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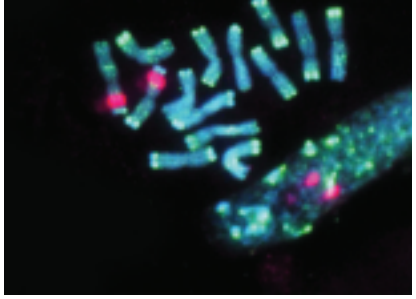
E.C.A. on Facebook

E.C.A. is now also on Social Media! For the present we are active on Facebook, but Instagram and Twitter may follow soon.
Each week you will find announcements of interesting articles, related to cytogenomics or to biology in general, and also pictures and stories from social events related to E.C.A. and its members. Also our E.C.A. conferences will be covered on Social Media.
You can see the weekly posts and announcements via the direct link

https://www.facebook.com/Cytogenetic/ or on the updated E.C.A. website **http://www.e-c-a.eu/**

Please contact us (mariano.rocchi@uniba.it; k.madan@lumc.nl) if you wish to share an interesting news item or a pertinent article.

The Board of the E.C.A. takes this opportunity to wish you all a happy and successful 2020.

European Cytogeneticists Association

12th EUROPEAN CYTOGENOMICS CONFERENCE

6 - 9 JULY 2019

SALZBURG CONGRESS
SALZBURG, AUSTRIA

www.eca2019.com

12th EUROPEAN CYTOGENENOMICS CONFERENCE SALZBURG 2019

Saturday, 6 July 2019

Permanent Working Group (PWG) Reports

PWG: CYTOGENOMICS.

Co-ordinators:
Joris Vermeesch (B)
Anna Lindstrand (S)

The focus of the working group on cytogenomics was structural variation analysis following whole genome sequencing. This is currently a major challenge and likely will be for the coming years. We focused on three topics, callers, databases and visualization.

The first speaker was **Nicole de Leeuw** who presented "Tools for structural variant detection from WGS data. She submitted an overview of different callers and their performance. The bioinformatics team in Radboud University Medical Center has evaluated numerous callers for the detection of structural variation (SV), including deletions, insertions, duplications, inversions and translocations, in the human genome. Four of these were selected for further SV testing using discordant pair-end reads and split reads: Pindel, Delly, Manta and Lumpy. These four callers were compared with each other by using the pilot genome from Genome In A Bottle (GIAB), looking at sensitivity, specificity and running time. Manta yielded the best test results and is currently used together with Control-FREEC for CNV calling and ExpansionHunter for the detection of short tandem repeats.

The second speaker was **Jesper Eisfeldt** who presented "Structural variation databases based on whole genome sequencing data". Jesper presented an overview of gnomAD-SV and SweGen-SVDB, two public structural variant frequency databases, as well as strategies for building local (private) structural variant databases. These databases were compared based on the number of filtered common variants in a cohort of 100 individuals, illustrating that local databases are necessary for filtering local technical artefacts as well as population specific variation.

The third speaker was **Johanna Lundin** who presented "VCF2Cytosure", a new tool for visualization of structural variants developed by researchers at the Karolinska Institute together with the Dept of Clinical Genetics at the Karolinska University Hospital. This tool converts VCF-files from whole genome sequencing into CGH-files suitable for uploading in the Cytosure Interpret software (Oxford Gene Technologies). In this software the structural variants are visualized in the same way as CGH data and can be analyzed and classified in a very user-friendly way suitable for a clinical setting.

PWG: CYTOGENETICS OF HAEMATOLOGICAL MALIGNANCIES.

Co-ordinators:
Bertil Johansson (S)
Harald Rieder (D)

About 50 participants joined the PWG meeting.

The meeting started with the presentation of an abstract entitled "A unique peripheral blood karyotype characterized by multiple complex chromosomal translocations is caused by homozygosity for the CHEK2 p.Gly167Arg variant" which was given by **Nivin Moustafa** (IL). She reported about two patients who were unrelated and were homozygous for the CHEK2 c.499G>A; p.Gly167Arg variant, located in the central forkhead-associated (FHA) domain of the protein. One patient presented with primary

multi-organ tumorigenesis including dozens of intestinal polyps since age 35 years, thymoma at age 49 years, breast cancer (BC) at 65 years, prostate cancer at 66 years, left renal cell carcinoma, angiomyolipoma of the right kidney and sigmoid gastrointestinal stromal tumor at age 67 years. Karyotyping on peripheral blood lymphocytes revealed multiple different non-clonal chromosomal translocations involving various chromosomes in 40%–60% of cells, which were mostly unbalanced. The patient's bone marrow and fibroblasts were normal and did not display the translocation phenotype. The second patient had early-onset acute myeloid leukemia, which was characterized by a clonal karyotype including a t(3;8)(q26;q24) in the bone marrow cells. The chromosome analysis of peripheral blood cells showed different non-clonal complex chromosome aberrations in about 30% of the cells. It was discussed that the homozygosity for p.Gly167Arg in CHEK2 increases the patients' susceptibility to DNA double strand breaks (DSB) and shifts the balance to non-accurate DSB correction. This possibly explains the increased susceptibility of homozygotes to either multiple primary tumours during their lifetime or early-onset tumorigenesis.

Oskar Haas (A) presented the paper entitled "Hyperdiploid acute lymphoblastic leukemia with genome-wide copy-neutral loss of homozygosity (CN-LOH)". He addressed a distinct subset of hyperdiploid ALL in children, which is characterized by pure tetrasomies. Such cases show a CN-LOH of all disomic as well as the duplication of both homologs of all tetrasomic chromosomes. It is generally believed that this distinct pattern can only derive from the duplication of a preexistent analogous hyper-haploid clone. As apparent indicator of poor prognosis, such hyperhaploid/-diploid ALL forms are nowadays stratified as high risk in ongoing treatment protocols. Oskar Haas showed, that hyperhaploid and analogous hyperdiploid forms cannot be distinguished based on array patterns alone. Therefore, he considered it essential and mandatory to use cytogenetic, FISH and/or DNA index analyses in addition to microarray investigations to clearly assess the pattern of the chromosomal gains and losses more accurately.

Harald Rieder (G) presented the abstract "Interactive online karyotyping to teach cyto-genetics in undergraduate medical education". The online karyotyping tool was available for the participants of the PWG meeting by using their own device. Chromosomes could be dragged by mouse or pencil to complete a karyogram. If a karyogram was completed it was submitted to the system to check for mistakes. In case of mistakes the system rejected the karyogram until it was corrected. After the completion of the karyogram, a karyotype according to the ISCN had to be provided which again was checked by the system. A total of seven different chromosome aberrations were included in the online tool and successfully used for teaching cytogenetics in undergraduate medical education. The meeting closed with several participants staying on to continue with online karyotyping.

PWG: CANCER CYTOGENOMICS, SOLID TUMOR STUDIES.

Co-ordinators:
Roberta Vanni (I)
David Gisselsson-Nord (S)

The Permanent Working Group (PWG) on "Cancer Cytogenetics, solid tumor studies" met in Salzburg on July 6, 2019 during the 12th European Cytogenomics Conference.

The aim of the meeting was to define the "State of the Art Seminar on Solid Tumor Cyto-genomics".

The Co- coordinator **Roberta Vanni** started the session by thanking the presenters of the selected abstracts - who had enthusiastically accepted to share their results with the community - and gave an overview of the development of the research in solid tumor cytogenetics. The emerging evolution of this field in the last years has paralleled the tremendous evolution of new molecular techniques, most prominently the next generation (massively parallel) sequencing. This trend was reflected in part by the abstracts

presented during the PWG. In line with this trend and the new title of the European ECA Conference (European Cytogenomics Conference), the participants agreed with the proposal to change the title of the Permanent Working Group to "Cancer Cytogenomics, solid tumor Studies".

The co-coordinator **David Gisselsson** introduced the speakers and moderated the discussion.

The first speaker, **Dr. Alla S Koltsova** from St. Petersburg State University, reported a study on cytogenetic abnormalities in uterine leiomyoma cells in which the frequency of karyotypically abnormal clones *in vivo* and *in vitro* was compared. The authors observed a statistically significant difference in frequency of abnormal cells between cultured and non-cultured samples and suggested that some of the abnormal clones (but not all) have a selective growth, in contrast to cells with chromothripsis which were less represented in cultured cells.

The second speaker, **Dr. Alvin Soon Tiong Lim**, from the Singapore General Hospital, Molecular Pathology, reported on the diagnostic role of MAML2 gene rearrangements, disclosed by FISH, in mucoepidermoid carcinoma (MEC) and highlighted that MAML2 testing is needed for tumors deviating from the conventional appearance of MEC and those resembling other tumor types.

The third speaker, **Dr. Aakila Sammy**, from the Brunel University London, discussed the results of a research on ovarian cancer analytic cytogenomics. Her group had investigated mis-localization and re-organization of specific ovarian cancer-related chromosomes and genes by analyzing nuclear motor myosin, which has a role in the adaptation of chromosome territories under different physiological conditions. Knocking down the protein, they compared the response of ovarian cancer cells, healthy ovarian cells and cells characterized by drug resistance. They concluded that genome organization represents an exploitable mechanism in which the protein may play a significant diagnostic and prognostic role.

Last but not least, **Dr. Eric Jeandidier**, of the Groupe Hospitalier de la Région de Mulhouse et Sud-Alsace Service de Génétique Mulhouse-France, reported on how they improved the detection of dicentric chromosomes and telomere dysfunction, the driving forces of chromosomal instability. Their approach consisted of sequential analysis of telomeres and centromeres using FISH followed by the M-FISH technique. This allowed the assessment of the potential role of telomere dysfunction and chromosomal instability in order to improve initial treatment strategy on an individual basis.

PWG: QUALITY ISSUES AND TRAINING IN CYTOGENETICS.

Co-ordinators:
Ros Hastings (UK),
MartineDoco-Fenzy (F)
Marta Rodríguez de Alba (E)

On behalf of the PWG on Quality and Training, Dr Ros Hastings presented a workshop on ISCN. The talk started by giving some of the common misconceptions, then detailing some of the basic ISCN rules before giving examples of frequent errors seen in the GenQA External Quality Assessments (EQA). Finally there was an interactive ISCN quiz for everyone to participate in.

Ros represents Europe on the Standing Committee for ISCN and at a recent committee meeting it was decided there would be a major revision with a new ISCN being published in 2020. The committee reviewed 222 suggestions for improvement and agreed to include some of the suggestions. The committee agreed there was a need for some triple-colour FISH examples plus more examples were needed in the rsa and sequence chapters. In addition, it was proposed that the chapter on comparative genomic hybridisation (13.6) could be removed and that section 8.4 on UPD should be moved to the array

chapter. Finally, it was proposed that a new chapter for Polar Body ISCN to describe haploid sets should be included.
It is also proposed that the new ISCN 2020 will include the following changes:

- A summary of the basic ISCN rules will be given at front of book.
- Sex chromosomes will be reported first for all techniques.
- Abnormalities will be described from pter to qter for all techniques so that the G-banded nomenclature is consistent with the 'arr' and 'seq' nomenclature.
- Nucleotides can be separated by commas (but not full stops) for arrays, rsa and sequence nomenclature.
- When an unbalanced derivative is inherited from a balanced rearrangement in the parent 'dmat' and 'dpat' will be used. The previous use of mat or pat for an unbalanced derivative is ambiguous as could also imply the parent is also unbalanced.
- Centres will have to specify which HGVS version is being used for 'seq' nomenclature.
- More p-arm examples will be given.

A pdf copy of the talk can be found on the GenQA website for any interested genetics centres on https://www.genqa.org/sites/default/files/PWG%20-%20ECA%202019.pdf
Dr Ros Hastings has now stepped down from this PWG as she has retired and 'returned' to work part time with GenQA. Thanks to all the ECA members who have supported this PWG at the ECA conferences over the years. I hope you will continue to do so under the able leadership of Dr Martine Doco-Fenzy who will now take the lead on this PWG.

Ros Hastings
Outgoing Chair of the PWG for Quality and Training.

The Board of the E.C.A. is grateful to Ros Hastings for her excellent work as a coordinator of this PWG for many years.

