EMBO reports

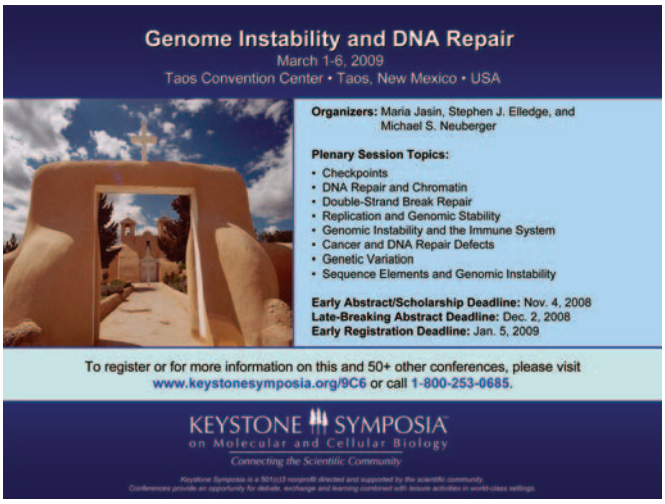
meeting report

Dealing with dangerous accidents: DNA double-strand breaks take centre stage

Symposium on Genome Instability and DNA Repair

Vincenzo Costanzo[1+], *Jayanta Chaudhuri*[2++], *Jennifer C. Fung*[3+++] & *John V. Moran*[4++++]

[1]London Research Institute, South Mimms, UK, [2]Memorial Sloan Kettering Cancer Center, New York, USA, [3]University of California, San Francisco, California, USA, and [4]Howard Hughes Medical Institute, University of Michigan Medical School, Ann Arbor, Michigan, USA



The Keystone Symposium on Genome Instability and DNA Repair took place between 1 and 6 March 2009, in Taos, New Mexico, and was organized by M. Jasin, S.J. Elledge & M.S. Neuberger.

Keywords: copy-number variation; DNA repair; genome instability; non-homologous end joining; recombination

EMBO *reports* (2009) **10**, 837–842. doi:10.1038/embor.2009.173

[1]Genome Stability, London Research Institute, South Mimms, Hertfordshire EN63LD, UK
[2]Immunology Program, Memorial Sloan Kettering Cancer Center, New York, New York 10021, USA
[3]Department of Obstetrics, Gynecology and Reproductive Sciences, University of California, San Francisco, California 94158, USA
[4]Howard Hughes Medical Institute, Departments of Human Genetics and Internal Medicine, University of Michigan Medical School, Ann Arbor, Michigan 48109, USA
[+]Corresponding author. Tel: +44 (0)1707 625748; Fax: +44 (0)1707 625801;
E-mail: vincenzo.costanzo@cancer.org.uk
[++]Corresponding author. Tel: +1 646 888 2357; Fax: +1 646 422 0453;
E-mail: chaudhuj@mskcc.org
[+++]Corresponding author. Tel: +1 415 514 4309; Fax: +1 415 514 4242;
E-mail: jennifer.fung@ucsf.edu
[++++]Corresponding author. Tel: +1 734 615 0456; Fax: +1 734 763 3784;
E-mail: moranj@umich.edu

Submitted 21 May 2009; accepted 23 June 2009; published online 17 July 2009

Introduction

DSBs occur when both strands of a DNA double helix are damaged by either the normal metabolic processes of the cell, or environmental factors such as ultraviolet light or radiation. Cells have evolved mechanisms to repair DSBs, to ensure that large pieces of the genome are not lost; however, the aberrant repair of DSBs can lead to genome rearrangements that scramble the information encoded in the DNA. The 2009 Keystone Symposium on Genome Instability and DNA Repair featured mechanistic studies of DSB repair, host responses to DSBs and recent phenomenology in human genomic disorders. This report highlights some of the key findings presented at the meeting. Space limitations have prevented us from discussing every contribution, and we apologize to those whose work is not highlighted in this report.

DSBs: formation, repair and signalling events

One way to understand the consequences of aberrant DSB repair is to identify and characterize genomic rearrangements. M. Stratton (Cambridge, UK) used second-generation whole-genome DNA-sequencing technologies to characterize the most common types of DNA rearrangement in a range of cancer cell lines and primary tumours. In addition to finding a large number of point mutations in these samples, preliminary analyses of the data indicate that many rearrangements result from intrachromosomal events, and, less frequently, interchromosomal breakage and rejoining events. Importantly, many of these rearrangements had not been seen previously because they are too small to be detected by conventional karyotype analyses. The analysis of junction sequences by the Stratton group also revealed that classical repair processes such as HR and NHEJ, probably have a role in the formation of these rearrangements. However, he also observed a distinct pattern of insertion events, which involved nearby, unlinked or unknown segments of DNA that might result from non-classical DSB repair events such as single-strand annealing, template switching or break-induced replication. Therefore, the mutations and DNA rearrangements that occur in tumour cells are more varied and complex than previously thought.

