) describe an evolutionarily conserved mechanism that promotes replication past G-quadruplexes*.

Unusual DNA structures are less favourable than the common DNA double helix, and form only if the duplex is stimulated to unwind, exposing single strands. The replication process provides plenty of stimuli for this. For example, during replication, replicative helicase enzymes efficiently separate the two strands to form a two-pronged replication fork (Fig. 1a). In addition, DNA polymerase enzymes — which use these DNA strands as templates for replication — function only in a particular direction along the DNA strand (5′ to 3′), whereas the two strands have antiparallel orientation. Consequently, synthesis along the leading-strand template is continuous, whereas synthesis along the lagging strand is discontinuous, proceeding in short increments known as Okazaki fragments. This leads to the presence of long, single-stranded regions at the fork, called Okazaki initiation zones, at which the next fragment will be synthesized.

Okazaki initiation zones provide convenient places for formation of unusual DNA structures, including G-quadruplexes. Earlier bacterial, yeast and mammalian studies implied that formation of such structures along the lagging-strand template slows down replication and increases the likelihood of chromosomal breakage and genomic rearrangements[2].

Formation of G-quadruplexes was also predicted[3] along the leading-strand template, in regions of single-stranded DNA that transiently arise between the replicative helicases and leading-strand DNA polymerase. These predictions were corroborated recently[4] when accumulation of G-quadruplexes was demonstrated during the S-phase of the cell

*This article and the paper under discussion[1] were published online on 8 May 2013.

cycle — the phase at which replication occurs.

Because formation of unusual DNA structures is a recurrent problem, cells have evolved defensive mechanisms to deal with them. The first line of defence is a protein that binds to single-stranded DNA; it is known as replication protein A (RPA) in eukaryotic organisms such as animals, plants and fungi, and is an essential component of the replication fork. RPA covers the Okazaki initiation zone and prevents DNA base pairing, thus efficiently stopping it from folding into various secondary structures.

But every guardian has its Achilles heel, and for RPA that is its low affinity for DNA that is made of repetitive purine (adenine and guanine) bases[5]. Therefore, G-quadruplexes, which are mainly made of guanines, can overcome this line of defence, and fold into stable secondary structures. When this happens, DNA polymerase suddenly faces a four-stranded obstacle in the template, which it cannot disentangle under normal conditions[6] (Fig. 1b). This stalls the entire replication fork, occasionally leading to fork reversal and chromosomal fragility[7].

Enter the second line of defence — accessory DNA helicases. Paeschke and colleagues' study is devoted to one family of these enzymes, called Pif1. These helicases, which untangle DNA moving in the 5′ to 3′ direction, were initially identified in the budding yeast *Saccharomyces cerevisiae*, but have since been found in organisms as diverse as bacteria and humans[8].

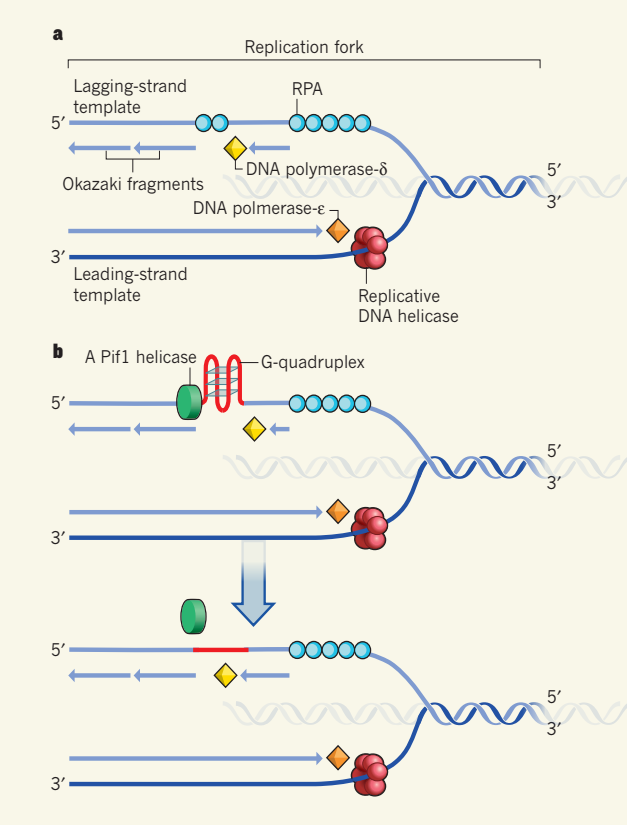

Figure 1 | The problem at G-quadruplexes. During DNA replication, replicative DNA helicases unwind the two strands into a leading- and a lagging-strand template to form a replication fork. Whereas DNA polymerase-ε promotes continuous replication along the leading-strand template, DNA polymerase-δ copies the lagging-strand template in short increments called Okazaki fragments, which are joined together later. **a**, Where the lagging strand is exposed — for instance, at Okazaki initiation zones — a single-strand DNA-binding protein, replication protein A (RPA), binds to it to ensure smooth progress of replication along the strand, and the fork as a whole. **b**, Where this strand contains DNA repeats rich in guanine bases, RPA is inefficient, allowing the strand to fold into secondary structures, including G-quadruplexes. Consequently, progress of DNA polymerase-δ is blocked, leading to stalling of the replication fork. Paeschke *et al.*[1] report that Pif1 helicases unwind G-quadruplexes, allowing replication to progress past this obstacle.