PWG: MARKER CHROMOSOMES

Coordinators:
Thomas Liehr (D)
Isabel Marques Carreira (P)

As is customary, the permanent working group meeting, PWG Marker Chromosomes was held on the first day of the 12th ECA-conference 2019 in Salzburg, Austria. This was the 7th meeting of the PWG. The session was again well appreciated and attended by >150 cytogeneticists from all over Europe.
Thomas Liehr (Jena, Germany) briefly introduced the topic of this specific PWG and outlined the programme. Five speakers, who had been selected from the abstracts submitted to the conference, gave a 5-8 minute presentation on small supernumerary marker chromosomes (sSMC) identified during routine diagnostics. **Wafa Slimani** presented 8 out of 33 sSMC cases characterized in her lab in Sousse (Tunisia) during the past 8 years: six cases of sSMC(15) and 2 cases with partial tri- or tetrasomy 9p; these were also discussed in the context of available literature. **Nadezda Shilova** talked about a man with the karyotype 47,XY,+invdup(22)(p11.2) 10]/ 46,XY [20] on whom extensive studies on meiotic segregation of this sSMC had been performed in their Moscow lab (Russia). As a result, they could show that the presence of this sSMC had no influence on the rate of aneuploidy in the sperm of this healthy sSMC carrier. The next presentation, by **Jadranka Vraneković** (Rijeka, Croatia), showed the difficulties of diagnosing a prenatal case of a Pallister-Killian syndrome with a unique feature, a bifid cardiac apex which has never been described before. **Yvonne Stratis** (Mainz, Germany) reported the first sSMC with a neocentromere resulting from a complex chromosomal rearrangement involving an insertion of chromosome-4 material into a chromosome 12, and a deletion of a part of chromosome 12. The deleted segment 12q2?3~q24.33 formed an sSMC. **Paolo Reho** (Florence, Italy) showed how low coverage whole genome sequencing in plasma-derived circulating cell free DNA can be used to detect low levels of

sSMC(X) or sSMC(Y) in cases of Turner syndrome, who were previously thought to have a karyotype 45,X in all cells.
Finally, **Thomas Liehr** (Jena, Germany) reported on the next project to be done in close cooperation with FACE2GENE and on, thanks to a financial support of NORD foundation, the establishment of computer-assisted recognition of patients with cat-eye-syndrome. This has been already implementted and published for Pallister-Killian and for Emanuel syndromes (PMID: 28661575). Just like the other speakers in this session, Dr. Liehr also emphasized the irreplaceable impact of (molecular) cytogenetics in a world of array-CGH and NGS; he referred to a recently published paper of his group (PMID: 30089300) in which it was shown that >80% of sSMC carriers would be missed if cytogenetics would be skipped and be replaced by array-CGH in the diagnosis of infertility. Finally, problems with maintenance and updating of the well-appreciated sSMC page (http://ssmc-tl.com/Start. html) and its sister pages (http://ssmc-tl.com/Start. html) were outlined. These problems will mean that sooner or later in 2019/2020 this page can no longer be accessed via that link. Just in case this issue cannot be solved, the page is already now available on http://molbiol.sci.am/ssmc/ssmc-tl.com/Start. html. Dr. Liehr is working on a new version and will announce where it can be found - this will be possible via the last mentioned link or via http://markerchromosomes.ag.vu, http://markerchromosomes.wg.am or https://www.uniklinikum-jena.de/humangenetik/en/Databases.html.

Overall, the meeting showed that there is a broad spectrum of different sSMCs, which can be detected in our routine pre- and postnatal studies. Thanks to all speakers for giving their excellent presentations, and it was really a pity that **Isabel Marques Carreira** (Coimbra, Portugal) could not come in time to the meeting due to travel problems!

PWG: PRENATAL DIAGNOSIS.

Co-ordinators:
Maria Rosario Pinto Leite (P)
Jean-Michel Dupont (F)

A meeting of the permanent working group on Prenatal Diagnosis was held on 6 July during the 12th European Cytogenetic Conference 2019 in Salzburg. Approximately 80 participants attended the session.
There were three presentations, including one last minute abstract on Uniparental Disomy risk estimate.
Rosário Pinto Leite and **Jean Michel Dupont** challenged the audience to take part in an innovating and dynamic experiment during the presentation: answering in real time a survey (quiz) about practices in CVS in Europe. The answers provided to each question were then compared with previous responses given by colleagues from several countries, including several laboratories in Portugal. This survey, of which results will be available later on, gives a first glimpse of the heterogeneous practices all over Europe with regard to CVS handling.
The presentation by **Aurélie Coussement**, from the Laboratoire de Cytogénétique, Cochin hospital in Paris, was titled “Back for the future - Lessons from the past for an updated management of the trophoblast”. It focused on the management of chorionic villus samples for chromosome analysis, starting in the 80s where only direct analysis was available from the cyto-trophoblast, to the presentday protocols including arrayCGH. She presented the pros and cons of the successive strategies used and showed that the association of direct analysis on cytotrophoblast associated with either FISH on mesenchymal cells (in case of aneuploidy) or array CGH in case of normal result give the best compromise between time to get a result and sensitivity.
Kamran Moradkhani, from Service de Génétique, CHU Nantes, presented “Risk estimation of uniparental disomy of chromosome 14 or 15 in a fetus with a parent carrying a non-homologous Robertsonian translocation”. In accordance with the results of this study, the authors do not

recommend prenatal testing for UPD for pregnancies when one of the parents is known to carry a nonhomologous ROB involving chromosome 14 and/or 15. A total of 1747 UPD testing were performed on fetuses during pregnancy for the presence of UPD(14) and/or UPD(15); the risk of UPD following prenatal diagnosis was estimated to be around 0.06 %, less than the risk of miscarriage following an invasive prenatal sampling. This study was published in July 2019 in Prenatal diagnosis (https://doi.org/10.1002/pd.5518)

PWG: CYTOGENETIC TOXICOLOGY AND MUTAGENESIS.

Co-ordinators:
José M. Garcia-Sagredo (E),
Emanuela VOLPI (UK)

The meeting of the Cytogenetic Toxicology and Mutagenesis PWG was held in the Salzburg Conference Centre on the first day of the 12th ECA Conference. The session was opened and chaired by the PWG Coordinators, **Emanuela Volpi** (University of Westminster, London) and **José Garcia-Sagredo** (Alcala' University). The central theme for this year's meeting was *the evaluation of chromosomal instability applied to diagnosis, prognosis and treatment of disease*.

from left to right: Jose Garcia-Sagredo, Ulrike Mau-Holtzmann, Emanuela Volpi, Isadora May Vaz, Radhia M'kacher and Eric Jeandidier.

The first two invited talks focused on chromosomal instability in cultured stem cells and its relevance in connection to the safe use of these cells for clinical applications.

The first speaker - **Isadora May Vaz** from the Pontifícia Universidade Católica do Paraná in Curitiba in Brazil - showed how the reprogramming and *in vitro* culture of induced pluripotent stem cells (iPSC) from karyotypically healthy mesenchymal cells can lead to a completely altered lineage through the emergence of clonal cytogenetic changes (Abstract 1131).

In line with the first talk, in her presentation titled 'Quality control: CHECK YOUR CULTURES! Karyotyping identifies genetic instability in iPSC' (Abstract 1157), the second speaker – **Ulrike A. Mau-Holzmann** from the University of Tuebingen in Germany – emphasized 'chromosomal instability' and the resulting gain of chromosomal aberrations during reprogramming, transdifferentiation or gene-editing as common features during iPSC culturing, and advocated periodical karyotyping as an effective quality control measure. A lively discussion ensued leading to a consensus on the necessity to develop means to increase awareness of this issue with the relevant communities of stem cells practitioners.

The next two talks were delivered by two collaborating colleagues from France, **Radhia M'kacher** (Cell Environment DNA damage R&D, Paris) and **Eric Jeandidier** (Groupe Hospitalier de la Région de Mulhouse et Sud-Alsace). Eric (Abstract 1202) presented development of a protocol based on centromere and telomere staining followed by M-FISH for the detection of dicentric chromosomes and the assessment of telomere dysfunction in connection with chromosomal instability in patients with haematopoietic malignancies. Radhia (Abstract 1200) reported their interesting results on the identification of telomere dysfunction and pericentromeric breakpoints in patients undergoing fertility treatment suggesting telomere status as a potential novel biomarker for the prediction of outcome in assisted reproduction.

The final talk was given by **Ivan Iourov** (Mental Health Research Centre, Moscow, Russia). Ivan presented the most recent data by Yurov's team on their longstanding project on somatic chromosomal mosaicism and instability in neurodevelopmental diseases (Abstract 1097), with novel empirical evidence, which is bound to generate renewed interest in this fascinating and challenging research area.

It has been an honor and a pleasure to organize this PWG meeting and to have the opportunity to meet talented, like-minded colleagues from different parts of the world to discuss research topics of common interest. We very much look forward to the next PWG meeting.

PWG: CLINICAL AND MOLECULAR APPROACHES TO CYTOGENETIC SYNDROMES

Co-ordinators:
Conny van Ravenswaaij (NL),
Cristina Skrypnyk (BRN),
Nicole de Leeuw (NL)

During this one-hour PWG meeting, **Nicole de Leeuw** started with an update on ECARUCA and the transition to DECIPHER to enable the continued sharing of the gathered content of the ECARUCA database, which contains detailed, curated clinical and molecular information from more than 5,000 patients with a rare, unbalanced chromosome aberration. A total of 3,646 patients with 4,365 genomic imbalances qualify to be submitted to DECIPHER. ECARUCA account holders who submitted cases in the past 15 years will be contacted to obtain adequate consent for submission in DECIPHER.

Next, selected abstracts from three ECA participants were presented, the first two focussing on the 3p26.3 region. Both copy number gains and losses in the 3p26.3 region have been associated with neurodevelopmental disorders. **Andreea Cristina Stanciu** from the University of Medicine and Pharmacy in Bucharest, Romania, gave a presentation entitled "The first case of 3p26.3 deletion containing only *CHL1* gene associated with ASD". She gave a brief summary on the 3p deletion syndrome (OMIM #613792) which is a rare contiguous gene disorder caused by deletions in the distal 3pter region. It is characterized by psychomotor retardation, developmental delay, dysmorphisms, microcephaly and ptosis. Only four well documented cases have been reported so far, each with a terminal deletion in 3p26.3 (500 kb - 1.1 Mb) encompassing only the *CHL1* gene. Three of these were inherited from an apparently unaffected parent. *CHL1* is highly expressed in the central and peripheral nervous systems and copy number variants involving this gene have been considered causative for impaired cognitive function. Andreea presented a 12-year-old boy with mild intellectual disability, speech delay, motor delay and hyperkinesia in whom array-CGH analysis disclosed an interstitial loss of 12 kb in 3p26.3 causing a partial deletion of the *CHL1* gene. Unfortunately, carrier testing in the parents was not (yet) performed.

Next, **Igor Lebedev** from the Research Institute of Medical Genetics in Tomsk, Russia, presented their work on "*CNTN6* expression in human IPSC-derived neurons from a patient with neurodevelopmental disorder and 3p26.3 microduplication and the same microduplication healthy carrier". This duplication was previously characterised by whole genome sequencing, which did not reveal any structural variations either within the *CNTN6* gene or its flanking regions. They next obtained cell lines of induced pluripotent stem cells (iPSC) from cells of the patient and his unaffected father carrying the same duplication (of maternal origin in the father). The iPSCs were differentiated in vitro into cortical neurons. The level of *CNTN6* gene expression in patient iPSCs was significantly lower than in the neurons from two healthy donors (without the duplication). The level of *CNTN6* expression in the carrier father iPSCs was comparable to that in neurons from the healthy donors. Allele-specific analysis of *CNTN6* expression revealed slightly preferred expression of the maternal allele in control iPSCs. This preference was more pronounced in

the father carrying a maternal duplication. The expression of the duplicated allele of paternal origin in the patient was significantly weaker than a normal one which caused an even more profound difference between maternal and paternal allele expression of *CNTN6*. The significant reduction of the *CNTN6* expression in neurons obtained from patient iPSCs can explain the similarity of the symptoms observed in patients with either a deletion or a duplication of the *CNTN6* gene.

Ana Sousa (CHULN-HSM, EPE Medical Genetics Department in Lisbon, Portugal) was the last speaker in this meeting and she talked about "*STAG1* haploinsufficiency: an emerging phenotype". She presented data on a 4-year-old boy with mild developmental delay, arched and sparse eyebrows, down-slanting palpebral fissures, a wide nose with short columella, and thick lips. An intragenic deletion of 206 kb, involving exons 2 to 12 of the *STAG1* gene (arr[GRCh 37]3q22.3(136184662_136390897)x1), was detected by array CGH analysis. *STAG1* encodes one of the components of the cohesion complex which is involved in chromosome segregation and gene transcriptional regulation. *STAG1* is one of eight genes participating in the cohesion pathway. A dysfunction in this pathway my lead to one of the cohesinopathies, which are rare neurodevelopmental disorders characterized by distinctive facial dysmorphism, growth retardation, developmental delay/intellectual disability (DD/ID), and limb abnormalities. So far, only 15 patients with a (partial) loss of *STAG1* or a pathogenic nucleotide variant in *STAG1* have been reported in the literature. The patient presented here shares common clinical features with these published patients and these features include DD/ID, ranging from mild to severe, and nonspecific facial dysmorphisms. No clear phenotypic differences were observed between patients with an intragenic *STAG1* deletion and those with a pathogenic nucleotide variant. It was concluded that *STAG1* is a new cohesinopathy gene that acts via a loss-of-function mechanism, but patients with *STAG1* haploinsufficiency are difficult to recognize due to the lack of a distinctive clinical phenotype.

PWG: ANIMAL, PLANT, AND COMPARATIVE CYTOGENETICS.

Co-ordinators:
J.S. (Pat) Heslop-Harrison (UK),
Valérie Fillon (F)

Ten abstracts were selected for a short oral presentation during the workshop. The studies cover various aspects of animal and plant cytogenomics.

Dimitij Dedukh (Saint Petersburg, Russia) discussed genome elimination before meiosis in diploid and triploid hybrids of water frogs. Indeed, looking at gonads of parental species and hybrid tadpoles, many micronuclei were seen but not associated with cell death (as no signals in germ cells with micronuclei were seen when using caspase3 immunolabelling). Genome elimination was gradual over several cell cycles. He suggested mechanisms of genome elimination: budding from interphases, lagging at mitosis, or a combination of both events.

Martina Flegrova (Budejovice, Czech Republic) talked about sex chromosomes of looper butterflies, screening for sex chromatin presence or absence. Extending her analysis by comparative genomic hybridization (CGH), she and her co-authors concluded that sex chromatin was not a reliable marker of W chromosome presence but can be used as an indicator of W chromosome rearrangements. She is now looking at the reproductive consequences of these changes in loopers and the sex determining genes.

