Carcinoma Ovarico: il ruolo del Biologo Molecolare

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SACRO CUORE DON CALABRIA I R C C S

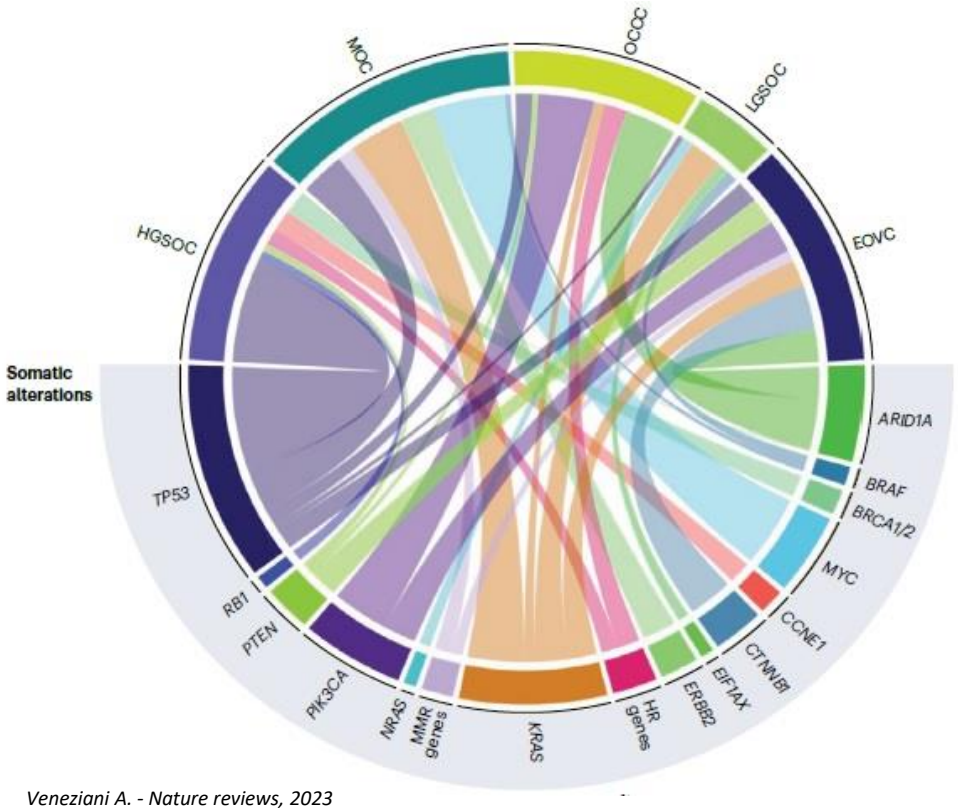
1922 - 2022 SACRO CUORE

Cancer Care Center
Numero per la Cura del Tumore
Numero Verde
800 143 143

Outline

- OC Molecular Heterogeneity
- Genomic Instability in HGSOC
- The importance of molecular diagnostics in HGSOC
- Current Laboratory Practice
- Challengies in HRD testing
- Hereditary OC

Molecular Landscape of OC



Veneziani A. - Nature reviews, 2023

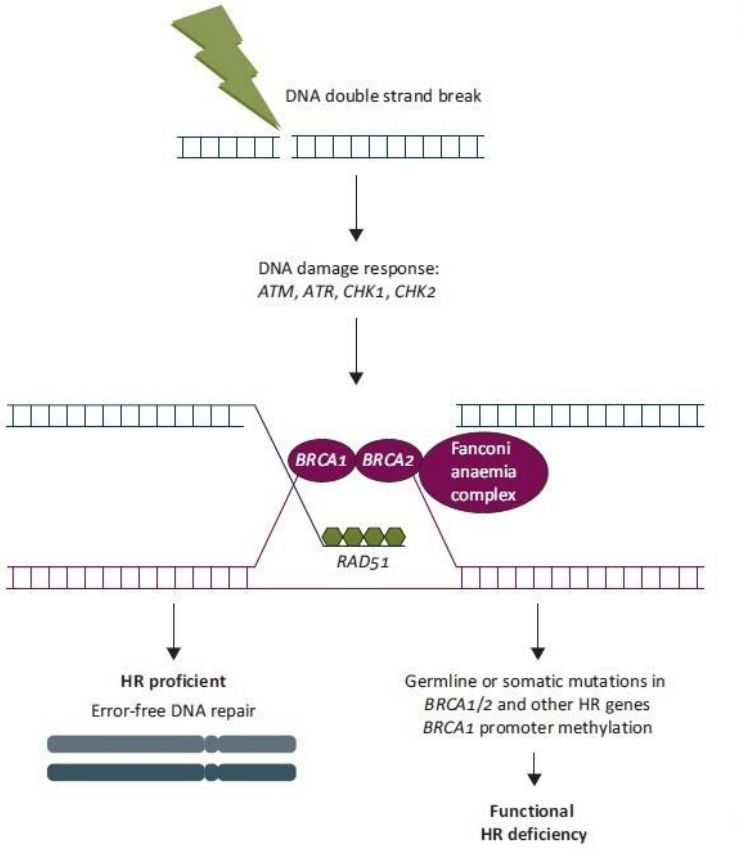
2020 WHO Classification and molecular features