Although intriguing, Stratton's findings are based on inference. Yet, understanding DSB repair necessitates learning more about

Glossary

A-complex	protein complex containing BRCA1 and NBA1, and other proteins
AID	activation-induced cytidine deaminase
ATM	ataxia telangiectasia mutated
BCL	B-cell leukaemia lymphoma
BRCA1	breast cancer 1
c-Myc	avian myelocytomatosis virus oncogene cellular homologue
CNV	copy-number variant
CpG	cytosine guanine
Crm-1	chromosome region-maintenance 1
CSR	class-switch recombination
DNA2	DNA nuclease 2
DSB	double-strand break
Dsbc1	double-strand break control 1
E2A	E2A immunoglobulin enhancer-binding factors E12/E47; also known as transcription factor 3 (TCF3)
EME1	essential meiotic endonuclease 1
ExoI	exonuclease I
GEN1	XPG-like endonuclease
HR	homologous recombination
IgH	immunoglobulin heavy-chain locus
I-Sce1	a rare cutting restriction endonuclease encoded by the *Saccharomyces cerevisiae* mitochondrial intron, omega
Ku70	70 kDa Ku antigen
Mms4	methyl methanesulphonate sensitive 4
Mre11	meiotic recombination 11
MRN	Mre11–Rad50–Nbs1 complex
Mus81	methyl methane sulphonate and ultraviolet sensitive 81
NBA1	new component of the BRCA1 A complex
NHEJ	non-homologous end joining
RAG1/2	recombination-activating gene 1/2
Rcr1	recombination regulator 1
SGS1	slow growth suppressor 1
Spo11	sporulation 11
V(D)J	variable-diversity-joining recombination
XRCC4	X-ray cross complementation 4
Yen1	yeast endonuclease 1

what happens to these lesions soon after they arise. Important contributions towards this goal came from L. Symington (New York, NY, USA) and G. Ira (Houston, TX, USA), who presented studies on the MRN complex, which has a 3'–5' exonuclease/endonuclease activity *in vitro* that seems to be incompatible with the proposed 5'–3' exonuclease activity that is needed to promote 3'-overhang formation *in vivo* (Mimitou & Symington, 2008; Zhu *et al* 2008). Symington and Ira demonstrated that this paradox might be resolved by the activity of the SGS1 helicase *in vivo*, which unwinds DNA and displaces single-stranded DNA in the 5'–3' direction. The single-stranded DNA is processed initially by the MRN complex and then by the DNA2 exonuclease/endonuclease, which acts redundantly with ExoI to degrade the 5' strand (Fig 1). Their findings seem to have resolved a longstanding question in DSB processing and explain how 3' overhangs are generated for use in HR-based DNA repair.

Another clue about the role of the MRN nuclease came from D. Ferguson (Ann Arbor, MI, USA). Ferguson used knock-in technology to create mice containing a nuclease-deficient allele of *Mre11* (Buis *et al*, 2008). Remarkably, these mice exhibited phenotypes that were indistinguishable from those of mice lacking the *Mre11* gene, including early embryonic lethality and genomic instability; however, in contrast to *Mre11*-knockout cells, ATM signalling is not affected in MRE11 nuclease-deficient cells. Therefore, unlike simple eukaryotes such as *Saccharomyces cerevisiae*, higher eukaryotes probably have a low tolerance for DSBs and use a range of recombination-based processes to repair these lesions rapidly in order to avoid cell death.

HR has been studied extensively and involves the formation of a Holliday junction (HJ) intermediate. The symmetrical cleavage of the HJ by an enzyme known as resolvase allows for the cleavage of the resultant duplexes (Liu & West, 2004). S. West (South Mimms, UK) reported the identification of the human (GEN1) and yeast (Yen1) HJ resolvases (Ip *et al*, 2008). Unlike previously identified nucleases—for example, Mus81–Mms4/EME1—GEN1 and Yen1 cleave model HJ substrates at symmetrical positions, yielding nicked duplexes that can be ligated without further processing. The identification of GEN1/Yen1 is a crucial breakthrough that will make its mark on many processes that involve HR-based repair.