Maria Kyulak (Saint Petersburg, Russia) described new tandem repeats in Japanese quail (*Coturnix japonica*), a poultry species considered a model in development, behavioural, vertebrate physiology and disease studies. 23 tandem repeats (representing at least 4.8% of the Japanese quail genome) were found. FISH with specific oligonucleotide probes revealed that

some of them contribute to the heterochromatin of the short arms of CJA3 and CJAa, and microchromosomes. In addition, repeat CjapSAT was found in pericentromeric heterochromatin of CJA 1-6 and 3 pairs of michrochromosomes, as well as in p- and q-arms of CJAW.

Francesca Dumas (Palermo, Italy), by using both classical and molecular cytogenetics, described the genomic organization of repetitive DNA in *Graphiurus platyops* and *G. ocularis* genomes. Karyotype reconstruction with G- and C-banding showed the same diploid number (2n=46), with only bi-armed autosomal chromosomes in *G. ocularis* but 5 acrocentric pairs in *G. platyops*. FISH of telomeric (TTTAGG)n probes in both species revealed signals at the centromeres of all bi-armed chromosomes and at the terminal positions, suggesting that their dispersion could be linked to different mechanisms, such as chromosomal rearrangements and telomeric sequence amplification). There are another 15 species in the genus, so the evolutionary order of changes and consequences for speciation may soon be elucidated.

Ioana Nicolae (Balotesti, Romania), reported on data from the last 5 years from cytogenetic investigation of both cattle and river buffaloes (all females) with reproductive disturbances. From 209 investigated animals (144 cattle and 65 buffaloes), 31 of them (22 cattle and 9 buffaloes) showed chromosome instability by using a sister chromatid exchange SCE-test. Also a case of 2n=49,X in a sterile buffalo female with prominent withers and tight pelvis was found. There was some suggesting environmental pollutants may account for the high levels of SCE detected.

Also from Romania, **Dana Pusta** (Cluj-Napoca, Romania) presented an overview of the most frequent chromosomal anomalies in domestic animals and their consequences oN the reproductive traits. Particular attention was reserved to the disorders of sexual development (DSD) in cats, dogs, horses and cattle.

Alsu Saifitdinova (Saint Petersburg, Russia) discussed the NOR-transposition in the genome of Japanese quail which shows three pairs of active NOR chromosomes, while vast majority of bird species have a single pair of NOR-chromosome. Using primers to the conserved region of the 18S ribosomal RNA gene, rDNA was amplified from the quail genome karyotype and fragments of them were localized by FISH on short heterochromatic arms of all acrocentric chromosomes in the complement. In addition, a set of the fragments was cloned, sequenced and analyzed bioinformatically and revealed chimeric sequences containing fragments of transposable elements, fragments of MHC genes and some others. Lampbrush chromosomes preparations were particularly valuable to show transposable elements containing rDNA derivatives that formed lateral loops. Following the seminal work on lampbrush chromosomes of the late Professor Herbert Macgregor (1933-2018), Alsu will now continue running the Lampbrush Chromosome website, now at http://spass-sci.ru/lbc.

Mariano Rocchi (Bari, Italy), the President of ECA, was pleased to be able to present work with Doron Tolomeo, who could not be present. He showed data on Cercopithecini monkeys, the most karyotypically diverse tribe of Old World monkeys where 2n varies from 48 to 72. Detailed BAC-FISH assays were used to study the chromosomes of four species: *Chlorocebus ethiops* (CAE), *Erythrocebus patas* (EPA), *Cercopithecus mitis albogularis* (CAL) and *Cercopithecus petaurista* (CPE). Starting from an ancestral form, four variant forms were traced after a series of common inversions as a selective advantage. Although heterozygous inversions should produce unbalanced gametes, crossing over may be suppressed. This may be due to multiple rearrangements that occurred in the centromeric regions of these chromosomes or the presence of an evolutionary new centromere in the last common ancestor of CAL and CPE.

Anna Zlotina (Saint Petersburg, Russia) refers to chromomere organization and genomic context, by using microdissection on chicken giant lamp-brush chromosomes (LBCs) LBCs. Subsequent FISH on LBCs allowed to map the microdissected regions precisely and to evaluate

their transcriptional activity in growing oocytes. The data on genomic context of individual chromomeres were obtained by high-throughput sequencing providing information on chromomeres' size and genomic boarders indicating that prominent marker chromomeres are about 4-5 Mb in size, while common chromomeres - 1.5-3.5 Mb. Analysis of genomic features showed that the majority of chromomeres combine gene-dense and gene-poor regions, while massive loopless DAPI-positive chromomeres lack genes and are remarkably enriched with different repetitive elements.

Stefan Mueller (Munich, Germany) reported new data on the Western mosquitofish (*Gambusia affinis*), a species collected from lakes at Mondsee, near the ECC Congress site in Salzburg. In particular, the genomic region from the *G. affinis* amt gene localized on the long arm of the W chromosome (Wq) by FISH was dissected using exonic PCR probes. By using intra- and interspecific CGH and comparative expressed sequence hybridization (CESH), as well as FISH with rDNA and oligonucleotide repeat probes, and immuno-fluorescence, it was possible to demonstrate that the long arm of the *G. affinis* W chromosome is enriched for repetitive sequences. The study suggests that certain expressed noncoding elements from the amt genomic region with architectural localization along Wq may play a role in sex specific gene dosis compensation in this ZZ/ZW system.

Overall, the Animal, Plant and Comparative Cytogenetics working group gave an exciting opportunity to hear about a diverse range of topics showing the value of cytogenomic work in evolutionary and developmental terms, with many implications for disease and speciation. The discussions started during the well-attended session continued through the conference and particularly during the main sessions during the Congress.

Participants of the PWG meeting at the 12th ECC

Opening lecture

Chairs: Mariano Rocchi - Dieter Kotzot

Joris Vermeesch: Somatic chromosomal mosaicim

Single cell genomics has revolutionized our understanding of cancer evolution. Professor Vermeesch, with his seminal paper on single cell genomic analysis published in Nat Med 15: 577-583, 2009, has revolutionized our understanding of the first developmental steps in embryos. His conclusions that the rate of mitotic error is significantly higher than previously assumed, and that chromosomal mosaicism is quite common in human cleavage-stage embryos, are now well established. In his presentation Professor Vermeesch illustrated how his group has progressed towards the mapping of chromosomal instability starting from the beginning of life, from the zygote to the elderly. He also developed methods to map haplotypes in single cells. The methods reconstruct genome-wide haplotype architectures as well as the copy-number and segregational origin of those haplotypes by employing phased parental genotypes and deciphering WGA distorted SNP B-allele fractions. Part of the findings he presented is now published by Professor Vermeesch and his colleagues (Masset et al., Multi-centre evaluation of a comprehensive pre-implantation genetic test through haplotyping-by-sequencing. Hum Reprod 34:1608-1619, 2019).

Session Reports

Sunday, 6 July, 2019

Plenary session 1: Recent advances in cytogenomics

Chairs: Mariano Rocchi – Thierry Lavabre-Bertrand

Claudia Haferlach: The future of cytogenomics in the diagnostics.

The 12th European Cytogenomic Conference celebrated the discovery of banding techniques introduced for the first time 50 years ago (see the presentation by Prof. Felix Mitelman in plenary session 2). It was a revolution that paved the way for detailed studies of chromosomal aberrations, particularly in cancer and in evolution. A few years later, molecular cytogenetics introduced the FISH technology, followed by microarray analysis and, more recently, by the next generation sequencing (NGS). Professor Haferlach beautyfully illustrated in detail how the NGS technology can be of great help in clarifying genomic alterations. She reported on a large cytogenomic study on hematological malignancies that shows how new technologies can provide very powerful tools for a better diagnosis, which can be exploited for a better medical treatment to improve patient outcomes. She concluded her speech by encouraging cytogeneticists to exploit these new tools.

Michael Speicher: Liquid biopsies in patients with cancer.

Cell-free circulating DNA (cfCD) analysis is a very hot field in various disciplines of medicine, prenatal diagnosis and cancer in particular. In fact, in the present conference there were a number of presentations or posters on circulating DNA (see for example the presentation of Prof. Chiu in plenary session 5). The great advantage of the technology consists in its minimal invasiveness. On the other hand, DNA analysis of the circulating DNA poses enormous challenges. With respect to cancer, many laboratories around the world are trying to lay a solid foundation to be used to detect the early appearance of cancer, its residual disease and relapse, and emergence of resistance to therapy. In his presentation, Prof.

Speicher illustrated his laboratory's experience in the characterization of the transcription factor binding sites from the analysis of the cfCD. They identified patient-specific as well as tumor-specific patterns. Prof Speicher and his colleagues have published the validity of this approach (Ulz P. et al.: Inference of transcription factor binding from cell-free DNA enables tumor subtype prediction and early detection. Nat Commun 10:4666, 2019).
These two high profile presentations were followed by three selected abstracts:

Pascal Chambon: A simple, universal and cost-efficient dPCR method for the targeted analysis of copy number variations.

Laïla El Khattabi: Next Generation Mapping, a novel approach that enables the detection of unbalanced as well as balanced structural variants.

Paolo Reho: Low-coverage whole genome sequencing in plasma circulating cell-free DNA analysis: the Turner syndrome experience.

Plenary session 2: 50 Years of chromosome banding
Chairs: Kamlesh Madan - José Garcia-Sagredo

The second plenary session of the conference was devoted to celebrating and commemorating the 50 years of banding techniques. We were honored to have two very interesting speakers: **Felix Mitelman** from Lund, who is well known for his pioneering work on chromosomal changes in cancer and who was involved from the beginning of the banding era, and **Darío G. Lupiáñez** from Berlin, who is known for his work on 3D spatial organization of DNA.

Felix Mitelman, in his talk *Chromosome banding: the end of the Dark Ages*, told us about the developments in cytogenetics since the discovery of Q- banding, which led to a profusion of banding techniques each with its own specific properties and applications. Chromosome banding has played a pivotal role in everything that followed in the advancement of clinical genetics and of basic research right up to sequencing and gene mapping. Numerous constitutional chromosome abnormality syndromes have been delineated and can be detected post and prenatally, establishing clinical cytogenetics as a medical speciality. Chromosome banding has played an extremely important role in cancer research. It has led to the identification of cancer-associated chromosome changes for specific tumor types and to pinpointing the location of cancer-initiating genes. This has proved to be a means of unravelling pathogenetic mechanisms in the development of cancer. The impact of chromosome banding simply cannot be overstated.
Felix Mitelman illustrated this point by taking us through the story of development of Gleevec – starting from the first discovery in 1960 of the presence of a short chromosome 22, the Philadelphia (Ph) chromosome, in cells of patients with chronoic myeloid leukemia (CML): after the discovery of banding mehtods it was found that the Ph chromosome was in fact a result of a chromosomal exchange between chromosomes 9 and 22; a decade later it was discovered that the breakage and reunion in the translocation creates a new gene *bcr:abl;* some years later this new gene was shown to produce an abnormal protein which triggers off cell proliferation leading to CML; with the cooperation of pharmaceutical companies a drug, Gleevec, was developed that could inhibit the action of the protein – thus curing the disease; Gleevec was approved for treatment of CML in 2001. Today patients with CML, which was a lethal disease, now show complete remission with improvement in quality of life and a nearly normal life expectancy. A beautiful example of how chromosome banding has contributed to medical cure of a potentially lethal disease!

In the second talk **Dario G. Lupiañez** spoke about the implication of 3D spatial organization of DNA in evolution and disease. He explained how TADs (topologically associating domains) are the basic structural units implicated in transcription regulatory elements. Structural variations that disrupt TADs may alter or modulate several mechanisms of gene regulation. These alterations of TADs may have a positional effect influencing the expression of genes at some distance from the breakpoints of some structural variations. Lupiañez showed several examples of malformations, such as brachydactyly. In summary, the alteration of spatial 3D genome is implicated both, in evolution and in the development of diseases.

Concurrent session 1: 3D chromatin organization and dynamics

Chairs: Jean-Michel Dupont - Darío G. Lupiáñez

That chromatin organization is of great importance for correct gene expression has become clear with increasing in-depth understanding of the mechanisms linking 3D structure and regulation of expression.

During this exciting session, three aspects of this expanding field were presented.

In the first talk, **Alexandre Reymond**, from the Center for Integrative Genomics in Lausanne University (Switzerland), presented the case of two closely related CNVs on chromosome 16p11.2, the proximal 600kb BP4-PB5 CNV and the distal 220kb BP2-BP3 CNV, both of which are involved in neurodevelopmental and neuropsychiatric disorders. Alexandre Reymond showed that normal gene expression is mediated by specific chromatin looping and that 3D organization in one region can be altered by a CNV (deletion or duplication) in another region. He further demonstrated how this specific CNV prone chromatin structure characterized by several segmental duplications could have evolved specifically in Homo sapiens through the duplication of an ancestral segment including the BOLA2 gene, driven by the evolutionary advantage of a better iron metabolism.

The next talk by **Mario Nicodemi**, from the Dipartimento di Fisica of Naple University (Italy), addressed one of the greatest challenges for medical applications of this new understanding of the importance of chromatin architecture: how to predict the consequences of any new structural variant (SV) discovered during the genetic workup of a patient? Using polymer physic-based computation, Mario Nicodemi demonstrated that 3D conformation of chromatin and presence of ectopic contacts in specific regions can be predicted, with confirmation of these predictions by Hi-C data from the EPHA4 and PITX1 loci. Development of such an in-silico analysis tool would be of invaluable help for a better understanding of SV consequences in order to provide patients with a more robust interpretation of genomic rearrangements.