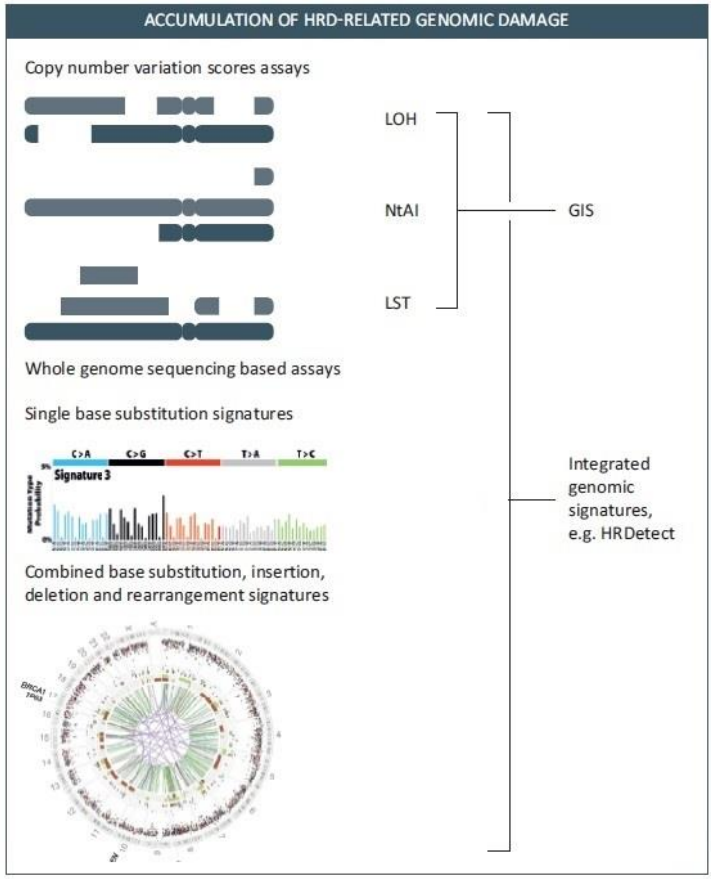
- **EOVC:** ~10%, alterations in MMR genes (Lynch), ARID1A, KRAS, CTNNB1, PTEN, POLE, FBXW7, SOX8, HER2, TP53.
- **OCCC:** ~10%, alterations in ARID1A, PIK3CA, TP53, TERT, MMR genes (Lynch).
- **LGSOC:** ~5%, alterations in MAPK (eg. BRAF), KRAS, AKT-MTOR pathway.
- **MOC**: ~5%, alterations in KRAS, TP53, CTNNB1, APC HER2amp, MMR genes (Lynch).
- **HGSOC**: ~70%, alterations in TP53, BRCA1/2, HR pathway genes, NF1, RB1, CCNE1.