S. cerevisiae has two Pif1-family helicases: Pif1 and Rrm3. Pif1 prevents replication-fork stalling at G-quadruplexes[9], whereas Rrm3 helps to resolve 'collisions' between transcription and replication processes along DNA[10]. Paeschke *et al.* demonstrate that Pif1-family helicases from various species unwind G-quadruplexes extremely quickly and efficiently. Many other DNA helicases were shown to unwind G-quadruplexes, but the Pif1 helicases seem to be the best for this task. Moreover, G-quadruplexes are by far these helicases' favourite substrates.

The authors also demonstrate that the absence of Pif1 helicases in yeast causes a drastic increase in gross chromosomal rearrangements, particularly those that originated at G-quadruplex-forming sequences. Most importantly, this high genetic instability was efficiently suppressed by expressing bacterial or human Pif1 helicases in the helicase-deficient yeast strain. Thus, unravelling G-quadruplexes seems to be a conserved function of Pif1 helicases that arose very early in evolution.

Most unexpected are the data that came from Paeschke and co-workers' characterization of the rearrangement events triggered by G-quadruplexes in helicase-deficient yeast strains. As in their previous set of experiments, the authors introduced 'reporter' genes into a non-essential arm of the yeast chromosome to track these events. In normal strains, most events that manifested as reporter inactivation were caused by the loss of the chromosomal region carrying the reporters. By contrast, in helicase-deficient strains, the reporters' activity was not lost owing to the loss of the corresponding genes, but because the genes were epigenetically silenced — that is, their histone proteins were chemically modified, without alteration to the DNA sequence. This silencing is similar to a genetic phenomenon known as position-effect variegation[11], which also depends on the activity of the histone-modifying deacetylase enzyme sirtuin-2. As the yeast clones continued to grow on selective media, they underwent genetic rearrangements leading to actual loss of the reporter genes.

Paeschke and colleagues' findings become curiouser and curiouser, with this provocative result prompting them to coin the term complex genetic–epigenetic events. It is difficult to explain this concept in known molecular terms. The authors speculate that the presence of a G-quadruplex motif could trigger local epigenetic silencing, which somehow can be extended to the rest of the chromosome when Pif1 helicases are absent, impairing replication. Theoretically, one can think of such potential mechanisms as altered histone deposition and/or accumulation of epigenetic marks on them owing to impaired replication. Many other scenarios should also be considered.

From this study, the Pif1 family of DNA helicases emerges as a principal player in rescuing genomes from the negative effects of G-quadruplexes — which include replication-fork impairment, epigenetic silencing of surrounding genes and gross chromosomal rearrangements. G-quadruplexes are extremely common elements of the human genome. They are required for the genome's normal activities (such as telomere maintenance and immunoglobulin-switch recombination), but are also implicated in its malfunction, resulting in genetic rearrangements and human disease, including cancer[12]. This will, no doubt, stimulate studies of Pif1 helicases in health and disease for years to come. ■

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1. Paeschke, K. *et al. Nature* **497,** 458–462 (2013).
2. Mirkin, E. V. & Mirkin, S. M. *Microbiol. Mol. Biol. Rev.* **71,** 13–35 (2007).
3. Lopes, J. *et al. EMBO J.* **30,** 4033–4046 (2011).
4. Biffi, G., Tannahill, D., McCafferty, J. & Balasubramanian, S. *Nature Chem.* **5,** 182–186 (2013).
5. Wold, M. S. *Annu. Rev. Biochem.* **66,** 61–92 (1997).
6. Woodford, K. J., Howell, R. M. & Usdin, K. *J. Biol. Chem.* **269,** 27029–27035 (1994).
7. Anand, R. P. *et al. Nucleic Acids Res.* **40,** 1091–1105 (2012).
8. Bochman, M. L., Sabouri, N. & Zakian, V. A. *DNA Repair* **9,** 237–249 (2010).
9. Ribeyre, C. *et al. PLoS Genet.* **5,** e1000475 (2009).
10. Ivessa, A. S. *et al. Mol. Cell* **12,** 1525–1536 (2003).
11. Perrod, S. & Gasser, S. M. *Cell. Mol. Life Sci.* **60,** 2303–2318 (2003).
12. Maizels, N. & Gray, L. T. *PLoS Genet.* **9,** e1003468 (2013).