The last talk was a selected presentation from abstracts: “Chromosome radial positioning in spermatogenic germ cells from mus musculus” by **Mireia Sole** from Barcelona University (Spain). This talk addressed the question of the specific organization of chromosomes in the nucleus during spermatogenesis in the mouse. FISH labelling of all chromosomes and confocal 3D imaging were used to analyze the dynamic position of individual chromosomes in 5 different stages of spermatogenesis. There were two main results. One was that the telomere clustering of chromosomes during the bouquet formation leads to a specific radial arrangement of chromosomes during the pachytene stage with the smallest chromosomes being at the periphery of the nucleus, as opposed to the classical nuclear arrangement where they are preferentially toward the nuclear center. The second was that the position of chromosomes is partly related to gene activity, including that of sex chromosomes during their activation /inactivation process, resulting in an excess of genetic material in the interior of the nucleus.

Concurrent session 2: Clinical cytogenomics
Chairs: Damien Sanlaville - Orsetta Zuffardi

There were three talks in this exciting Clinical Cytogenomics Session.
The first speaker was **Nicole de Leeuw**, from the Department of Human Genetics of Radboud University Nijmegen, Netherlands. She presented an overview of CNV and diseases: in constitutional diagnostics
Nicole de Leeuw first gave an overview of the different types of DNA variations and imbalances identified in the human genome such as chromosomal aneuploidy, balanced and unbalanced chromosomal rearrangements, CNV (Copy Number Variants), Indel, repeat extensions and SNV (Single Nucleotide Variants). She also mentioned the different techniques to detect CNV namely karyotype, FISH, aCGH but also WES (Whole Exome Sequencing).
After this introduction, she showed several rare and recurrent CNVs found in patients with ID and/or CMA such as CNV on chromosome 22s, 16p or 15q and explained the importance of LCRs in such chromosome abnormalities.
She stressed the problem of the penetrance of such CNVs and cited the publication of Girirajan published in 2012 in NEJM (Phenotypic heterogeneity of genomic disorders and rare copy-number variants).
She then presented several examples:

- Importance of exome sequencing in prenatal context: a foetus at 20+5 weeks showed skeletal dysplasia. A WES was performed which allowed the identification of a del 1q21.1 in the foetus and the father. Exome sequencing identified a heterozygous SNP in the mother in 5'UTR region of *RBM8A* gene leading to the prenatal diagnosis of TAR syndrome.
- Common recurrent CNV: del 2q13 of 170 kb including *NPHP1* gene causing adult onset ESRD (end-stage renal disease)
- Rare recurrent 14q32 microdeletion encompasses imprinting gene regions with a clinical feature of UPD14
- Line and Alu mediated (non)recurrent deletions in Alveolar capillary dysplasia with misalignment of pulmonary veins (ACDMPV). In a study by Szafranski, 38 patients out of 45 have a microdeletion at least near the *FOXF1* enhancer (LINE- and Alu-containing genomic instability hotspots at 16q24.1 associated with recurrent and non-recurrent CNV deletions causative for ACDMPV, Hum Mut 2018)
- Intragenic hemizygous loss in Xp11.2: deletion of 94 kb in *DMD* genes compatible with DMD or BMD phenotype.
- Rare intragenic copy number gain in 10 kb in *PAX6*
- Same intragenic gain in TTN in 2 sibs with muscle weakness and fatigue: Limb-girdle muscle dystrophy type 2J

For the importance of the WES, Nicole De Leeuw referred us to the publication of Pfundt et al, titled "Detection of clinically relevant copy-number variants by exome sequencing in a large cohort of genetic disorders" (Genetics in Medicine 2017). CNVs identified by WES allow an overall increase in diagnostic yield of ~2%.
Following the WES, she showed the importance of WGS in constitutional disorders with the example of *IQSEC2* gene and its importance in characterizing chromosomal breakpoints at the molecular level. She referred to the publication of Redin et al., (The genomic landscape of balanced cytogenetic abnormalities associated with human congenital anomalies, Nat Genet 2017).
Finally, Nicole de Leeuw concluded that WGS would be the most efficient approach to identify the cause of development disorders.

The second speaker was **Malte Spielmann** from Human Molecular Genomics at Max Planck Institute in Berlin who spoke about the effect of structural variation in the three-dimensional genome.

Recent studies have shown that SVs can not only affect gene dosage but also modulate basic mechanisms of gene regulation. So, when we perform a WGS and we identify a CNV loss without OMIM gene, what can we conclude? Malte Spielmann presented the mechanism which could correlate such a deletion to the phenotype and the technique that could help us in this situation (Malte Spielmann at al., Structural variation in the 3D genome Nat Rev Genet 2018). For the mechanism, Malte Spielmann stressed the interaction between the enhancer and the gene (Kragesteen et al., studied Pitx1, a regulator of hind limb development, and showed that dynamic changes in chromatin conformation can restrict the activity of enhancers, Nat Genet 2018). Malte Spielmann also presented several examples of disrupting or fused TADs responsible for human syndromes (Breaking TADs: How Alterations of Chromatin Domains Result in Disease, Trends Genet 2016).

As far as the technique is concerned, Malte Spielmann explained the importance of HiC to study chromatin interactions. He mentioned the main technical points and the limits and importance of this technique. He showed that HiC is a good method to detect SVs (Structural Variants) at a reasonable cost (1300 euros per sample). He also stressed that HiC can detect all the SVs.

That Hi-C is good for the detection of large SVs in patient samples was shown in 10 individuals with developmental delay (DD). Hi-C was performed on 3 cell types: fibroblasts, amniocytes and lymphoblastoid cell lines. For example, it was found that a duplication of a TAD boundary at the *SOX9* locus causes neo-TAD formation and is associated with Cooks syndrome.

To finish, Malte Spielmann conclude that:

- Changes in the regulatory landscape are a major cause of congenital disease
- Hi-C can be used to identify SVs
- The dynamics of chromatin interactions is important and we need a better knowledge of the 3D genome.

The last talk was a selected abstract of **Jesper Eisfeldt** about the evolution of cytogenetics to cytogenomics with the interest of whole genome sequencing as a genetic test in rare disease diagnostics.

Concurrent session 3: Structural organization of the human genome

Chairs: Joris Vermeesch - Nicole de Leeuw

Megan Dennis, from the University of California, Davis, in her lecture titled “The role of duplicated genes in human brain evolution and disease”, presented her view of the underlying genetic contributors to unique human adaptive traits. She posits that role of human-specific segmental duplications as a possible source of neurological innovation and disease has remained largely understudied. By performing Pacific Biosciences long-read sequencing, Megan Dennis and collaborators identified over 30 gene families mapping within human-specific segmental duplications. Targeted long-read sequencing in diverse human populations is currently being performed to accurately detect variants in these human-specific segmental duplications. The results of these studies will offer important insights into if/how these genes contribute to innovative neurological features that distinguish modern humans from related great ape species.

Francesca Antonacci from the University of Bari, Italy, presented a talk, titled “Inversion variants in the human genome”, focused on the importance of genomic inversions in predis-position to disease and evolution.

Despite their relevance, inversions represent a relatively unexplored form of structural variation. Combining molecular cytogenetics, genomic approaches, and sequencing techniques, Fran-cesca Antonacci’s group recently characterized

several inversion polymorphisms in the human population associated with regions predisposed to disease- causing microdeletions. Recent advances in the discovery of structural variation using single cell strand sequencing and its applications in identifying inversions in primate genomes were also shown. In particular, combining single cell strand sequencing with cytogenetics, Francesca Antonacci and colleagues identified many hotspots of genomic instability that show a great rearrangement activity in primates, likely implicated in evolutionary innovations, as well as medical conditions.

Anna Lindstrand from the Karolinska Institutet, Sweden, ended the plenary session with her presentation "Cytogenetically visible inversions are formed by multiple molecular mechanisms". She presented her experience utilizing short-read whole-genome sequencing and/or Sanger sequencing to characterize cytogenetically detected chromosomal inversions at the nucleotide level. All large inversions, characterized in detail, showed little to no microhomology in the breakpoint junctions; this is similar to what is commonly seen in reciprocal translocations. Also gene disruption frequency was similar to the frequency obtained for balanced translocations. In summary, Anna Lindstrand showed that high-coverage short-read WGS can detect a substantial fraction of copy number neutral inversions and resolve the breakpoints at the nucleotide level. Moreover, NAHR is likely not the major mechanism underlying the formation of large chromosomal inversions, as most inversions were mediated through mechanisms other than ectopic recombination.

Concurrent session 4: Human infertility

Chairs: Elisabeth Syk Lundberg - Sevilhan Artan

The session included two invited speakers and one selected oral presentation.

The invited speaker **Pierre Ray** (University Grenoble Alpes, France) discussed the problem of male infertility in humans, which is characterized by multifactorial etiology, often with a strong genetic component. The most commonly identified causes of male infertility concern chromosomal defects affecting the gonosomes, as well as structural rearrangements, often associated with infertility. However hundreds of genes are expressed in the testes and are necessary for spermatogenesis. Several major genes have been identified using homozygosity mapping and whole exome sequencing has recently allowed improved possibilities to more efficiently identify genetic causes of male infertility.

The second lecturer, **Terry Hassold** (School of Molecular Biosciences, Washington State University, USA)**,** addressed the old problem of how aneuploidy arises in humans. Aneuploidy is the most common genetic complication of pregnancy, with approximately 0.2-0.3% of newborn infants being trisomic and studies of preimplantation embryos suggest that a large proportion of fertilized human eggs have extra or missing chromosomes. Most of these errors result from the fertilization of a chromosomally abnormal egg by a normal sperm. There are multiple routes to female-derived aneuploidy including errors occurring during the long meiotic arrest stage or as part of the meiotic cell cycle checkpoint machinery. Recent studies confirm huge differences between human males and females in the way in which chromosomes find and synapse, in the packaging of chromatin, and in the control of the meiotic recombination pathway. Further, they indicate that errors in fetal oogenesis - especially those that lead to failure to recombine or abnormally located crossovers - are surprisingly common in humans.

The selected abstract was presented by **Harita Ghevaria** (Preimplantation Genetics Group, University College London, United Kingdom), who had used next generation sequencing (NGS) to detect premeiotic errors in human oocytes. NGS provided accurate information regarding the frequency of aneuploidy that is due to premeiotic errors compared with that caused by errors at MI

of oogenesis. The overall frequency of premeiotic errors in this study, including 18 women and 68 oocytes, was approximately 16%, but the frequency varied between the women. On an individual basis this seemed to be influenced by genetic factors.

Monday, 8 July, 2019

Plenary session 3: Tumor Cytogenomics I
Chairs: Felix Mitelman – Roberta Vanni

The session included two invited speakers and four selected oral presentations.

The first invited speaker, **Fredrik Mertens** (Lund, Sweden), presented data on the patterns of clonal evolution in sarcoma subtypes arising through three pathogenetic mechanisms, i.e., sarcomas with complex genomes (myxofibrosarcomas, MFS), gene fusion-driven myxoid liposarcomas (MLS), and amplicon-driven well-differentiated liposarcomas (WDLS) from which the group had access to multiple samples during tumor progression; a further requisite was that at least one year should have elapsed between first and last sampling. They also studied multiple samples from some of the primary lesions, in order to evaluate intra-lesional heterogeneity. Clonal heterogeneity was assessed through a combination of chromosome banding, single nucleotide polymorphism (SNP) array, and whole-exome sequencing analyses. They could show that the type of clonal evolution – i.e., whether nucleotide or chromosome level mutations predominate – and the rate by which new mutations accrue vary considerably among the three sarcoma types. In MFS, tumor progression was usually accompanied by accumulation of both chromosome and nucleotide level aberrations. Primary MLS display little intratumoral heterogeneity and few new mutations are found in local recurrences or metastases. WDLS, on the other hand, showed extensive inter-cellular variation in terms of chromosome level aberrations; this variation, however, had only minor impact on the predominant clone in each tumor. Furthermore, no significant single nucleotide variants were seen in primary tumors or at relapse. Thus, there is a considerable variation among sarcomas caused by different pathogenetic mechanisms with regard type of clonal evolution and to the rate by which new mutations become predominant.

The second lecturer, **David Gisselsson** (Lund, Sweden), reported his research team's work on the evolutionary processes in three prototypical childhood cancers, neuroblastoma, Wilms tumor and rhabdomyosarcoma and used the detected genetic variation to reconstruct the evolutionary history of each tumor. They find that approximately 90% of primary tumors show a history of branching evolution, sometimes leading to regional variation in genetic markers that are used for treatment decisions, e.g., 1q gain in Wilms tumors and the presence of structural vs. numerical aberrations in neuroblastoma. This variation could be traced back to four distinct evolutionary trajectories of which the ones indicating intense tumor cell competition are associated with a higher risk of relapse. Timelines inferred from the spatial distribution of genetic changes indicate that the genetic alterations associated with prognosis emerge at different points in evolutionary history for different tumor types, for example at the initiation of clonal expansion for neuroblastoma but closer to presentation for Wilms tumor. Tumors with a high relapse risk exhibit extensive branching into novel clones, leading to a rich sub-clonal underground that can act as a reservoir of genetic variation. In line with this, they found that chemotherapy typically leads to a replacement of the original clones by a set of collaterally related survivors. Similarly, the clones of

metastatic relapse are usually distant collateral relatives of the primary tumor's main clones. However, relapse manifesting at multiple sites typically have a common single cell ancestor, indicating that a specific anatomic locus acted as a bridgehead for further metastatic spread. From a clinical standpoint, the data indicate that (1) certain genetic markers used for treatment decisions today show a variation that in fact prevents unequivocal classification of tumors as positive or negative, (2) mutation analysis for the purpose of targeted therapy of relapsed tumors warrant resampling and reanalysis because data from the primary tumor can be misleading, and (3) strategies to prevent death from incurable relapse should focus more on early anatomic localization and elimination of the first relapsing clone to prevent dissemination to additional sites.