Genomic Instability in HGSOC

~ 50% HGSOC exhibit Homologous Recombination Deficiency

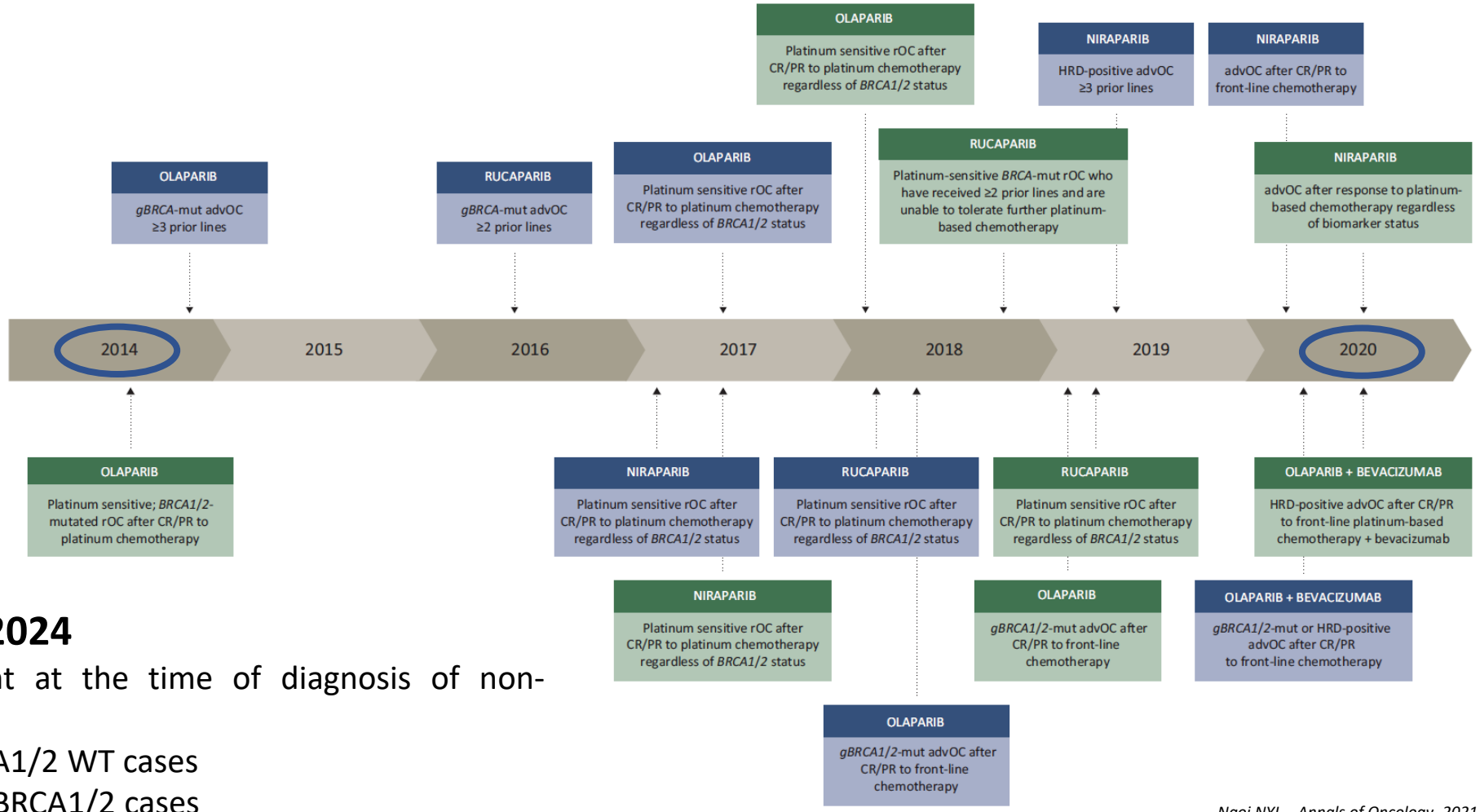
- 50% of HRD HGSOCs is explained through germline or somatic BRCA1/2 mutations
- …and the other 50%?





Miller E. – Annals of Oncology, 2020

HGSOC molecular diagnostics and PARPi



Ngoi NYL – Annals of Oncology, 2021

ESMO recommendations 2024

- Somatic BRCA1/2 assessment at the time of diagnosis of non-miconous OC
- HRD score assessment in BRCA1/2 WT cases
- Germline assessment for mutBRCA1/2 cases
- NO somatic assessment for other HR genes
- Germline assessment of HR genes in case of suspected familiarity

ESMO NGS Recommendations 2024

ESCAT

ESMO Scale for Clinical Actionability of Molecular Targets

ESMO GOOD SCIENCE BETTER MEDICINE BEST PRACTICE

ANNALS OF ONCOLOGY driving innovation in oncology

SPECIAL ARTICLE

Recommendations for the use of next-generation sequencing (NGS) for patients with advanced cancer in 2024: a report from the ESMO Precision Medicine Working Group

M. F. Mosele[1,2], C. B. Westphalen[3], A. Stenzinger[4], F. Barlesi[1,2,5], A. Bayle[5,6,7,8], I. Bièche[9], J. Bonastre[7,8], E. Castro[10], R. Dienstmann[11,12,13], A. Krämer[14,15], A. M. Czarnecka[16,17], F. Meric-Bernstam[18], S. Michiels[7,8], R. Miller[19,20], N. Normanno[21], J. Reis-Filho[22†], J. Remon[2], M. Robson[23], E. Rouleau[24], A. Scarpa[25], C. Serrano[11], J. Mateo[11] & F. André[1,2,5*]

