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Incidence and clinical significance of chromosomal abnormalities of heterochromatin area of human chromosome 9

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Introduction:

Cytogenetic abnormalities of the heterochromatin area of chromosome 9 are considered to be clinically insignificant variants of human karyotype. Nevertheless, many recent studies discuss possible pathologic effect of selected variants, especially association with reproduction failure. Our objectives are to determine frequency of heterochromatine variants of chromosome 9 among patients karyotyped because of the reproduction failure; to estimate the population frequency of these variants in the Czech population and to analyze the clinical significance of these variants, especially concerning the reproduction failure.

Reproduction failure cohort

Institute of Biology and Medical Genetics, Charles University, 1st Faculty of Medicine and General University Hospital, Prague, Czech Republic
2007 – 2010

***n* = 1036**

- Includes cases of idiopathic reproduction failure only
- Karyotyping is the standard part of the diagnostic process
- Includes 463 pairs and 110 singles (506 females and 530 males)
- The cytogenetic examination identified chromosomal pathology (like robertsonian translocation or numerical gonosomal abnormalities) to be the possible cause of reproduction failure in 17 individuals (1,64 %)

Amniocentesis cohort

Institute of Biology and Medical Genetics, Charles University, 1st Faculty of Medicine and General University Hospital, Prague, Czech Republic
2003 – 2010

***n* = 995**

- Includes fetuses, that were karyotyped prenatally following the amniocentesis procedure
- The only indication for the karyotyping was advanced age of the mother (35 or more years)
- Fetuses with any abnormal findings (abnormal ultrasound or abnormal screenings) were excluded
- Includes 518 females and 477 males

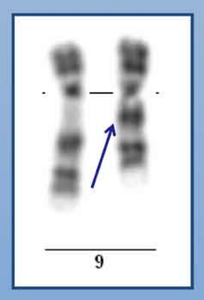
Methods:

Two separate patient cohorts were created especially for this analysis. All subjects were karyotyped because of a clinical indication and informed consent was obtained from all individuals.

All samples (either peripheral blood or amniotic fluid) were cultivated using the standard laboratory protocols. Standard G-banding method was used for the chromosome analysis. Each sample was analyzed by at least two different cytogeneticists.

Statistical analysis was done using the R-software (www.R-project.org)

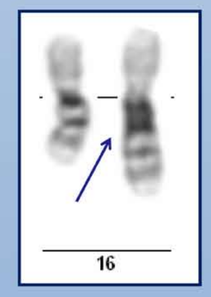
Inv(9)(p12q13)



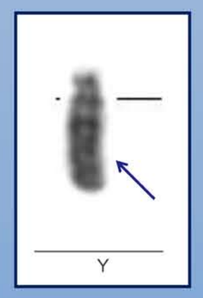
9qh-

Results

Heterochromatin variant	Reproduction failure cohort (n = 1036) Number	Reproduction failure cohort (n = 1036) Frequency (%)	Amniocentesis cohort (n = 995) Number	Amniocentesis cohort (n = 995) Frequency (%)	Relative risk analysis and statistical comparison (Fisher exact test) Relative risk	*p value*
1qh+	15	1.45	10	1.01	1,44	0.4239
9qh+	40	3.86	21	2.11	**1,83**	**0.0264**
9qh-	4	0.39	8	0.80	0,48	0.2568
inv(9)(p12q13)	18	1.74	9	0.90	1.92	0.1844
inv(9)(p12q21)	0	0.00	1	0.10	0.00	-
16qh+	26	2.51	17	1.71	1.47	0.2212
16qh-	0	0.00	2	0.20	0.00	-
Yqh+	9	0.87	4	0.40	2.16	0.2666
Yqh-	15	0.45	14	1.41	1.03	1.0000
Chromosome 9 variants in total	62	5.99	39	3.92	**1.53**	**0.0500**
Chromosome 16 variants in total	26	2.51	14	1.41	1.78	0.1571
Chromosome Y variants in total	24	2.32	18	1.81	1.28	0.7171
Total (All heterochromatin variants)	**127**	**12.26**	**86**	**8.64**	**1.48**	**0.0059**



16qh+



Yqh+

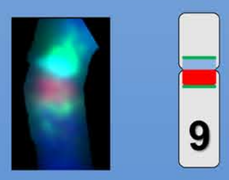
Molecular cytogenetic analysis of inv(9)

Centromeric alpha satelite *(Abbott)*

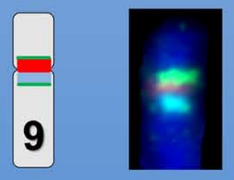
9p12 + 9q12 BAC RP11-211-N8 *(BlueGnome)*

Centromeric III satelite *(ZytoVision)*

Normal 9



Inv(9)



- We are using 3 different FISH probes for detail analysis of human chromosome 9 pericentric area
- Homologous sequences on the short and long arm of chromosome 9 (9p12 and 9 q12) are detected by the specific BAC FISH (RP11-211-N8 clone)
- There are at least 6 different sub-variants of basic inv(9) finding that could be distinguished using multiprobe FISH method (Starke et al., 2004)
- Potential association of reproduction failure with certain sub-variant of inv(9) wasn't analyzed yet
- This analysis is the main goal of the next phase of our project

Conclusions and References

- We have found that the association between heterochromatin variants of human chromosome 9 and reproduction failure is statistically significant.
- Our results in the Czech population correspond to findings of other authors.
- However, the causal mechanism for these observations remains unknown for now.
- It is certain that the majority of these heterochromatin variants is benign. However, some cases could be possibly harmful.
- The distinguishing between benign and harmful variant is impossible on the standard G-banding level. Molecular cytogenetic analysis is necessary for more detail analysis.

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