Several talks also addressed how DSBs are formed during the normal cell cycle. Torsional stress can occur during DNA replication-fork elongation, especially when two replicons fuse together at termination or when the replication forks encounter transcription units. The topoisomerase-mediated resolution of these constraints then allows the completion of S phase, and subsequent chromosome condensation and segregation. However, interfering with this process can result in entangled DNA molecules and DNA breaks, leading to intra-S-phase DNA lesions and/or segregation abnormalities. Using genomic approaches, M. Foiani (Milan, Italy) characterized regions where replication termination occurs in *S. cerevisiae* and identified cellular pathways that are required for the resolution of two converging forks. Importantly, these replication-termination zones might represent replication-risk elements that are prone to genome rearrangements, unscheduled recombination events and even DNA breakage.

The efficient activation of the DNA-damage response might also be important for inducing chromatin modifications that promote an effective repair of DSBs. Following a DSB, ubiquitination of the damaged chromatin attracts BRCA1 complexes. How BRCA1 acts at the sites of DNA damage remains ill defined. The Ludwig laboratory (New York, NY, USA) showed that the ubiquitin ligase activity of BRCA1 is not essential for HR or the response to DNA-damaging agents, and therefore seems to be dispensable for DSB repair (Reid *et al*, 2008). However, it is clear that BRCA1 forms distinct subcomplexes. S. Elledge (Boston, MA, USA) reported the identification of a new component of the BRCA1 A-complex, NBA1, which is required for the recruitment of BRCA1 to sites of DNA damage. NBA1 also has a crucial role in ionizing-radiation resistance and cell-cycle control. Intriguingly, analyses of NBA1 and the other components of the BRCA1 A-complex revealed marked similarities to the lid complex of the 26S proteasome (Wang *et al*, 2009). Does the BRCA1 A-complex recruit the 26S proteasome to DSBs? If so, this groundbreaking finding is likely to shed light on the role of BRCA1 in DNA repair and cancer.

Chromosomal translocations are also a characteristic of many cancers. To identify proteins that affect translocation, M. Jasin (New York, NY, USA) used the rare endonuclease I-SceI to introduce DSBs at two different chromosomal loci in either wild-type mouse cells or mutants lacking proteins that are important for NHEJ—such as XRCC4 and Ku70 (Weinstock *et al*, 2007). Jasin reported a three- to fivefold increase in translocations in both *Xrcc4*-deficient and