Table 7. List of genomic alterations level I/II according to ESCAT in advanced ovarian cancer

Gene/Signature[a]	Alteration	Estimated prevalence	ESCAT score	Drug class matched	References
BRCA1/2	Germline pathogenic/likely pathogenic variants Somatic pathogenic/likely pathogenic variants	15%-17% 5%-7%	IA	PARP inhibitors	Bell et al., *Nature* 2011[109] Mirza et al., *N Engl J Med* 2016[110] Coleman et al., *Lancet* 2017[111] Pujade-Lauraine et al., *Lancet Oncol* 2017[112] Moore et al., *N Engl J Med* 2018[113] González-Martin et al., *N Engl J Med* 2019[114] Ray-Coquard et al., *N Engl J Med* 2019[115] DiSilvestro et al., *J Clin Oncol* 2023[116] González-Martin et al., *Ann Oncol* 2023[117]
HRD[a]	HRD	50% high-grade serous ovarian cancer	IA	PARP inhibitors	Mirza et al., *N Engl J Med* 2016[110] Coleman et al., *Lancet* 2017[111] González-Martin et al., *N Engl J Med* 2019[114] Ray-Coquard et al., *N Engl J Med* 2019[115] González-Martin et al., *Ann Oncol* 2023[117]

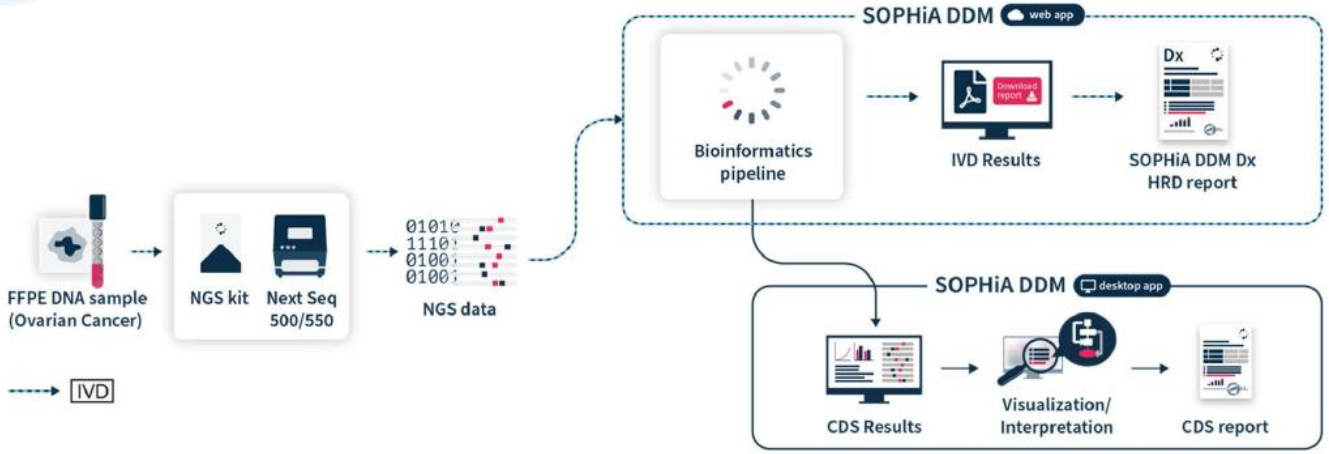
ESCAT, ESMO Scale for Clinical Actionability of molecular Targets; HRD, homologous recombination deficiency; PARP, poly (ADP-ribose) polymerase.
[a]Signature.

Current Laboratory Practice - Somatic

Diagnosis of HGSOC
UO Anatomia Patologica e Biologia Molecolare

Somatic Molecular Testing
UO Anatomia Patologica e Biologia Molecolare

Clinical Assessment / Pre-Germline Test Genetic Counselling
UO Oncologia / UO Ginecologia /UOS Genetica Medica



- SNVs
- InDels
- Splice Variants
- HRD Status
- CNVs

28 HRR Genes

BRCA1, BRCA2, PTEN, PALB2, BRIP1, RAD51C, RAD51D, RAD51B, ATM, MRE11, PPP2R2A, CHEK2, CHEK1, AKT1, RAD54L, NBN, BARD1, FANCL, FANCA, ESR1, PIK3CA, CCNE1, FANCD2, CDK12, FGFR3, TP53, FGFR2, FGFR1.