The first selected abstract was presented by **Roberto Valli** (Varese, Italy) who reviewed data **on e**xpression studies in a large series of 97 patients with Shwachman-Diamond syndrome (SDS) in relation to clonal chromosome anomalies in the bone marrow. Two clonal chromosome changes are frequent in the bone marrow cells of SDS patients: an isochromosome for the long arm of chromosome 7, i(7)(q10), and an interstitial deletion of the long arm of chromosome 20, del(20q). Both these clonal anomalies have been shown to be favorable prognostic signs. Expression analysis of bone marrow cells in relation to the presence of clonal chromosome anomalies were studied in five cases with del(20q), one case with i(7)(q10), and two cases with other anomalies. The study was performed by microarray technique considering the whole transcriptome, and three gene subsets, selected as relevant for bone marrow function. The results were compared with those of nine patients with SDS without clonal anomalies, and of nine healthy subjects. There was a significant difference between gene expression in the bone marrows of SDS patients and healthy subjects, both at the whole transcriptome level and of the gene sets selected. The deletion del(20q), with the gene *EIF6* consistently lost, even in patients with the smallest losses of material, apparently changes the transcription pattern: a low proportion of abnormal cells leads to a pattern similar to SDS patients without acquired anomalies, whereas a high proportion gives rise to a pattern similar to healthy subjects; this may explain the favorable prognosis in SDS patients with del(20q). The only case with i(7)(q10) showed a transcription pattern similar to healthy subjects, paralleling the positive prognostic role of this anomaly.

The next speaker, **Paola Caria** (Cagliari, Italy), presented data on the three-dimensional (3D) telomere organization in papillary thyroid carcinomas. Well-differentiated thyroid cancer (WDTC) is the most common endocrine malignancy and papillary thyroid cancer (PTC) represents a large group of WDTC with two main histologic variants: classic PTC (cPTC) and follicular variant PTC (FV-PTC). FV-PTC is divided into two sub-groups based on two different morphological aspects: infiltrative and encapsulated nodules. Recently, the encapsulated variant has been reclassified as noninvasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP), because it shows features similar to non-malignant lesions. At the molecular level, PTC may show *BRAF* mutations and/or *RET7PTC* rearrangements (BRAF-like nodules) and *RAS* mutations (RAS-like nodules). Several studies using quantitative 3D telomere imaging have shown that cancer cells have an altered 3D telomere organization, in contrast to normal cells. To evaluate if specific telomere architecture may characterize PTC histologic variants, quantitative fluorescence in situ hybridization (Q-FISH), 3D imaging and 3D analysis were performed in 16 thyroid lesions: 5 cPTC, 3 FV-PTC, 4 NIFTP and 4 FTA (follicular thyroid adenoma), using normal thyroid tissue (NT) as control. Moreover, *RET/PTC* rearrangements and BRAF expression (indicative of *BRAFV600E* mutation) were investigated by FISH and immunofluorescence, respectively. The research group found different telomere profiles in tumors compared to control ($p<0.05$), and

telomere profiles of FTA close to NT. The comparison of 3D telomere profiles of the tumors demonstrated that NIFTP has longer telomeres than cPTC and FV-PTC ($p<0.001$). No correlation between molecular alterations and 3D telomere profiles was observed. The data suggest that 3D telomere organization might have diagnostic utility and might help the clinical management of NIFTP patients.

The third speaker, **Jordi Camps** (Barcelona, Spain), presented the results of a study on patterns of acquired uniparental disomy revealing bi-allelic inactivation of tumor suppressor genes (TSG) in gastrointestinal cancers and in advanced colorectal adenomas. Somatically acquired uniparental disomies (aUPDs), also known as copy number neutral loss of heterozygosity (cnLOH), are frequent events in solid tumors and have been associated with cancer-related genes. The research team aimed at integrating aUPD profiles with whole-exome sequencing mutational data in a tumor type-specific manner. Using TCGA datasets for 1,032 gastrointestinal cancers, including colon (COAD), rectum (READ), stomach (STAD), esophageal adenocarcinoma (EAC) and esophageal squamous cell carcinoma (ESCC), they showed that gastrointestinal cancers display tumor type-specific profiles of aUPD. By inferring genome ploidy, they demonstrated that an increased number of aUPD events were present in samples with higher DNA ploidy compared to near-diploid tumors. The integration of whole-exome sequencing and aUPD provided evidence of bi-allelic inactivation of TSGs and activation of oncogenes in a tumor type-specific manner. Of note, *APC* was the most recurrently inactivated gene in COAD and READ by the presence of homozygous mutations as a consequence of aUPD. Likewise, *ARID1A* and *NOTCH1* were bi-allelically inactivated by aUPD in STAD and ESCC, respectively. Furthermore, while *TP53* showed inactivation caused by aUPD at chromosome arm 17p across all tumor types, copy number losses at this genomic position were also frequent. When the group studied the presence of aUPD in premalignant lesions of the colorectum, they observed that 31% of samples showed aUPD at chromosome arm 5q. All samples with aUPD displayed a homozygous mutation in *APC*. Finally, the presence of mosaic UPD was detected at a higher frequency in peripheral blood lymphocytes of patients with colorectal cancer in a case-control study.

The last talk was given by **Sabrina Haslinger** (Vienna, Austria) who presented data on *ZNF384* gene fusions in 15 cases of B-cell acute lymphoblastic leukemias identified by systematic FISH and array screening. The subdivision of childhood B-cell precursor acute lymphoblastic leukemia (BCP-ALL), based on specific genetic features, such as gene fusions or ploidy and copy number aberrations, provides the basis for treatment stratification and decisions. The so-called "B-other" group embraces all cases with rare recurrent abnormalities that are hitherto less well defined. Because they include potential candidates for targeted and personalized therapies, they are currently the main focus of interest. One of these recently identified subgroups that accounts for approximately 4% of B-ALL and up to 10% of “B-other” ALL cases, involves the *ZNF384* gene, which has been found to be fused to at least ten different partners. These cases have commonly a CD10 negative (pro-B/BI) or CD10 low immunophenotype with myeloid markers and a distinct gene expression pattern. To search for *ZNF384*-positive cases, the group screened all “B-other” cases that were enrolled in the ALL-BFM 2009 study with SNP/CGH arrays as well as an additional selected cohort with a *ZNF384*-specific dual color break-apart FISH probe set. They found fifteen patients with a *ZNF384* fusion, which constitutes approximately 5% of all “B-other” ALL cases. Eight of them had an *EP300-ZNF384* fusion, three a *TCF3-ZNF384* fusion and one had an *EWSR1-ZNF384* fusion. The remaining three had novel fusion partners, two of which were ascertained as *CCAR1* and *NIPBL*. Four of them had *IKZF1* deletions and all but one are in remission, supporting the notion that *ZNF384*-positive cases seem to respond well to current therapies.

Concurrent session 5: Tumor cytogenomics II
Chairs: Claudia Haferlach - Harald Rieder

In this session **Liran Shlush** from the Weizman Institute in Rehovot, Israel presented his work on the evolution from age-related clonal hematopoiesis to AML and the related risk factors. He summarized the current knowledge on age related hematopoiesis and the so far identified risk factors on the genomic level for progression to hematological malignancies. Further, he discussed the additional evaluation of blood parameters and their value for the identification of patients at risk for the development of AML. This might set the stage to develop a strategy to diagnose AML before disease symptoms occur. Liran Shlush looked even more into the future by proposing a clinical trial aiming at the prevention of hematological malignancies by administering drugs such as IDH inhibitors to probands with age related clonal hematopoiesis at risk harboring mutations in *IDH1/2* or other targetable genes.

The second speaker **Floris Foijer** (Groningen, The Netherlands) spoke about how single cell DNA sequencing can be used to quantify karyotype heterogeneity in cancer. Tools were presented that quantify and visualize chromosomal instability and the resultant aneuploidy at high resolution in primary cancer cells through single cells whole genome sequencing. Further the latest results from mouse models in which chromosomal instability was provoked in a cancer and a non-cancer setting, which can be used to study karyotype evolution in developing tumors were discussed. In addition, examples of single cells whole genome sequencing of human cancer samples were shown to exemplify how this technique can provide insight into the karyotype heterogeneity in a human background and how these data compare to findings in mice.

The third talk was selected as an oral presentation from submitted abstracts and was given by **Karla Svobodova** (Prague, Czech Republic). She presented her work on the identification of cryptic aberrations in MDS with clonal evolution and discussed the clinical relevance. MDS cases were analyzed by a variety of different cytogeneomic techniques (G-banding analysis, interphase-FISH, 24-color FISH, aCGH/SNP microarray, NGS). Clonal evolution was identified in 36/469 MDS patients. While the early genetic changes in the diagnostic samples were frequently MDS specific, most progression-related aberrations identified after clonal evolution were MDS non-specific. Copy number neutral loss of heterozygosity (CN-LOH) was detected in 19% of patients, which pointed to homozygous mutations in these regions in some cases. Based on the results of this study the conclusion was drawn that a combination of cytogenomic methods allows detection of many cryptic genomic changes and contributes to the identification of genes/genomic regions that offer potential therapeutic targets in patients with progressive MDS.

Concurrent session 6: Animal and plant cytogenomics I
Chairs: Pat Heslop-Harrison - Valerie Fillon

In the Salzburg Congress, two sessions were dedicated to animal and plant cytogenetics. Four invited speakers gave talks in a wide field of topics including plant repeatome, meiosis in pig, satellite evolution in Bovids and transposable element mobilization. The oral selected abstract gave an overview on the use of high-resolution fibre-FISH for the characterisation of genome structure.

Alain Pinton (Toulouse, France) opened the 6th session with a synthesis of the key results his team has obtained on chromosome rearrangements and meiosis in pigs. Since 1968, chromosomal controls are carried out at INRA on bovine (1000/yr), pig (2500/yr) and other species like wild boars, horses, cats and dogs (400/yr). Up to now, 43,500 pigs have been karyotyped at the age of 6-10 months leading to the detection of 208 structural rearrangements among them 29 involving sex chromosomes. Most rearrangements are reciprocal translocations (90%), but inversions (7%) and Robertsonian translocations (2%) were also found. In boars, 0.5% carry balanced structural rearrangements which would reduce fertility by 40%. Balanced abnormalities give rise to reproduction problems like spermatogenesis disturbances, or unbalanced gametes usually leading to embryonic death. Alain presented the effect of different rearrangements on reproduction by analyzing the meiotic process (meiotic pairing and recombination during pachytene) and the male and female meiotic segregation products.
Thus, unbalanced gamete frequency is varies depending on gender and the translocations. The recombination rate is also different between genders with more recombination in female in rcp12/14. Contrasting with humans, inversions only have a small impact in pigs. Moreover, the analysis of the synatonemal complexes by immunocytochemistry has shown that because of impairing (accumulation of gamma-H2AX histone) some genes can be transcriptionally repressed or over expressed with the consequence of meiotic arrest.

The second talk was presented by **Ilya Kirov** (Moscow, Russia) on the plant repeatome: cytogenetic, transcriptomic and proteomic. Repeatome is the complement of all types of repeated sequences in a genome including mobile elements, satellite DNA and others. It has been possible to identify rapidly high copy tandem by NGS bio-informatic analysis in onion, rose, moss and sunflower. Numbers of high-copy tandem repeats may be much higher than thought first. Despite the repeatome being, for a long time, considered as 'junk' and 'parasitic' DNA, it could have a functional role in the cell. The repeat abundance and distribution differ according the species as shown by FISH-based karyotyping in onion, *Allium*. Moreover, the centromere assembly is challenging and needs new pipelines for bio-informatics and long-read sequencing data. Transcribed repeats are mostly located in heterochromatin regions. They are transcribed into lncRNAs and their expression is tightly controlled by epigenetics landmarks. Retrotransposons are expressed in somatic cells (seeds, roots). Numerous transcripts have been detected in sunflower and moss. Different isoforms exist in sunflower and most of the transcripts are polyadenylated. But among the 100 members of a new active Ty1 retrotransposon in the sunflower genome, Tyrran, only two are expressed. More surprisingly, some of the retrotransposon transcripts are translated. But 70% of retrotransposon encoded proteins are truncated proteins and 30% have no known peptides. There is still a lot to discover to understand how the repeatome could contribute to the cellular mechanisms.

Plenary session 4: Chromosomal Imbalances
Chairs: Mariano Rocchi - Konstantin Miller

In her talk "The trisomy legacy: from numerical to structural abnormalities" **Orsetta Zuffardi** showed, by a whole genome approach, that some de novo unbalanced structural aberrations originate in a two-step event. The first is a maternal nondisjunction event leading to a trisomic zygote. This is followed by anaphase lagging of a supernumerary chromosome in a micronucleus where chromothripsis is ignited (illustrated by a big bang which brought the audience to full attention) shattering the chromosome into pieces. Some pieces are lost while others are re-stitched in a disordered assembly and are rescued by attaching themselves to a non-chromothripsed chromosome, either terminally or interstitially. Examples of small de novo supernumerary marker chromosomes, de novo unbalanced translocations and insertions were presented. The mechanism could also induce imprinting disorders and autosomal recessive disorders associated with iso/heterouniparental disomy.