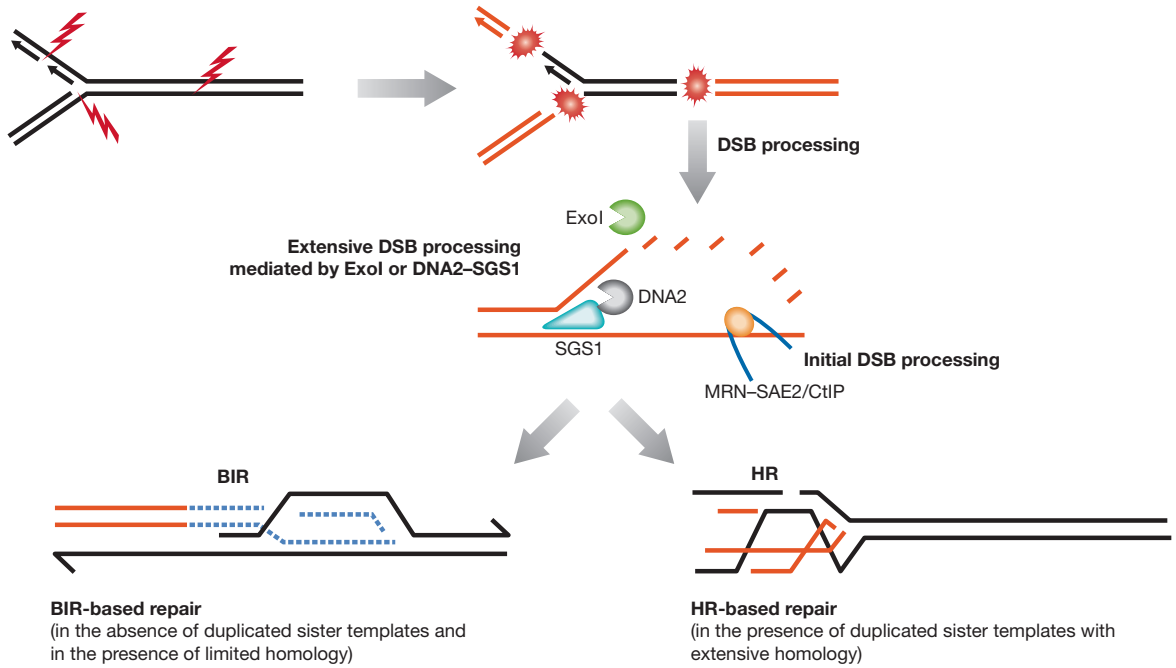


Fig 1 | Responses to DNA double-strand breaks in S phase. DSBs that arise during the S phase are rapidly processed by a helicase (SGS1) and exonucleases/endonucleases (DNA2, ExoI and MRN–SAE2/CtIP) to generate a 3' overhang. HR, which does not induce errors, or BIR, which is error prone, are used in higher eukaryotes to repair the lesion depending on the context in which the DSBs arise. BIR, break-induced replication; DNA2, DNA nuclease 2; DSB, double-strand break; ExoI, exonuclease I; HR, homologous recombination; MRN, Mre11–Rad50–Nbs1 complex; SAE2/CtIP, sporulation in the absence of Spo11/CTBP-interacting protein nuclease; SGS1, slow growth suppressor 1.

Ku70-deficient cells, and found that the translocation frequency could be suppressed to wild-type levels after the reintroduction of XRCC4 and Ku70, respectively, into the mutant cells. The sequence characteristics of the translocation junction—which include microhomology—were similar for the wild-type and mutant cell lines, pointing to the involvement of alternative pathways of NHEJ in translocation. Jasin also reported on a new technique that uses engineered zinc-finger nucleases to induce and quantify translocations in human cells without the need for cell cloning. By using this system, Jasin could determine that the aberrant repair of the DSBs leads to translocations—as occurs in mouse cells—approximately one-half of which show microhomology at the breakpoint junctions (Brunet *et al*, 2009). Therefore, this system can be used to analyse the role of DNA-repair proteins in human cancers.

DNA repair in lymphocytes

A characteristic of B lymphocytes is their ability to recognize a vast array of pathogens. This diversity is achieved, in part, by V(D)J recombination and CSR (Fig 2). Both processes proceed through DSB intermediates and are among the rare instances in which DNA lesions are deliberately introduced at defined genomic regions. The regulation of the enzymes that generate these lesions and of the proteins that repair the resultant DSBs were discussed in depth at this meeting.

Artemis is an endonuclease that cleaves hairpin intermediates at coding ends during V(D)J recombination (Sekiguchi & Ferguson, 2006). Inactive alleles of Artemis lead to severe combined immunodeficiency, whereas hypomorphic alleles of Artemis can lead to immunodeficiencies of varying severity (Revy *et al*, 2005). J. Sekiguchi (Ann Arbor, MI, USA) generated knock-in mice that phenocopy a premature-truncation mutation in the human Artemis protein. The mice had a severe reduction in B and T lymphocytes, and a pronounced accumulation of hairpin-coding ends. The mutant cells also exhibited DSB-repair defects, indicating that the carboxy terminus of Artemis is essential for V(D)J recombination and the maintenance of genomic stability (Huang *et al*, 2009).

CSR occurs between repetitive switch regions, which precede each immunoglobulin constant-region gene (Chaudhuri *et al*, 2007). The B cell-specific AID protein deaminates cytidines to uridines within transcribed switch regions, initiating a series of reactions that leads to the generation of the DSBs required for CSR (Muramatsu *et al*, 2007). Mutations in AID that block its nuclear export severely impair CSR, but do not affect cytidine deamination. To determine whether nucleocytoplasmic shuttling is required for CSR, M. Neuberger (Cambridge, UK) mutated the AID nuclear-export signal and/or replaced it with one from a heterologous protein (Geisberger *et al*, 2009). Many of the mutant proteins underwent nuclear export, but were severely impaired for CSR, which showed that export is necessary but not sufficient for CSR. However, some mutants that allowed nuclear export and CSR showed increased protein stability, which indicated that the AID nuclear-export signal might mediate interactions with other proteins—for example, exportin and/or Crm-1—to generate the high density of lesions at switch regions required for CSR.

Chromosomal translocations between the immunoglobulin loci and oncogenes are characteristics of B-cell lymphomas. To determine why some genes are targeted for translocations, M. Lieber (Los Angeles, CA, USA) assembled a database of breakpoints from the most