HRD Testing – A New Coplexity Level

BRCA Status NEGATIVE Edit

Proposed BRCA status
Negative

Clinically relevant BRCA variants Edit

No variant detected

General Information **Insertion**
BRIP1(NM_032043.3):c.1018_1019insCT
p.(Leu340ProfsTer9)

PharmGKB
No data available

Germline Classification
Pathogenic
13 points = 13 P - 0 B

Genes
BRIP1

Transcripts
NM_032043.3 - frameshift
MANE Select

ClinVar Pathogenic/Likely Pathogenic ★★☆☆
4 | 1

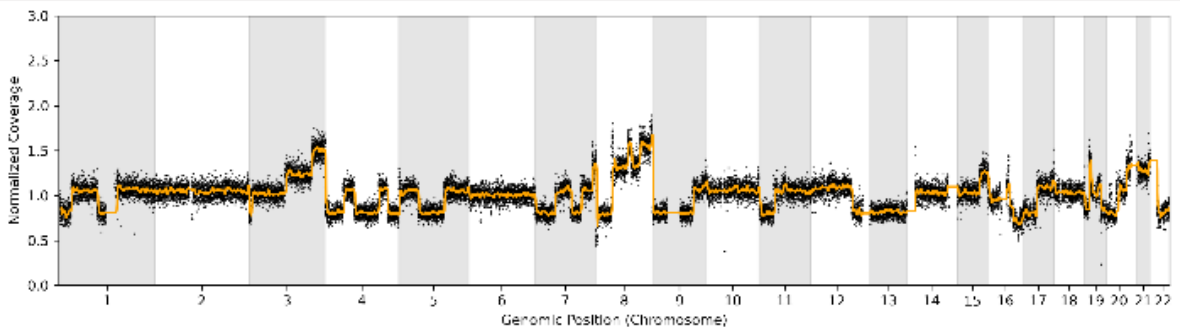
Genomic integrity status NEGATIVE Edit

Proposed GI status (For Ovarian cancers)
Negative

Genomic integrity index
-2.9

QA Status
High

Low-pass WGS data Download full report



CASE 1 | **BRCA neg** | **HRD neg** | **HRR PV**

- Therapy 
- Genetic counselling 

SCREENING GENES SNVs/INDELs CNVs FUSIONS WARNINGS

Interpretat
HRD_v1

Variant List - sorted by: ref

save column preferences

SOPHiA Filters

P...	In ...	BRCA Score	BRCA Pathogenicity	Actionability	T...	Gene	Coding consequence	c.DNA	Protein	Depth	VF%	Genome p...	ref	alt	Chr...	Exon ID	Positi...
A				T1 T2 T3 T4 D P	INDEL	BRIP1	frameshift	c.1018_1019insCT	p.(Leu340Profs*9)	5927	73.9	59878735	A	AAG	17	8	101