The second invited speaker **Iben Bache** reported on the long-term outcomes of prenatally detected de novo balanced chromosomal rearrangements (BCR). Fortyone patients with a prenatally detected BCR with normal first-trimester screening and ultrasound scan from a nationwide Danish registry from 1970 to 2008 were included in this long-term follow up with a mean period of 17 years of observation. Compared to a matched control group, the BCR-group showed a signifycantly higher frequency (26.8%) of neurodevelopmental or neuropsychiatric disorders; 7.3% had milder congenital malformations. Chromosomal microarray showed no imbalances in 97% of the BCR carriers, illustrating its limited prognostic value. Mate-pair sequencing (MPS), on the other hand, revealed disrupted genes and regulatory domains. The evaluation of the MPS-results indicates a significant value of the method. However, its relatively low sensitivity shows the challenges still associated with prenatal risk prediction of long-term morbidity. In conclusion the previously known risk figures of an untoward outcome of 6-9% should be upwardly revised to approximately 27% and sequencing of the chromosomal breakpoints should be used as the first-tier diagnostic test in prenatally detected de novo BCR.

The selected presentation by **Aafke Engwerda** showed the successful approach of collaborating with a Facebook group on a specific chromosome aberration for the phenotype-genotype analysis of terminal 6q deletion. A cohort of 30 individuals from the study website was combined with 55 cases from the literature. The terminal 6q26q27 region includes thirty-nine protein encoding genes of which seven show haploinsufficiencies. The phenotype includes a variety of clinical findings. The most frequent ones, such as short stature, respiratory problems, cardiac and kidney abnormalities, are shared by one third of the patients. The results showed several subgroups in which deletions such as those for the PARK2 gene or the OKI gene were associated with specific combinations of clinical findings. The study demonstrated that social media may contribute to collecting data in rare chromosome aberrations to allow a better interpretation of the clinical spectrum in correlation to the genotype.

Concurrent session 7: Animal and plant cytogenomics II
Chairs: Trude Schwarzacher - Leopoldo Iannuzzi

Raquel Chaves (Lisboa, Portugal) opened the 7th session with a talk on the satellite evolution in Bovidae. Satellite DNA was composed of constitutive heterochromatin and is a hotspot for

structural rearrangements. Some species share similar sequences. In Bovidae, satellite are located in centromeric regions and can be involved in the occurrence of Robertsonian translocations. Thus Bovidae are good model to understand evolution. Combining PCR, sequencing, FISH and *in silico* approaches, Raquel and her team have studied the role of satellite in centromere function. Flanking regions of satellites are enriched in transposable elements, related to the centromeric activity. It was also possible to revisit the Roberstonian translocation 1:29 and to demonstrate it involves satellite DNA reorganization at the centromere level. When the fusion occurs, there is a change in the order of satellites, together with a loss of satellites. During evolution, this Robertsonian translocation has been fixed in antelope species *Tragelaphus* and a loss of satellites is observed. The genomic era, with the synergy between *in silico* and *in situ* approaches, opens up new perspectives, not only to reveal the fundamental features of satellite DNAs, but also to uncover a universal framework for understanding the roles of repetitive DNAs as a whole within the biology of chromosomes and genomes.

In the 2nd lecture, **Vincent Colot** (Paris, France) talked about transposable element mobilization: where, how and with what consequences? Transposable elements (TEs) are major engines of genome evolution. They create mutations and affect genes and genomes in multiple ways. We know little about the spectrum, genomic distribution and impact of heritable mutations created by transposable elements mobilization - how much do they contribute to heritable mutations? Population genomics studies in Arabidopsis indicate that natural selection is a key player in shaping the transposable elements landscape. DNA methylation is a key epigenetic mechanism that contributes to the stable transcriptional silencing of TEs in plants. Demethylation is essential for complex machinery ensuring maintenance of DNA methylation in three sequence contexts over TEs in plants. TE insertions accumulate in ddm1-derived epiRILs. Most transpositions take place during the propagation of epiRILs. Most of the TE insertions are contributed by a few initial donor TEs - a master copy in the starting material. Once triggered, TE mobilization generates insertions at rates that exceed small-size mutations. TE retrotransposons are distributed with more overt bias across the genome but are nevertheless often around 5'UTR, exon, intron and 3'UTR, much lower in flanking 500 bp and beyond. Local insertion preferences are associated with specific chromatin features. Amazingly, H2A.Z promotes and guides AtCopia93 integration - so H2A.Z contributes to minimize the deleterious impact of AtCopia93 mobilization. Transposons can be highly mutagenic.

In the final talk, **Fengtang Yang** (Cambridge, United-Kingdom) gave an overview on the characterization of complex genomic structure and variation by high-resolution fibre-FISH. This technique is performed on extended chromatin and gives 1000-fold more resolution than FISH on metaphase chromosomes. Data on relative length, position and copy number are really obtainable by FISH on fibres obtained by alkaline lysis. The probes are generated with Genome Plex WGA amplification and labelled using the same process of reamplification. Molecular combining allows quantitative sizing of overlaps and gaps. It is possible to define the order, the copy number and the haplotypes. The more useful applications concern the sizing of gaps in reference genomes, the characterisation of complex Copy Number Variation and Structure Variants, the verification of rearrangements after CRISPR cas9 technology, the validation of fusion genes identify by RNAseq and the assistance of de novo assembly of regions with gene families. An example was given on the characterization of structural variation of the human glycophorin locus in poster 6.P3.

Concurrent session 8: Accreditation, quality control and education
Chairs: Konstantin Miller - Albert Schinzel

In his talk on European certification and continuous education of Clinical Laboratory Geneticists working in cytogenetics **Thomas Liehr** gave an overview of the various tasks in human genetic testing and of the specialists involved such as Medical Geneticists, Clinical Laboratory Geneticists, Technologists, Genetic Counsellors and Genetic Nurses. To harmonize professional education, the European Board of Medical Genetics European Certification has started registration of Clinical Laboratory Geneticists (CLG) based on core capacities and continuous education. Results of a survey in 35 European and some non-European countries on the rights and duties of CLGs were presented. An official EU-recognition of the qualification is desired.

The second invited speaker **Thomas Eggermann** reported on the challenges and opportunities in next generation sequencing (NGS) in the context of quality assurance. NGS, as basic tool of human genetic testing, needs to be embedded in a quality system that requires precisely defined quality parameters, a structured database and a validated bioinformatic pipeline, validation of the wetlab procedures, and participation in quality assessment schemes. Examples for a possible validation of NGS were given. For accreditation according to DIN EN ISO 15189 (or 17025 where applicable) the matrix, parameter/gene, method and platform have to be considered. In conclusion, it was stated that accreditation is an adequate tool for quality assurance in NGS-based diagnostics and that in the diagnostic context only genes with known relationship between aberrant variant and phenotype should be analysed.

The selected abstract titled **'**What should laboratory specialists in clinical genetics know about chromosomes ten years from now?' was presented by **Ron Hochstenbach.** He pointed out that Whole Genome Sequencing (WGS) as the future first-tier genetic test is able to detect a variety of genetic variants, structural rearrangements and uniparental disomy in a single diagnostic test. The laboratory specialist needs to be aware, however, of the types and frequencies of pathogenic genome variants that remain undetected by WGS. One survey has shown that about 8% of clinically relevant abnormal results would be missed by WGS based on the current industry standard. These include balanced chromosomal rearrangements with breaks in repetitive DNA sequences, low level sex chromosome mosaicism and rare abnormalities such as r(20). With the development in sequence technology, it is to be expected that the spectrum of missed abnormalities will change. It remains a challenge for laboratory specialists in the future to recognize the need for follow-up of WGS-data and to assure the competence in classical cytogenetics to perform the tests according to the state of the art.

Tuesday, 9 July 2019

Plenary session 5: Prenatal diagnosis
Chairs: Damien Sanlaville - Maria Rosario Pinto Leite

There were four exciting talks in this session on prenatal diagnosis. Two talks were by invited speakers and two had been selected from the submitted abstracts.

The first invited speaker was **Rosa Chiu**, from the department of chemical pathology; she is the assistant dean for research at the Chinese University of Hong Kong. The title of her talk was "Cell-free DNA analysis as a tool for "non-invasive cytogenetics"**.**

The first part of her talk was about the general principle of NIPT. Rosa Chiu reminded us that fetal DNAfc (cell-free DNA) comes from the placenta. So, in a blood sample of a pregnant woman there is a mixture of both maternal DNA and placental DNA. Rosa Chiu outlined the technical principle of NIPT (Chiu PNAS 2008, Chiu NMJ 2011) and reminded us that the main purpose of it was to detect trisomy 21 (Flat positive < 0.1 %, detection rate > 99%). This strategy has led to substantial reductions in the number of invasive procedures, both chorionic villus sampling and amniocentesis (Warsof Prenat Diag 2015). Rosa Chiu then presented the several possible analyses that can be done from cell-free DNA. In fact, it is possible to detect all chromosome aneuploidy including rare autosomal trisomies, RAT (Pertile Sci Transl Med 2017). She also provided several examples including the detection of segmental aneusomies such as tetrasomy 12p detected in a foetus with omphalocoele. Nevertheless, NIPT is not a perfect test. Roso Chiu illustrated this point by presenting the article from Huijsdens-van Amsterdam et al. (EJHG 2018), which shows that iso-chromosome 21 is overrepresented among false negative cell-free DNA screening results involving Down syndrome. Eight out of 29 (28%) Down syndrome cases with a false-negative "non-invasive prenatal test" (NIPT) were associated with a 21q;21q rearrangement, compared to 2% reported in live-born children with Down syndrome.

In the second part of her talk, Rosa Chiu stressed the difference between fetal and maternal cell-free DNA. In fact, plasma DNA molecules show a predictable fragmentation pattern reminiscent of nuclease-cleaved nucleosomes, with the fetal DNA showing a reduction in a 166–base pair (bp) peak relative to a 143-bp peak, when compared to maternal DNA (Lo, Science Translational Medicine, 2010). The difference in size between these two types of DNAfc is explained by the localisation of cutting nucleosome DNA. Cell-free DNA in human plasma is nonrandomly fragmented and reflects genome wide nucleosomal organization. The ends of plasma DNA molecules select different genomic locations in fetal or maternal derived DNA (Chan et al, PNAS, 2016). In the same article Chan et al. show that it is possible to detect pathogenic variation of nucleotides. They successfully identified causative *de novo BRAF* mutation through the maternal plasma DNA analysis.

Rosa Chiu also presented two exciting articles. One is from Jiang at al. (Liver-derived cell-free nucleic acids in plasma: Biology and applications in liquid biopsy Journal of Hepatology 2019). In this article Jiang et al. show that the analysis of methylation and fragmentomic patterns of cfDNA has made it possible to determine the origin of the tissue of cfDNA.

The second article is from Serpas et al (PNAS 2019, Dnase1l3 deletion causes aberrations in length and end-motif frequencies in plasma DNA). Serpas et al, found that the deletion of Dnase1l3 in mice resulted in aberrations in the fragmentation of plasma DNA. They demonstrate that DNASE1L3 plays a role in circulating plasma DNA homoeostasis by enhancing fragmentation and influencing end-motif frequencies.

The second invited speaker was **Leen Vancoillie**, from the lab of Joris Vermeesch in the Katholic University of Leuven (KUL) in Belgium. Leen Vancoille spoke about the landscape of pathogenic copy number variations in healthy, reproducing females.

Given that 90% of cell-free DNA is of maternal origin, NIPT also gives results of genome-wide screening of pregnant mothers for maternal copy number variations (CNV's). These secondary variants may provide an opportunity for the pregnant mother and her fetus to optimize follow-up and management.

Since November 2013 NIPT, using an in-house developed and optimized genome-wide analysis, has been accredited in KUL genomic centre. NIPT is also reimbursed by the Belgian Social Security for all pregnant women since 2017. Common fetal trisomies have been detected in 1/205 pregnant women. In 2016, national guidelines for managing incidental findings detected by NIPT, including maternal incidental findings were published in Belgium (https://www.college-genetics.be/assets/recommandations/fr/guidelines/BeSHG%20NIPT%20Incidental%20Findings%20-%202016.pdf).

Leen Vancoille presented one-year experience reporting maternal secondary variants in a series of 26,123 NIPT. Autosomal rearrangements with a dominant effect were observed in 21 pregnancies (incidence 1/1243). These included five microdeletions involving a tumour suppressor gene (*NF1*, *BRIP1*, *MSH6*, *BRCA1* and *DICER1*). Four different genomic disorders were detected namely 3q29 microdeletion (n=1), PWS/AS duplication (n=1), CMT1A (n=3) and HNPP (n=7). There were two large deletions: a mosaic 17Mb 9q21 deletion and one non-mosaic 9.5Mb 4qter deletion.

Two thirds of actionable CNVs were X-linked (65/ 95). The recurrent *STS* deletion was the most common (27 cases), followed by intragenic *DMD* deletions or duplications (n=11) and *SHOX* deletions (n= 4). More details are available on *DMD* CNV in the article published by Brison et al, in Gentics in Medecine (Maternal copy-number variations in the DMD gene as secondary findings in noninvasive prenatal screening, 2019). Moreover, the recurrent int22h1/int22h2-mediated Xq28 duplication syndrome was observed 6 times; the reciprocal deletion 4 times. Six large X chromosomal deletions were detected.

In addition, 12 cases of cancer in the mother were detected between 2013 and 2019 following NIPT procedures.

These study shows that 1/275 women carry a clinically significant CNV. So, genome-wide NIPT has the potential of informing pregnant women of significant reproductive risks.

The first selected abstract was presented by **Ming Chen**, from Changhua Christian Hospital Dept. GenomicMedicine Changhua-Taiwan. Ming Chen presented the results of her study titled "A silicon-based coral-like nanostructured microfluidics to isolate rare cells in human circulation: validation by SK-BR-3 cancer cell line and its utility in circulating fetal nucleated red blood cells". Circulating fetal cells (CFC) are of importance in the field of non-invasive prenatal diagnosis research. Chen et al., developed a chip-based microfluidic platform to capture the extremely rare cell population in human circulation namely "Coral Chip" for cell capture, coupled with an automatic cell picker. The recovered cells can be subjected to subsequent cytogenetic and molecular genomic analyses.