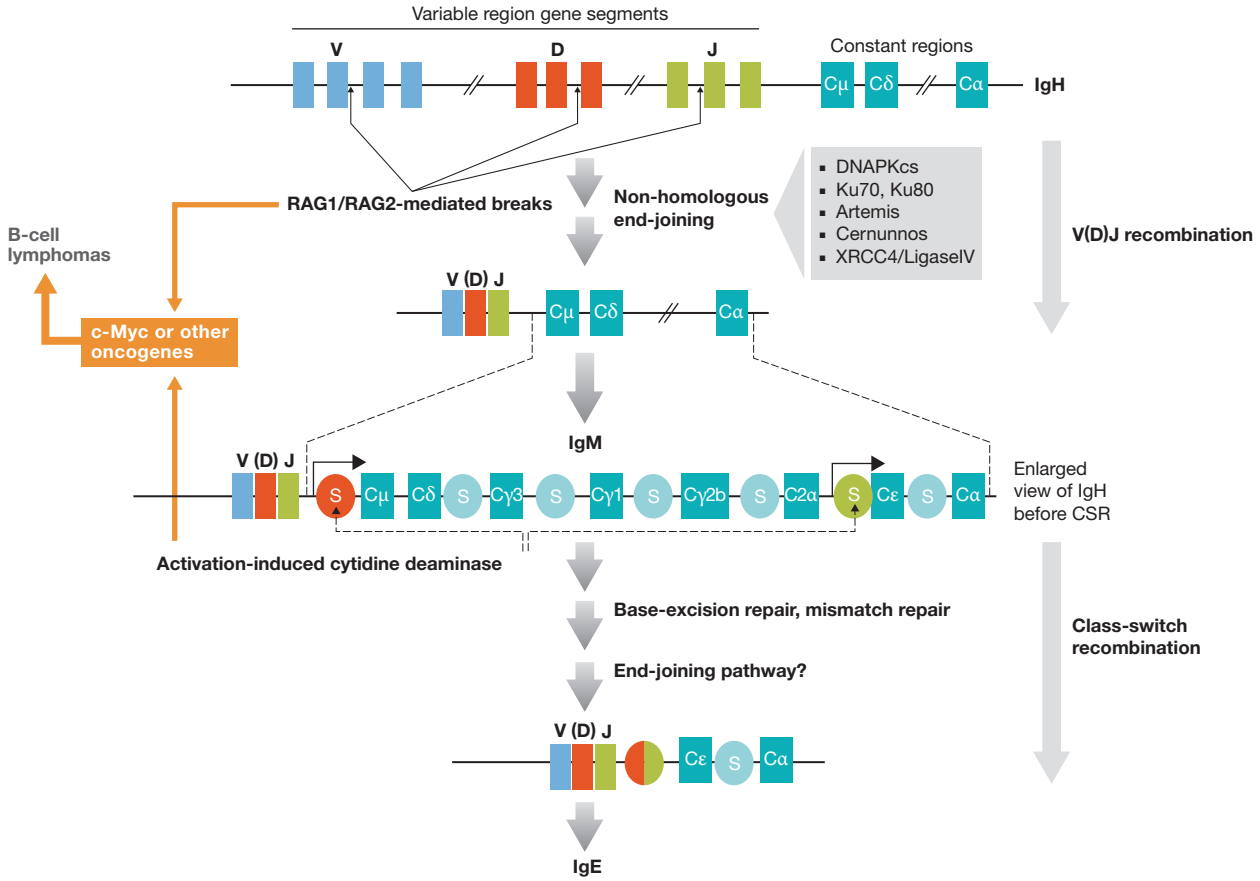


Fig 2 | DNA recombination in B cells. The V, D and J segments are assembled during V(D)J recombination (only the heavy chain locus is shown). The reaction is initiated by the RAG1/RAG2 endonuclease. Components of the NHEJ machinery ligate the resultant DSBs. The end product of V(D)J recombination is a B cell expressing IgM at its surface. Class-switch recombination occurs between repetitive switch (S) regions (ovals) that precede each constant-region gene. Each S region is an independent transcription unit, which is shown by the thick arrows. AID deamination at S regions followed by the activities of base-excision repair and/or mismatch repair lead to the formation of DSBs that subsequently are ligated by end joining. The factors that ligate DSBs at the S regions require elucidation. RAG1/RAG2 endonuclease and AID deaminase have been implicated in the ontogeny of B-cell lymphomas when their activities are aberrantly targeted to non-*Ig* genes such as *c-Myc* and other oncogenes. AID, activation-induced cytidine deaminase; c-Myc, avian myelocytomatosis virus oncogene cellular homologue; CSR, class-switch recombination; DNAPKcs, DNA-dependent protein kinase catalytic subunit; DSB, double-strand break; Ig, immunoglobulin; Ku70, 70 kDa Ku antigen; Ku80, 80 kDa Ku antigen; NHEJ, non-homologous end joining; RAG1/RAG2, recombination-activating genes 1 and 2; V(D)J, variable-diversity-joining recombination; XRCC4, X-ray cross complementation 4.

common chromosomal rearrangements (*bcl1*, *bcl2* and *E2A*) in human leukaemias and lymphomas. His analyses revealed that CpG nucleotides, which constitute only 1% of the human genome, account for 40–70% of the translocations in early-stage, but not late-stage, B-cell lymphomas (Tsai *et al*, 2008). These translocations probably arise owing to AID activity on methylated cytidines (Morgan *et al*, 2004), followed by RAG1/RAG2 activity at the mispaired bases.