HRD Testing – A New Coplexity Level

BRCA Status NEGATIVE Edit

Proposed BRCA status
Negative

Clinically relevant BRCA variants Edit

No variant detected

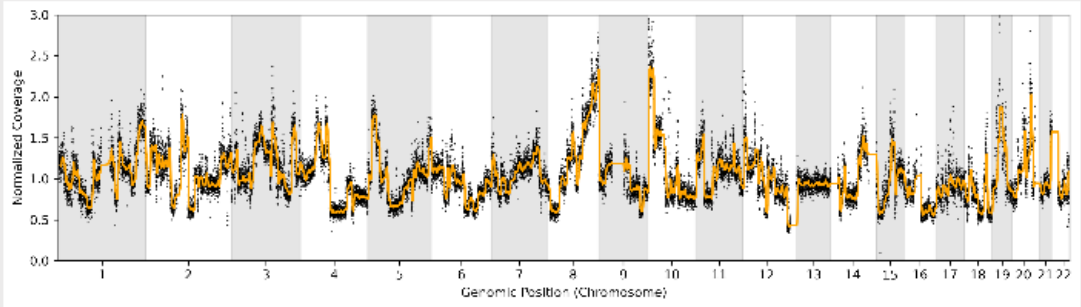
Genomic integrity status POSITIVE Edit

Proposed GI status (For Ovarian cancers)
Positive

Genomic integrity index
11.1

QA Status
High

Low-pass WGS data Download full report



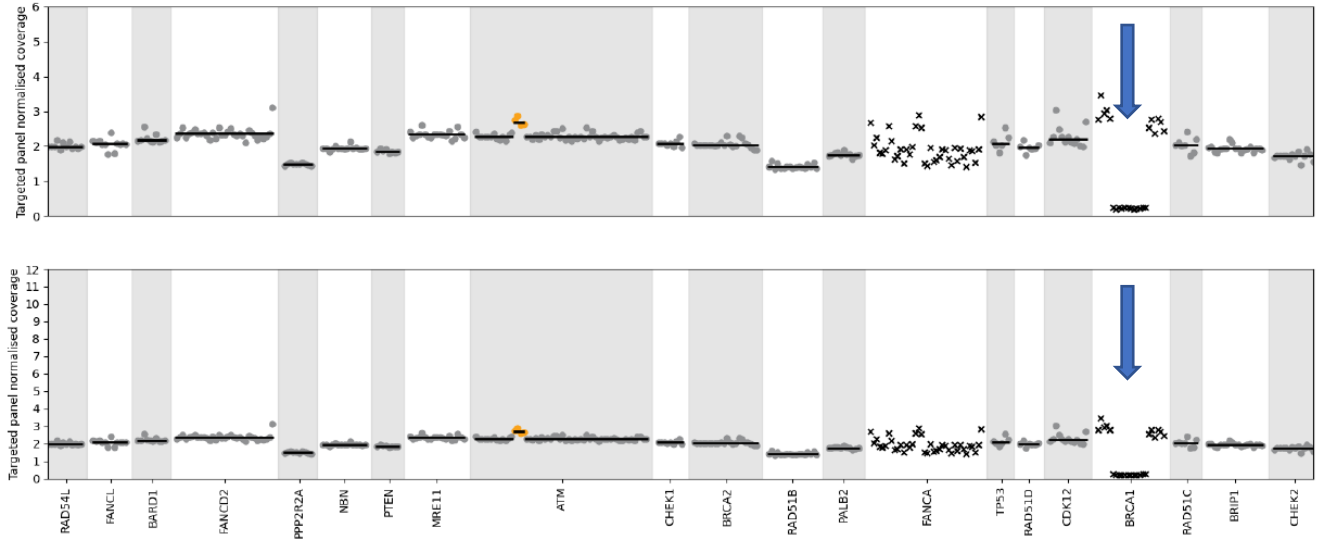
1. CNV ANALYSIS REPORT

SOPHiA GENETICS™

1.6 Results for sample 459668-0-S4

Gene	Gene QA status	Gene-level status	Exon-level status	Segment regions	Segment coverage level	Segment level interpretation
ATM	Pass	Exon-level event	Exon gain	ex2-3 - ex15	2.3	Normal
				ex16 - ex19-20	2.7	Exon gain
				ex21-22 - ex63	2.3	Normal
FANCA	Rejected	Rejected	Rejected	-	-	-
BRCA1	Rejected	Rejected	Rejected	-	-	-
CCNE1	Pass	Gain	NA	ex2 - ex11	4.1	Gain

BRCA1 Ex8-18DEL



CASE 2	BRCA neg	HRD POS	HRR neg

- Therapy
- Genetic counselling
- High-risk families loss!

