Our group aims at identifying and characterizing the biological processes that maintain genomic integrity and ensure the faithful transmission of genetic information during reproduction, as well as endogenous and exogenous events that enhance genome instability. We are focusing our work on two biological situations in which DNA double-strand breaks (DSBs) form in the budding yeast *Saccharomyces cerevisiae*.

First, we study the exchange of genetic material by recombination which occurs between the parental chromosomes during yeast meiosis. Using genome wide molecular methods, we were able to show that each yeast chromosome has a unique map of meiotic DSBs with alternating ‘hot’ and ‘cold’ domains where recombination occurs more or less frequently, and correlate with enriched region of histone H3-K4 trimethylation, (Figure 1). We recently identified the Spp1 protein, a member of the COMPASS complex, as linking histone methylation to the Mer2 protein, a key protein of the differentiated chromosomal axis required for DSB formation.

Also, we have developed a Gal4-Spo11 fusion protein, which allows us to modify the usual DNA cleavage sites along the chromosomes and diversified this method to target different region of the yeast genome. This method to modify meiotic recombination profiles has been licensed to MEIOGENIX to be tested in plants and mice.
Second, we study the mechanisms of genome instability and mutagenesis. We established the mutational landscape of several yeast mutator strains using next generation sequencing and bioinformatics methods to identify the variants. We also study the instability of human tandem repeated DNA sequences (minisatellites) inserted in the yeast genome.

In the *S. cerevisiae* genome, as in the human genome, tandem repeated minisatellite DNA sequences are unstable during meiosis when they may undergo expansion and/or contraction of the number of tandem repeats. To investigate the mechanism(s) underlying tandem-repeat instability, we introduced two human minisatellite CEB1 and CEB25 alleles into the *S. cerevisiae* genome. We found that deletion of the RAD27/FEN1 gene, which is involved in DNA replication and repair, causes a high level of instability of the CEB1-1.8 allele in cells growing mitotically, indicating that replication defects destabilise these repeated sequences. It gives rise to a large variety of length variants due to expansion and contraction of the repeat units. We have also found that the mitotic stability of CEB1 depends on the activity of the Pif1 helicase. *In vitro* and in vivo analyses showed that CEB1 repeats formed stable G-quadruplex (G4) secondary structures and that the Pif1 protein unwinds these structures efficiently. This was further confirmed by using the PhenDCs G4-quadruplexes ligands developed by the group of M-P. Teulade-Fichou (Institut Curie, Orsay); These molecules specifically destabilized CEB1 in wild-type treated cells and yielded CEB1 rearrangements similar to that in *pif1D* cells. Mechanistically, the instability of CEB1 occurs during leading-strand replication. The steps leading to CEB1 rearrangements are illustrated in Figure 2. The formation of CEB1 G-quadruplexes also stimulates gross chromosomal rearrangements.

We currently pursue a structure-function analysis of the CEB1 and CEB25 G-quadruplexes upon mutagenesis of the guanine tracts and loop regions. We found that short loops leads to more thermodynamically stable G-quadruplex, in correlation with their higher instability in vivo.
Key publications

Year of publication 2015

Aurèle Piazza, Michael Adrian, Frédéric Samazan, Brahim Heddi, Florian Hamon, Alexandre Serero, Judith Lopes, Marie-Paule Teulade-Fichou, Anh Tuân Phan, Alain Nicolas (2015 May 10)

Short loop length and high thermal stability determine genomic instability induced by G-quadruplex-forming minisatellites.
The EMBO journal : 34 : 1718-1734 : DOI : 10.15252/embj.201490702

Year of publication 2014

Michael Adrian, Ding Jie Ang, Christopher J Lech, Brahim Heddi, Alain Nicolas, Anh Tuân Phan (2014 Apr 17)

Structure and conformational dynamics of a stacked dimeric G-quadruplex formed by the human CEB1 minisatellite.
Journal of the American Chemical Society : 6297-305 : DOI : 10.1021/ja4125274

Alexandre Serero, Claire Jubin, Sophie Loeillet, Patricia Legoix-Né, Alain G Nicolas (2014 Jan 21)

Mutational landscape of yeast mutator strains.
Proceedings of the National Academy of Sciences of the United States of America : 1897-902 : DOI : 10.1073/pnas.1314423111

Year of publication 2012

Laurent Acquaviva, Lóránt Székvolgyi, Bernhard Dichtl, Beatriz Solange Dichtl, Christophe de La Roche Saint André, Alain Nicolas, Vincent Géli (2012 Nov 15)

The COMPASS subunit Spp1 links histone methylation to initiation of meiotic recombination.
Science (New York, N.Y.) : 215-8 : DOI : 10.1126/science.1225739

Aurèle Piazza, Alexandre Serero, Jean-Baptiste Boulé, Patricia Legoix-Né, Judith Lopes, Alain Nicolas (2012 Jun 7)

Stimulation of gross chromosomal rearrangements by the human CEB1 and CEB25 minisatellites in Saccharomyces cerevisiae depends on G-quadruplexes or Cdc13.
PLoS genetics : e1003033 : DOI : 10.1371/journal.pgen.1003033

Year of publication 2011

Judith Lopes, Aurèle Piazza, Rodrigo Bermejo, Barry Kriegsman, Arianna Colosio, Marie-Paule
Teulade-Fichou, Marco Foiani, Alain Nicolas (2011 Jun 28)

**G-quadruplex-induced instability during leading-strand replication.**

*The EMBO journal*: 4033-46 : [DOI: 10.1038/emboj.2011.316]