The *c-Myc* oncogene is also a common translocation partner of the *IgH* in patients with Burkitt lymphomas and mice with plasmacytomas (Ramiro *et al*, 2007). To determine whether AID generates DSBs at *c-Myc*, D. Robbiani (New York, NY, USA) used I-SceI to introduce DSBs upstream of the *IgH*-switch region and/or the *c-Myc* locus. In the absence of I-SceI, all *c-Myc/IgH* translocations were AID dependent. By comparison, *c-Myc/IgH* translocations were observed in activated AID-deficient B cells only when I-SceI-induced DSBs were generated at both loci. Interestingly, Robbiani showed that the breaks at *c-Myc* are limiting under physiological conditions (Robbiani *et al*, 2008). In this context, F. Alt (Boston, MA, USA) found that *c-Myc* expression in the translocated *IgH/c-Myc* allele requires a transcriptional enhancer at the 3' end of the *IgH* locus.

The role of NHEJ in CSR remains ill defined. Both Alt and J-P de Villartay (Paris, France) showed that conditional inactivation of *Xrcc4* or *DNA ligase IV* in mice allows significant levels of CSR. K. Yu (East Lansing, MI, USA) arrived at a similar conclusion by studying a B-cell line in which *DNA ligase IV* was inactivated. In each study, most switch junctions showed DNA-sequence microhomology. Therefore, an alternative end-joining pathway is also active in CSR. Indeed, Alt found that *Xrcc4*-deficient splenic B cells accumulate AID-dependent DSBs at the *IgH* locus on chromosome 12 and RAG-dependent DSBs at the Ig light-chain

gene on chromosome 16, which could lead to chromosome 12/16 translocations by alternative end joining. Therefore, although it is not strictly required for CSR, classical NHEJ is important for maintaining genome stability during CSR.

Together, these talks reiterated that although targeted DNA alterations are essential for immune-system development, mistargeted RAG and/or AID activity could lead to the chromosomal translocations that are characteristic of B-cell lymphomas. Similarly, DNA-repair proteins that process the DSBs during lymphocyte differentiation have to be finely choreographed, as aberrant repair might have catastrophic consequences.

Meiotic control of recombination

During meiosis, recombination is initiated at DSBs that are created by the conserved Spo11 protein (Keeney & Neale, 2006). Repair of these DSBs by HR leads to either a reciprocal exchange of DNA between homologues (a crossover) or a non-reciprocal transfer of genetic information from one homologue to another (a non-crossover). Crossovers are important as they provide a physical link between homologues that ensures correct chromosome segregation. The regulation of meiotic recombination ensures that each pair of homologues receives at least one crossover event.

Several talks focused on genome-wide approaches to analyse the initiation and/or resolution of meiotic-recombination events. Previous work in several organisms showed that meiotic DSBs are not distributed randomly throughout the genome, but tend to cluster in 'hotspots'. What causes this non-random distribution remains poorly understood. S. Keeney (New York, NY, USA) described a high-resolution method to analyse global DSB distributions in *S. cerevisiae* using Spo11-associated oligonucleotide sequences. These experiments refine previous genome-wide analyses of meiotic DSBs, paving the way for a more detailed analysis of their regulation. Although significant overlap exists between the crossover and DSB maps, there are distinct differences, indicating that crossover placement is subject to additional levels of regulation. Phenomena that can potentially account for these differences include crossover interference or crossover homeostasis, as discussed by J. Fung (San Francisco, CA, USA), and interference between non-crossover and crossover events as presented by E. Mancera (Heidelberg, Germany). Interference imposes a more regular spacing of crossovers, whereas crossover homeostasis limits the number of crossovers to a tight fluctuation about the average.

Two talks highlighting recent work in mice suggested that *trans*-acting factors can influence crossover distribution. B. de Massy (Montpellier, France) and P. Petkov (Bar Harbor, ME, USA) reported the identification of overlapping regions of chromosome 17 that influence the activity of recombination hotspots elsewhere in the genome. These loci, *Dsbc1* (Grey *et al*, 2009) and *Rcr1* (Parvanov *et al*, 2009), neither act through a general activation or suppression of all hotspots, nor alter the overall number of crossovers; instead, they seem to affect the activity of specific hotpots differentially. In both cases, the loci were shown to affect the initiation of recombination before the crossover/non-crossover decision is made.