Hereditary OC

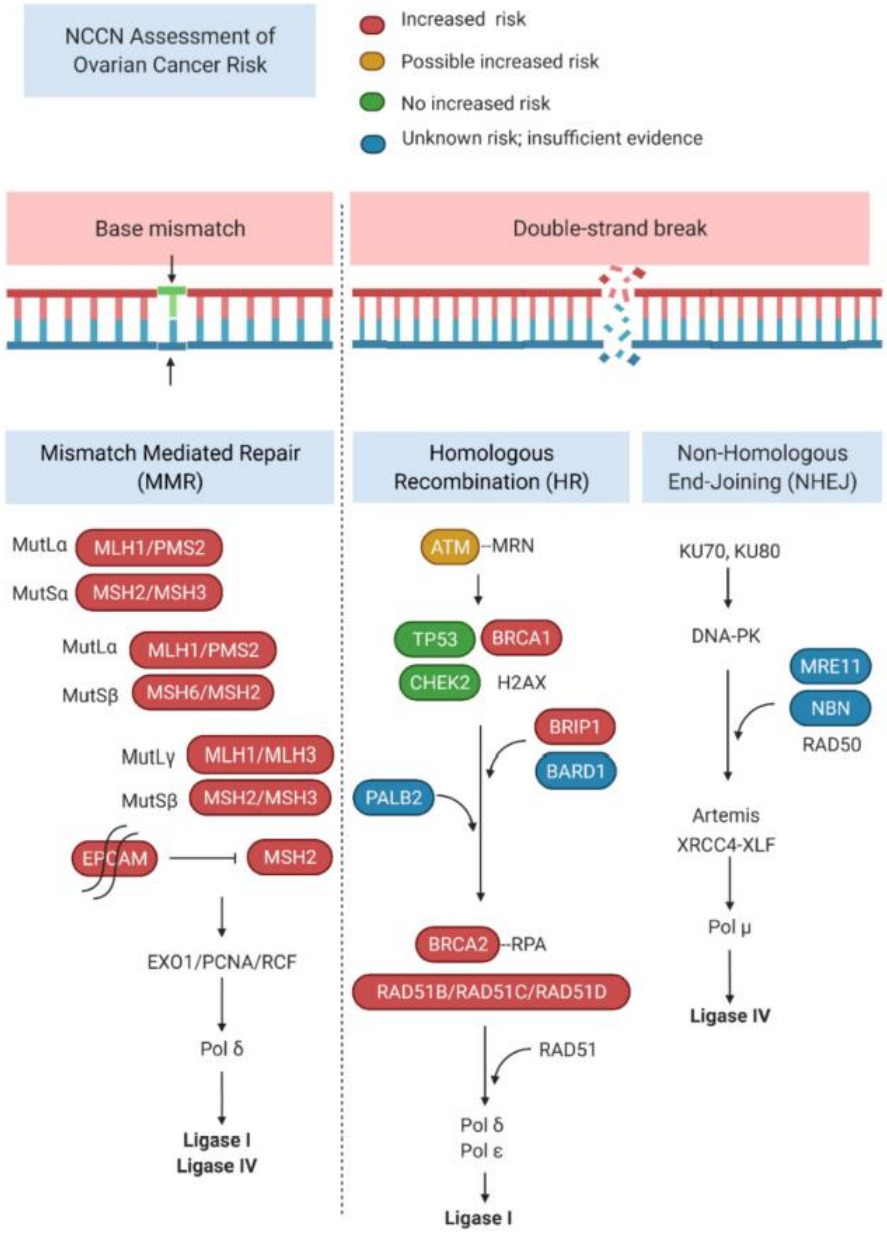
25% OC patients exhibit BC/OC family history. Of those:

- 40% are BRCA1/2 PV carriers
- 25% are carriers of PV in other genes

DSB – HR - HBOC Syndrome

- HIGH RISK genes: BRCA1 (~40%), BRCA2 (~18%)
- MODERATE RISK genes: RAD51C/D, PALB2, BRIP1, ATM.

- **MMR** (Mismatch Repair) – Lynch Syndrome
- HIGH RISK genes: MLH1 (~11%), MSH2-EPCAM (~17%), MSH6 (~10%), PMS2 (?).



Samuel et al. - Cells, 2022

Hereditary OC

NOTA 9. CRITERI DI "SOSPETTA EREDITARIETÀ" PER L'INVIO ALLA CONSULENZA GENETICA ONCOLOGICA TRADIZIONALE

- Per la valutazione dei criteri di sospetta ereditarietà è fondamentale che il clinico faccia riferimento sia alla storia personale della paziente, sia alla storia familiare di malattia.

Numero di soggetti affetti presenti in famiglia	Condizione
1 caso dei seguenti tumori:	Tumore della mammella in età ≤ 40 anni[a]
	2 tumori primari della mammella in età ≤ 50[b]
	Tumore della mammella maschile
	Tumore della mammella e dell'ovaio[c] nella stessa paziente
	Tumore della mammella di tipo "triplo negativo" (qualsiasi età)[a]
	Tumore epiteliale ovarico non mucinoso e non borderline, carcinoma della tuba o tumore primitivo peritoneale
2 casi in una qualsiasi combinazione dei seguenti tumori[d]:	Tumore della mammella ≤ 50 anni
	2 tumori primari della mammella[b]
	Tumore epiteliale ovarico non mucinoso e non borderline, carcinoma della tuba o tumore primitivo peritoneale
	Adenocarcinoma del pancreas
	Tumore della prostata[e]
3 casi in una qualsiasi combinazione dei seguenti tumori:	Tumore della mammella (qualsiasi età)
	Tumore della prostata ≤ 60 anni
	Adenocarcinoma del pancreas
	Tumore dell'ovaio
-	Presenza nella famiglia di VP nota