Chen reported the validation results of the capture efficiency of their system by the SK-BR-3 cell line study (n=4), as well as the validation of the system to be used in non-invasive prenatal diagnosis (NIPD). Spiking tests of SK-BR-3 breast cancer cells were used for the evaluation of capture efficiency. Peripheral bloods from 14 pregnant women whose fetuses had non-maternal genomic markers were tested for the capture of circulating fetal nucleated red blood cells (fnRBCs). Captured cells were subjected to fluorescent *in situ* hybridization (FISH) on chip or recovered by an automated cell picker for molecular genetic analyses. The capture rate for the spiking tests is estimated as 88.1%. fnRBCs

were successfully captured from 2 mL of maternal blood in all pregnant women.

These results demonstrated that the Coral Chip system has high capture efficiency and can be used for fnRBC capture that is suitable for the genetic diagnosis of fetuses without invasive procedures.

The second selected abstract was presented by **Celine Dupont** from Robert Debré University Hospital, Paris. She reported the experience of six years of molecular cytogenetics in prenatal diagnosis and discussed the benefits, lessons and perspectives. Dupont presented retrospectively analyzed results including 4056 results of prenatal BoBs and 494 results of chromosomal microarray (CMA) for patients referred for prenatal diagnosis between 2011 and 2017 in Robert Debré Hospital (Paris). BoBs identified an abnormal result in 13,6% of cases, mainly trisomy 21 and 18. Microarray identified CNV in 13,4% of cases, among which 5.1% of cryptic CNVs. Based on their experience they propose an adapted chromosomal diagnosis test and genetic counselling according to ultrasound fetal abnormality.

Interestingly they confirmed that IUGR is associated with Williams's syndrome and high nuchal translucency with 1p36 microdeletion.

Plenary session 6:

Chairs: Mariano Rocchi – Jean-Michel Dupont

Frank Pellestor: Chromoanagenesis: cataclysms behind complex chromosomal rearrangements.

Chromothripsis was first described in 2011 in cancer cells with highly complex chromosome abnormalities. Franck Pellestor reviewed the numerous studies that have shed light on this new phenomenon since then. According to the mechanism leading to the complex rearrangement, three different phenomena are described: the original, chromothripsis, and the two newly described chromoanasynthesis and chromoplexy; these are grouped under the name of 'chromoanagenesis'. The last part of the talk mainly emphasized the relationship between chromoanagenesis and gametogenesis and early embryo development.

Adriana Di-Battista: Balanced X-autosome translocations and premature ovarian failure are associated with altered expression of growth factors, junction organization and immune pathways.

The Premature Ovarian Failure (POF) has been documented in several patients with X/autosome translocations. Adriana Di-Battista reported patients with the X-breakpoint located within the region Xq13-Xq21. The translocation did not disrupt the genes mapping in this region, but many genes, not mapping in this region, were differentially expressed. The conclusion was that the results could be explained by assuming a "position effect".

Farewell session

Chair: Mariano Rocchi

Stylianos Antonarakis: Chromatin and single cell genomics, to understand the gene dosage imbalance in aneuploidies.

The keynote lecture by Prof. Stylianos Antonarakis was an extraordinary presentation focused on the phenomenon of biallelic expression of genes. That is to say, that in a single cell only one of the two alleles is preferentially expressed. This is in some way similar to the mosaics generated in all females by the inactivation of one X (Mary Lyon's hypothesis). The phenomenon

has been already reported occasionally, but the data brought by Prof. Antonarakis were extraordinary. In fact, in recent months various papers by Prof. Antonarakis have been published on this subject in prestigious journals.

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The abstracts of the
12th European Cytogenomics Conference
can be found at:

https://molecularcytogenetics.biomedcentral.com/articles/supplements/volume-12-supplement-1

12th ECC POSTER PRIZES AND FELLOWSHIPS

We were delighted to award the
2019 European Cytogeneticists Association Poster Prizes to:

4.P3 **Doron Tolomeo** (Bari, Italy: Independent evolution in one homolog of the 20 21 syntenic association in Cercopithecini monkeys (possibly) involving evolutionary neocentromere seeding (co-authors: Giorgia Chiatante, Roscoe R Stanyon, Mariano Rocchi, Nicoletta Archidiacono, Luca Sineo Oronzo Capozzi)

1.P19 **Anna Lengyel** (Budapest, Hungary): Neurodevelopmental disorders associated with recurrent copy number variations of the short arm of chromosome 16 (co-authors: Eva Pinti, Henriett Piko, Eszter Javorszky, Mariann Tihanyi, Gyorgy Fekete, Iren Haltrich).

3.P12 **Wisam Habhab** (Tübingen, Germany): Pre and postnatal findings in a patient with a rec(8)(qter >q21.11 p23.3 >qter) due to a paternal inv(8)(p23.3q21.11) (co-authors: Sylke Singer 1, Angelika Rieß, Karl Oliver Kagan, Thomas Liehr, Karin Schäferhoff, Andreas Dufke, Ulrike Mau-Holzmann, Martin Kehrer).

2.P15 **Alla S Koltsova** (st. Petersburg, Russia): Cytogenetic abnormalities in uterine leiomyoma cells in vivo and in vitro (co-authors: Anna A Pendina, Olga A Efimova, Olga V Malysheva, Natalia Yu Shved, Thomas Liehr, Moneeb A K Othman, Maka I Kakhiani, Vladislav S Baranov).

6.P7 **Anelisa Gollo Dantas** (Sao Paulo, Brazil): Upregulation of mir 155 in a 22q11.2 deletion syndrome cohort (co-authors: Marcos Leite Santoro, Natália Nunes, Malú Zamariolli, Diogo Cordeiro Queiroz Soares, Sintia Belangero, Chong Ae Kim, Maria Isabel Melaragno).

The following colleagues received the 2019 fellowships of the E.C.A. for the 12th ECC:

Vera Lima (Porto, Portugal): Chromosome 18p11.31p11.23 Triplication (co-authors: Joel Pinto, Francisco Valente, Cristina Godinho, Sergio Castedo, Alberto Barros, Sofia Dória).

Alla S Koltsova (St. Petersburg, Russia): Cytogenetic abnormalities in uterine leiomyoma cells in vivo and in vitro (co-authors: Anna A Pendina, Olga A Efimova, Olga V Malysheva, Natalia Yu Shved, Thomas Liehr, Moneeb A K Othman, Maka I Kakhiani, Vladislav S Baranov).

Andreea Cristina Stanciu (Bucharest, Romania): The first case of 3p26.3 deletion containing only CHL1 gene associated with ASD (co-authors: Ioana Streata, Mihai Ioana, Ina Focsa, Magdalena Budisteanu).

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COMMITTEE

ECC SCIENTIFIC PROGRAMME COMMITTEE

E.C.A. News

- Renewal of the Board in 2020. The following members are due for replacement or re-election at the General Assembly in 2020: E. Syk Lundberg (Sweden), J-M. Dupont (France) J. Garcia-Sagredo (Spain), M. Rocchi (Italy), R. Vanni (Italy).
- Nomination of individual candidates for the Board together with their motivation and a CV may be sent to the President, Prof. M. Rocchi (mariano.rocchi@uniba.it) before 1 March 2020.
- As stated in the statutes, lists for the board election may be sent to the President.

E.C.A. Fellowships

- The **E.C.A.** offers two **Fellowships** for each of the following courses:
  - **European Advanced Postgraduate Course in Classical and Molecular Cytogenetics** to be held in Nîmes March 2020.
  - **Goldrain Course in Clinical Cytogenetics** to be held in Goldrain Castle (South Tyrol, Italy) September 2020.
- The fellowships **include the course fees and the accommodation** during the lectures in Nîmes or in Goldrain but **do not include travel expenses** for either of the courses or for accommodation during the practical training for the Nîmes course.
- Applications with CV, list of publications and a letter of support should be addressed to the appropriate course organizer. The Educational Advisory Council of the E.C.A. will select the successful candidates.

MINUTES OF THE E.C.A. GENERAL ASSEMBLY, SALZBURG, JULY 2019

Minutes of the E.C.A. General Assembly held on Monday 8th July 2019 in the Europa Saal, Salzburg Congress, Austria.

Approximately 46 members of the Association were present.

The President Mariano Rocchi opened the Assembly at 18.31 and welcomed those attending. The voting for board members was closed and a group, not including any candidates, was appointed to count the ballots.

The Minutes of the General Assembly held 16th June 2018 in the Room White1 of Milano Congressi, Milano, Italy, and published in the Newsletter NL42 page 17 were approved.

The two new candidates for Board Membership, Juan Blanco and Ron Hochstenbach, were introduced.

The Officers reported the state of the Association to the membership.

The General Secretary overviewed the membership of the E.C.A., with 68 new members joining since 2018. Currently, there are 919 members, 119 technologists, 128 associated members and 24 honorary members. The General Secretary noted that the Newsletter is now electronic.

The Treasurer reviewed the finances of the E.C.A. for 2018. The outcome for the Florence Conference 11th ECC was in balance. The reserves of the Association were suitable to ensure the stability of the Association and in line with the Financial Policy. The General Assembly approved the accounts.

The President announced the results of the ballot for election of Board Members. A total of 87 votes (including those received by mail) were received; 87 voted 'yes' and the list comprising S. Artan (Turkey), J. Blanco (Spain), R. Hochstenbach (Netherlands), K. Miller (Germany), F. Mitelamn (Sweden) was duly elected.

The President and Chair of the Scientific Committee, Mariano Rocchi, overviewed the present 12th European Cytogenomics Conference of the E.C.A. in Salzburg. A member asked about general organization without clear pointer to abstracts. Participant lists cannot be distributed under data protection regulations, but names and addresses of authors can be obtained from the abstracts, available at https://link.springer.com/article/10.1186/s13039-019-0439-z.

There being no other business, the President closed the General Assembly at 18.50.

MINUTES OF THE E.C.A. BOARD MEETING, SALZBURG, JULY 2019

A meeting of the E.C.A. Board of Directors was held on 8th July 2019 at Congress Salzburg, Austria.
The Board Members present were: Sevilhan Artan, Juan Blanco, José M. Garcia-Sagredo, Jean-Michel Dupont (Treasurer), Pat Heslop-Harrison (2nd Vice-President), Ron Hochstenbach, Thierry Lavabre-Bertrand, Rosario Pinto Leite, Elisabeth Syk Lundberg, Kamlesh Madan (First Vice-President), Konstantin Miller (General Secretary), Felix Mitelman, Harald Rieder, Mariano Rocchi (President), and Roberta Vanni.

The President, Mariano Rocchi, opened the meeting at 18:50.

1. The Minutes of the meeting of the E.C.A. Board of Directors held on 16th March 2019 at Hotel Vatel, Nîmes were approved.
2. Juan Blanco and Ron Hochstenbach were congratulated on their election to the Board and welcomed to the meeting. Nicole de Leeuw and Juan Cruz Cigudosa, leaving the board, were thanked for their service.
3. The proceedings of the General Assembly of the Association, held immediately before the Board Meeting, were noted.
4. The Board will meet on 14th March 2020 in Nimes.

The President closed the meeting at 19.00

E.C.A. PERMANENT WORKING GROUPS (PWG)

PWG: CLINICAL AND MOLECULAR APPROACHES TO CYTOGENETIC SYNDROMES.

Co-ordinators:

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PWG: MARKER CHROMOSOMES.

Co-ordinators:

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PWG: CYTOGENETICS OF HAEMATOLOGICAL MALIGNANCIES.

Co-ordinators:

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PWG: CANCER CYTOGENETICS, SOLID TUMOR STUDIES.

Co-ordinators:

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PWG: CYTOGENETIC TOXICOLOGY AND MUTAGENESIS.

Co-ordinators:

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PWG: ANIMAL, PLANT, AND COMPARATIVE CYTOGENOMICS.

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PWG: PRENATAL DIAGNOSIS.

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PWG: QUALITY ISSUES AND TRAINING IN CYTOGENETICS.

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PWG: CYTOGENOMICS.

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Mitelman Database of Chromosome Aberrations and Gene Fusions in Cancer

The *Mitelman Database of Chromosome Aberrations and Gene Fusions in Cancer* has been transferred to the Institute for Systems Biology (ISB) Cancer Genomics Cloud (ISB-CGC) funded by the US National Cancer Institute (NCI), and is now available at the location:

https://mitelmandatabase.isb-cgc.org/

The updated web site offers some major advances, in particular:

1) Fast search. Powered by Google BigQuery, a cloud-enabled parallel query engine, each search can be executed faster than most web applications with conventional database engines.
2) User interface enhancements. The user interface for the new web site has been simplified with a more responsive front-end, and is easier to use. Major changes added to the web site include the autocomplete drop-down list for Gene, Topography, and Morphology inputs.
3) Capability to download search results. Users can now download query results into a TSV text file.
4) Easier on-line navigation. Search results are simpler to navigate, through use of text filtering, paginations on the search result pages, and user-defined sorting of columns in the search results.

The last update on October 15, 2019, contains information on cytogenetic abnormalities in 69,551 cases and 22,091 unique gene fusions involving 12,044 genes.

EUROPEAN CYTOGENETICISTS ASSOCIATION (E.C.A.)