Copy-number variants in the human genome

Several presentations also focused on how CNVs have influenced the human genome. CNV is a broad term used to describe DNA rearrangements that arise from chromosomal duplications, deletions, inversions, translocations and mobile genetic-element insertions. J. Lupski (Houston, TX, USA) introduced the concept of genomic disorders and discussed how non-allelic recombination events between highly similar, but non-identical, low-copy number repeated DNA sequences can result in distinct genomic disorders (Lupski, 2009). He then proposed a fork-stalling and template-switching mechanism that might be important in generating non-recurrent DNA rearrangements in human cells (Lee *et al*, 2007). Interestingly, the junction sequences at many genomic rearrangements were composed of a patchwork of sequences derived from various intrachromosomal locations—a phenomenon that was also touched on in Stratton's talk. It will be interesting to see whether some mutations observed in tumours occur by a similar mechanism.

M. Hurles (Cambridge, UK) discussed how his colleagues at the Sanger Institute (Cambridge, UK), in Boston (MA, USA) and in Toronto (Canada) have amassed a near complete map of common CNVs in the human genome. The map comprises approximately 11,000 CNVs that are more than 1 kb in length and some generalizations are becoming evident. First, it seems that CNVs are relatively common and might arise by distinct molecular mechanisms—for example, non-allelic homologous recombination, fork stalling and template switching, and/or retrotransposition—that often leave signatures in the resulting alleles. Second, genome architecture might predispose certain regions of human DNA to these types of rearrangement.

The idea that sequence context might predispose certain chromosomal regions to rearrangements was addressed by B. Emanuel (Philadelphia, PA, USA), who reported that nearly perfect palindromic AT-rich DNA-repeat sequences on human chromosome 22q11 might form stem–loop and/or cruciform-like structures, predisposing them to disease-producing meiotic DNA translocations with other genomic regions such as 11q23 and 8q24.1. These translocation partners also contain AT-rich palindromes, providing support for the role of palindromic sequences in genomic instability. S. Lewis (Boston, MA, USA) discussed *in vitro* findings in yeast and mammalian cells, and showed that extended palindromic sequences could be extruded from DNA to form cruciform structures. The processing of these cruciforms in the cell could lead to DSBs, resulting in both subtle and large-scale genomic DNA rearrangements. Lewis also presented a model called palindrome revision, and suggested that subtle rearrangements at extruded palindromic sequences might act to 'disable' them before they can lead to more serious genome disruptions. It will be interesting to see whether small central deletions at other palindromic sequences in human DNA render them 'immune' to subsequent gross chromosomal rearrangements.

Concluding remarks

The 2009 Keystone Symposium on Genome Instability and DNA Repair provided an opportunity to present data, exchange ideas and spend time with like-minded colleagues. The meeting highlighted the many pathways used by cells to deal with genotoxic stress effectively, and how they are used to repair deliberate lesions introduced during the processes of immune-gene diversification and meiosis. Indeed, the knowledge obtained from *in vitro* experiments and *in vivo* studies in model organisms—only a few of which could be mentioned here—should be instrumental in determining how the complex genomic rearrangements observed in cancer and human genomic disorders arise. We predict that this will be a fertile area of research for years to come, and thank M. Jasin, S. Elledge and M. Neuberger for bringing us together to discuss cutting-edge issues on this topic.

ACKNOWLEDGEMENTS
V.C. is supported by Cancer Research UK, the European Research Council, the Lister Institute for Preventive Medicine and the European Molecular Biology Organization Young Investigator Programme. J.C. is supported by the Damon-Runyon Cancer Research Foundation, the Bressler Foundation and the National Institutes of Health (1R01AI072194-01). J.C.F. is supported by an American Cancer Research Scholar Award (RSG CCG 110,688). J.V.M. is supported by the Howard Hughes Medical Institute and the National Institutes of Health (GM60518).

CONFLICT OF INTEREST
The authors declare that they have no conflict of interest.