PDTA Tumori Eredo-Familiari di Mammella e Ovaio – ROV 2024

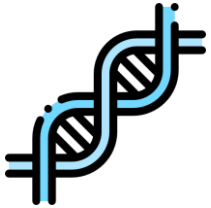
Cancer Type and Specific Population	More Strongly Recommended (higher relative risk of cancer or highly actionable)	Less Strongly Recommended (moderate relative risk of cancer or potential impact for therapy/change in medical management)
Ovarian cancer (epithelial)	*BRCA1, BRCA2, BRIP1, EPCAM, MLH1, MSH2, MSH6, PALB2, PMS2, RAD51C, RAD51D*	*ATM*

Tung N - JCO, 2024

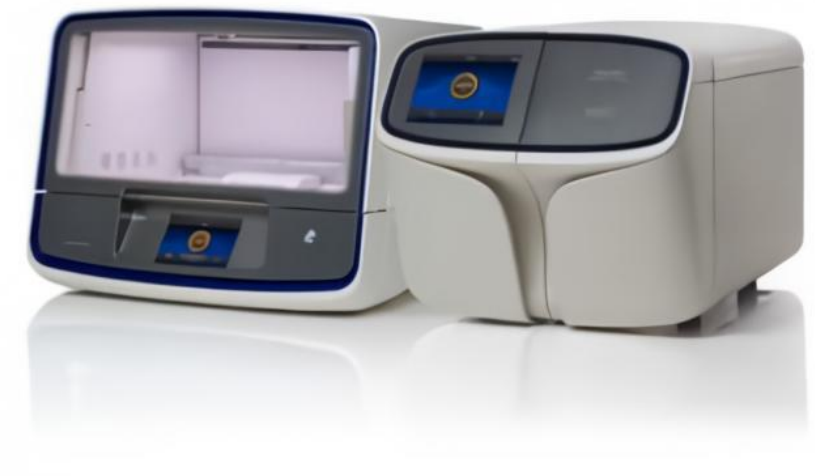
Current Laboratory Practice - Germline

Pre-Test Genetic Counselling
UOS Genetica Medica

Germline Molecular Testing
UO Anatomia Patologica e Biologia Molecolare



Post-Test Genetic Counselling
UOS Genetica Medica



- SNVs
- InDels
- CNVs
- Splice Variants

EMQN
The European Molecular Genetics Quality Network

SUMMARY OF YOUR PERFORMANCE IN THIS SCHEME

Assessment Category	Performance* (mean score)
Genotyping	2.00
Interpretation	2.00
Patient Identifiers and Clerical Accuracy	2.00
Scheme result (SATISFACTORY or POOR)	SATISFACTORY

ONCOGENETIC Panel	Geni analizzati
Mammella	*BRCA1, BRCA2, TP53, PTEN, PALB2, ATM, CHEK2, STK11, CDH1, BARD1*
Ovaio	*BRCA1, BRCA2, EPCAM, MLH1, MSH2, MSH6, PMS2, PALB2, BRIP1, RAD51C, RAD51D,ATM*
Colon-retto/poliposi	*APC, BMPR1A, GREM1, EPCAM, MLH1, MSH2, MSH3, MSH6, MUTYH, NTHL1, PMS2, POLD1, POLE, PTEN, RNF43, RPS20, SMAD4, STK11*
Rene	*c-MET, VHL, FH, PTEN, FLCN, TSC1, TSC2, SDHA, SDHB, SDHC, SDHD, BAP1, MITF*
Pancreas	*CDKN2A, STK11, BRCA1, BRCA2, PALB2, EPCAM, MLH1, MSH2, MSH6, PMS2, ATM, TP53*
Iperparatiroidismo primario	*MEN1, CDKN1B, RET, CDC73, CASR, GNA11, AP2S1, CDKN1A, CDKN2B, CDKN2C*
Melanoma familiare	*CDKN2A, CDK4, POT1, BAP1, MITF, TERT*
Stomaco	*CDH1, EPCAM, MSH2, MSH6, MLH1, PMS2, EPCAM, PTEN, STK11, TP53, APC*
Prostata	*BRCA2, BRCA1, ATM, CHEK2, PALB2, BRIP1, HOXB13, EPCAM*, MLH1*, MSH2, MSH6, PMS2*
Feocromocitoma/paragangl ioma	*FH, NF1, RET, SDHB, SDHD, VHL, MEN1, SDHA, SDHAF2, SDHC, TMEM127, MAX*

Take Home Message

- Genomic Instability is the hallmark HGSOC
- ESCAT1 biomarkers should be tested at the same time
- HRD data interpretation may be challenging
- HRD testing could be the doorway to germline testing
- Genetic Counselling and germline testing are common practice for OC patients

Thank You!

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