European Advanced Postgraduate Course in Classical and Molecular Cytogenetics

Director: Professor Jean-Michel Dupont, Paris - France

Objectives

This course was started by Professor Jean Paul Bureau 23 years ago and has been held in Nîmes under his directorship ever since. It is designed to provide advanced training in constitutional, haematological, and oncological cytogenetics to medical graduates, pharmacists, pathologists, biologists, health professionals and researchers, with an academic qualification. The students will be trained to identify genetic abnormalities for diagnosis and prognosis, and for fundamental and applied research using both classical and molecular cytogenetic techniques. The course is co-organized by E.C.A. and two French Universities, either as ***a Diploma (Basic = only the lectures or Advanced = lectures + practical training)*** or as a stand-alone course (lectures only)

Practical information

Lectures: A ten-day course held in February/March of each year.
Venue: Faculty of Medicine, Nîmes, France.
Official language: English.

Practical training (**only for students registered for the advanced Diploma**): A training of maximum 2 months in a cytogenetic laboratory. A list of laboratories is provided during the theoretical course.

Assessment **:** The assessment for the **basic diploma** will be on the basis of a one-hour examination held at the end of the lecture course**.** The knowledge of the students for the **advanced diploma** will be assessed in September by a written test (three questions) and an oral examination including a presentation (10-15 min) related to the practical training. The University will award a diploma to only those students who have passed.

All participants (including those for the stand-alone course) will receive a certificate of attendance by the E.C.A.

Topics (see opposite page).

Accommodation

A special price is available for participants in the 4* Vatel hotel close to the course venue. We highly recommend that all participants stay in this hotel where all the lecturers will be hosted in order to promote interactions during the course.
Accommodation is included in the stand-alone course fee

Registration

Registration opens in September and closes on January 30th.
To register please send a letter of application together with your CV by e-mail to one of the organizers mentioned below. If you are accepted you will receive a registration form.

Prof. Jean-Michel DUPONT
Laboratoire de Cytogénétique
Hôpital Cochin
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sylvie.mendez@aphp.fr

Prof. Thierry LAVABRE-BERTRAND
Laboratoire de Biologie Cellulaire et Cytogénétique Moléculaire
Faculté de Médecine Montpellier-Nîmes
Avenue Kennedy
30900 Nîmes, France
tlavabre@univ-montp1.fr
marie.martinez-lucon@umontpellier.fr

Registration fees

Diploma: From €360 to €1734 depending on the status of the student. Accommodation **NOT included**
Stand-alone course: €1300 (E.C.A. members) or €1400 (Non E.C.A. members); accommodation **included** on a shared double room basis. Extra fee for a single room on request.

2020 Course provisional program

This approximately 55 hour theoretical part of the course attempts to cover the field of cytogenetics in the broadest sense. The topics can be divided into the following categories:

Technical aspects:

Classical Cytogenetics: Cell culture techniques; Chromosome staining methods (Q-, G-, C-, R- banding and high resolution banding);

Molecular Cytogenetics: Methods and principles of Fluorescence In Situ Hybridization (FISH) and MFISH; Array CGH; Application of Massively Parallel Sequencing to Cytogenetics; Production and use of molecular probes; Database use in Cytogenetics;

Laboratory quality assessment.

Clinical cytogenetics:

Basics: Frequency of chromosome disorders; Cell cycle, mitosis and meiosis, gametogenesis; Heterochromatic and euchromatic variants; Numerical chromosome abnormalities; Structural abnormalities: translocations, inversions, insertions, deletions, rings, markers; Risk assessment for balanced abnormalities; X inactivation; numerical and structural abnormalities of the X and the Y; Mosaicism; Chimaeras; ISCN 2016.

Clinical: Phenotype of common autosomal and gonosomal aneuploidies; Chromosome abnormalities in recurrent abortions; Cytogenetics and infertility; Microdeletion syndromes; Uniparental disomy and its consequences; Genomic imprinting; Genetic counselling and ethical issues in cytogenetics.

Prenatal diagnosis: Indications, methods and interpretation; Risk assessment for chromosomal abnormalities; Non-invasive methods using foetal nucleic acids and foetal cells in maternal blood; Pre-implantation diagnosis.

Cancer Cytogenetics: Molecular approach to cancer cytogenetics; Predisposition to cancer, Chromosome instability syndromes; Chromosome mutagenesis; Solid tumors; Clinical application in onco-haematology.

Other:

Genome architecture; Structure of chromatin; Structure of metaphase chromosomes, Mechanisms of chromosome abberations; Origin of aneuploidy; Evolution and plasticity of the human genome; Animal cytogenetics; Plant cytogenetics.

The students will have the opportunity to evaluate the course.

The **European Cytogeneticists Association** offers **two scholarships** for **the European Advanced Postgraduate Course in Classical and Molecular Cytogenetics** to candidates of excellence. The Education Committee of the E.C.A. will select the suitable candidates.

The scholarship includes registration to the course and accommodation in Vatel Hotel in a shared double room but **does not include travel costs nor University registration.**
Scholarships will not be allocated to students whose registration is paid by a third party institution.

March 2020 Course provisional program

This approximately 55-hour theoretical part of the course attempts to cover the field of cytogenetics in the broadest sense. The topics can be divided into the following categories:

Technical aspects:

Classical Cytogenetics: Cell culture techniques; Chromosome staining methods (Q-, G-, C-, R-banding and high resolution banding);

Molecular Cytogenetics: Methods and principles of Fluorescence In Situ Hybridization (FISH) and MFISH; Array CGH; Application of Massively Parallel Sequencing to Cytogenetics; Production and use of molecular probes; Database use in Cytogenetics;

Laboratory quality assessment.

Clinical cytogenetics:

Basics: Frequency of chromosome disorders; Cell cycle, mitosis and meiosis, gametogenesis; Heterochromatic and euchromatic variants; Numerical chromosome abnormalities; Structural abnormalities: translocations, inversions, insertions, deletions, rings, markers; Risk assessment for balanced abnormalities; X inactivation; numerical and structural abnormalities of the X and the Y; Mosaicism; Chimaeras; ISCN 2013.

Clinical: Phenotype of common autosomal and gonosomal aneuploidies; Chromosome abnormalities in recurrent abortions; Cytogenetics and infertility; Microdeletion syndromes; Uniparental disomy and its consequences; Genomic imprinting; Genetic counselling and ethical issues in cytogenetics.

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Goldrain Course 2019

The Goldrain Castle, located in Italy‘s northern-most province – South Tyrol, became „home“ to the traditional Course in Clinical Cytogenetics; it was held for the 14th time this year. The course was organized by University of Zürich, European Cytogeneticist Association and The European Society of Human Genetics. The thirty-seven participants and sixteen lecturers were there to discover the mysteries of cytogenetics and I was lucky to be one of them.

Goldrain, a renaissance castle, was first built between the 12th and the 14th century, however it was re-built and enlarged during the 16th century. The castle nowadays serves as an educational and cultural centre of Venosta Valley. The location of the course, an area of fascinating medival castles (with the highest density in Europe) surrounded by fruit orchards and vineyards gives it a very special, unique atmosphere. On the whole, the course was an unforgettable experience, associated with travelling by local trains through picturesque mountain villages. The course was, as the name suggests, focused on cytogenetics. An inter-national community of physicians, laboratory workers and scientists (coming not only from the countries of the European Union but also from Egypt, Saudi Arabia, Malaysia, Pakistan, South Africa and Vietnam) thoroughly discussed the topics in 48 lectures and 10 workshops. All of these were taught by experts coming mainly from European countries but also from the US. In between the lectures and workshops, as well as in the evenings, everyone had a chance to continue discussions over a cup of tasty coffee (or, for those who prefered it, over a glass of delicious wine).

A wide range of topics from various fields - ranging from basic and molecular cytogenetics to cytogenomics were discussed. The lectures included clinical aspects as well as laboratory techniques and ethical issues. The course was divided into various sessions, covering clinical cytogenetics (dysmorphology, prenatal diagnosis, counselling for chromosomal aberrations and possibilities of non-invasive prenatal aneuploidy testing and pre-implantation genetic diagnosis and much more) as well as laboratory techniques such as FISH, MLPA, QF-PCR, SNP arrays, next generation sequencing and an introduction to CRISPR/Cas9.

Participants and lecturers of the 2019 Goldrain course

During the workshops the students were divided into small groups for discussion and practical work. Each of us gained new experience via working with ISCN systems and databases for interpreting array results; there were also interesting cases presented to us during the ethical workshop – focusing on the discord between ethics and the possibilities of contemporary medicine. Some of the students took the opportunity to present their scientific work or unusual cases in front of the community of students and faculty members to receive some valuable feedback.

There was an interesting excursion in the middle of the course. It took the course attendees to the Benedictine monastery Marienberg. Located at elevation 1,340 metres above the sea level, it is Europe's highest situated Benedictine abbey. The monks are nowadays focused on education of adults and also provide guided tours. We had an opportunity to discover the secrets of monastic life according to the Rule of St. Benedict in a local museum. The tour also included an impressive movie about the Romanesque representations of angels in the crypt. After the exhibition, during a walk from Schlinig (1700 m elevation) to Schliniger Alm (1850 m), we marvelled the breath-taking beauty of local mountains as well as green fields. During another free afternoon, many of us took the opportunity to continue to discoverthe magic of the region by visiting the city of Meran, or by taking a hike to St. Martin. To conclude, I would like to recommend this course to all cytogeneticists, as well as clinical geneticists and researchers, who wish to broaden their understanding of clinical cytogenetics. The course participants acquired a lot of knowledge, as well as practical skills and last but not least, it was an amazing experience of international bonding.

In addition to the academic part of the course, the Goldrain Castle also provided us with a homey accommodation and delicious food, including traditional South Tyrolean meals. I am very thankful for this great opportunity to learn many new things and to meet amazing people.

I would like to thank professor Albert Schinzel for organising this wonderful course and his wife and the faculty members for sharing their knowledge and experience; I also thank the other participants – for the shared fun and long, interesting discussions.

Natalie Friedova, MD (Prague, Czech Republic)

15th Goldrain Course in
Clinical Cytogenetics
August 22 to August 30, 2020

LOCATION

Goldrain Castle, Goldrain, South Tyrol, Italy
Website of the venue: **www.schloss-goldrain.it**

COURSE DESCRIPTION

The course is focused on phenotypic findings, mechanisms of origin and transmission, correlations of clinical patterns with chromosomal imbalance and modern ways of diagnosis of the latter. Special attention is paid to an understanding how deletions and/or duplications of chromosomal segments cause developmental defects. The course also addresses the optimal application of the diagnostic possibilities, both pre- and postnatally and including molecular cytogenetic methods for a precise determination of segmental aneuploidy.

TOPICS

Dysmorphic findings in chromosome aberrations: formation and interpretation – The adult and elderly patient with a chromosome aberration – Follow-up studies in patients with chromosome aberrations – Clinical findings associated with chromosome aberrations – Microdeletion syndromes: clinical pictures – prenatal cytogenetic diagnosis – Mosaics and chimeras – imprinting and uniparental disomy - Epidemiology of chromosome aberrations – Chromosome aberrations in spontaneous abortions and stillborns – Harmless chromosome aberrations – Risk assessment in structural chromosome aberrations Extra small supernumerary chromosomes – Genomic variation: a continuum from SNPs to chromosome aneuploidy – Pre-implantation cytogenetic diagnosis – Ultrasound findings indicative of chromosome aberrations – Ethical issues in the context of cytogenetic diagnosis – Non-invasive prenatal cytogenetic diagnosis.
ISCN - Practical exercises in cytogenetic nomenclature – Accreditation of cytogenetic laboratories - Accreditation of cytogenetic laboratories – Optimal use of available techniques in clinical cytogenetics – NGS – SNP arrays and Array-CGH: principles, technical aspects; evaluation of the results – MLPA - QF-PCR - FISH techniques and their interpretation – Introduction and practical exercises with database for phenotypical and variant interpretation - Students presentation of cases with difficult-to-interpret chromosome aberrations. Introduction to modern genetic editing techniques. - Practical exercises will be offered with the ISCN system for chromosome aberrations and with cytogenetic, genomic, and phenotypical databases.
- Students will have the opportunity to present their own observations and cytogenetic findings which are difficult to interpret.
- The students will have the opportunity to perform a test at the end of the course.

DIRECTOR

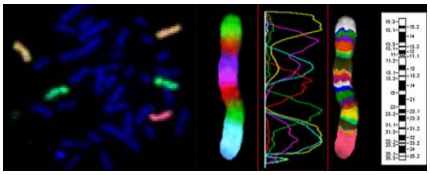
A. Schinzel (Zurich, Switzerland)

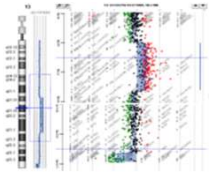
FACULTY

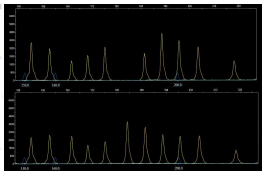
D. Bartholdi (Berne, Switzerland), A. Baumer (Zurich, Switzerland), P. Benn (Farmington CT, U.S.A.), J.M. Dupont (Paris, France), N. Kurtas (Florence, Italy), E. Klopocki (Würzburg, Germany), K. Madan (Leiden, The Netherlands), K. Miller (Hannover, Germany), R. Pfundt (Nijmegen, The Netherlands), G. van Buggenhout (Leuven, Belgium), M. Vismara (Zurich, Switzerland), J. Wisser (Zurich, Switzerland), O. Zuffardi (Pavia, Italy) and others

Full fee is Euro 1600 for a single room or Euro 1450 (VAT included) in a 2-bed-room. It includes tuition, course material, free access to internet during the course, accommodation for 7 nights, all meals, beverages during the breaks and a ½ day excursion.

For further questions please write directly to Albert Schinzel at **schinzel@medgen.uzh.ch**







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