REFERENCES
Brunet E, Simsek D, Tomishima M, DeKelver R, Choi VM, Gregory P, Urnov F, Weinstock DM, Jasin M (2009) Chromosomal translocations induced at specified loci in human stem cells. *Proc Natl Acad Sci USA* **106:** 10620–10625

Buis J, Wu Y, Deng Y, Leddon J, Westfield G, Eckersdorff M, Sekiguchi JM, Chang S, Ferguson DO (2008) Mre11 nuclease activity has essential roles in DNA repair and genomic stability distinct from ATM activation. *Cell* **135:** 85–96

Chaudhuri J *et al* (2007) Evolution of the immunoglobulin heavy chain class switch recombination mechanism. *Adv Immunol* **94:** 157–214

Geisberger R, Rada C, Neuberger MS (2009) The stability of AID and its function in class-switching are critically sensitive to the identity of its nuclear-export sequence. *Proc Natl Acad Sci USA* **106:** 6736–6741

Grey C, Baudat F, de Massy B (2009) Genome-wide control of the distribution of meiotic recombination. *PLoS Biol* **7:** 327–339

Huang Y, Giblin W, Kubec M, Westfield G, St Charles J, Chadde L, Kraftson S, Sekiguchi J (2009) Impact of a hypomorphic Artemis disease allele on lymphocyte development, DNA end processing, and genome stability. *J Exp Med* **206:** 893–908

Ip SC, Rass U, Blanco MG, Flynn HR, Skehel JM, West SC (2008) Identification of Holliday junction resolvases from humans and yeast. *Nature* **456:** 357–361

Keeney S, Neale MJ (2006) Initiation of meiotic recombination by formation of DNA double-strand breaks: mechanism and regulation. *Biochem Soc Trans* **34:** 523–525

Lee JA, Carvalho CM, Lupski JR (2007) A DNA replication mechanism for generating nonrecurrent rearrangements associated with genomic disorders. *Cell* **131:** 1235–1247

Liu Y, West SC (2004) Happy Hollidays: 40th anniversary of the Holliday junction. *Nat Rev Mol Cell Biol* **5:** 937–944

Lupski JR (2009) Genomic disorders ten years on. *Genome Med* **1:** 42

Mimitou EP, Symington LS (2008) Sae2, Exo1 and Sgs1 collaborate in DNA double-strand break processing. *Nature* **455:** 770–774

Morgan HD, Dean W, Coker HA, Reik W, Petersen-Mahrt SK (2004) Activation-induced cytidine deaminase deaminates 5-methylcytosine in DNA and is expressed in pluripotent tissues: implications for epigenetic reprogramming. *J Biol Chem* **279:** 52353–52360

Muramatsu M, Nagaoka H, Shinkura R, Begum NA, Honjo T (2007) Discovery of activation-induced cytidine deaminase, the engraver of antibody memory. *Adv Immunol* **94:** 1–36

Parvanov ED, Ng SH, Petkov PM, Paigen K (2009) Trans-regulation of mouse meiotic recombination hotspots by Rcr1. *PLoS Biol* **7:** 340–349

Ramiro A, San-Martin BR, McBride K, Jankovic M, Barreto V, Nussenzweig A, Nussenzweig MC (2007) The role of activation-induced deaminase in antibody diversification and chromosome translocations. *Adv Immunol* **94:** 75–107

Reid LJ, Shakya R, Modi AP, Lokshin M, Cheng JT, Jasin M, Baer R, Ludwig T (2008) E3 ligase activity of BRCA1 is not essential for cell viability of homology-directed repair of double-strand DNA breaks. *Proc Natl Acad Sci USA* **105:** 20876–20881

Revy P, Buck D, le Deist F, de Villartay JP (2005) The repair of DNA damages/modifications during the maturation of the immune system: lessons from human primary immunodeficiency disorders and animal models. *Adv Immunol* **87:** 237–295

Robbiani DF *et al* (2008) AID is required for the chromosomal breaks in c-myc that lead to c-myc/IgH translocations. *Cell* **135:** 1028–1038

Sekiguchi JM, Ferguson DO (2006) DNA double-strand break repair: a relentless hunt uncovers new prey. *Cell* **124:** 260–262

Tsai AG, Lu H, Raghavan SC, Muschen M, Hsieh CL, Lieber MR (2008) Human chromosomal translocations at CpG sites and a theoretical basis for their lineage and stage specificity. *Cell* **135:** 1130–1142

Wang B, Hurov K, Hofmann K, Elledge SJ (2009) NBA1, a new player in the Brca1 A complex, is required for DNA damage resistance and checkpoint control. *Genes Dev* **23:** 729–739

Weinstock DM, Brunet E, Jasin M (2007) Formation of NHEJ-derived reciprocal chromosomal translocations does not require Ku70. *Nat Cell Biol* **9:** 978–981

Zhu Z, Chung WH, Shim EY, Lee SE, Ira G (2008) Sgs1 helicase and two nucleases DNA2 and Exo1 resect DNA double-strand break-induced gene conversion. *Cell* **134:** 981–994

Vincenzo Costanzo (top left), Jayanta Chaudhuri (top right), Jennifer C. Fung (bottom left) & John V. Moran (bottom right)

