Biomarqueurs moléculaires dans les cancers de l’ovaire

Juin 2017

Alexandra Leary, MD, PhD

Gynecology Unit
& Translational Research Laboratory INSERM U981
Gustave Roussy Cancer Center
Liens d’intérêt

• Board : Clovis, AstraZeneca
• Transports : AstraZeneca
• Investigator in clinical trial sponsored by : Clovis, AstraZeneca
Outline

• **Predictive biomarkers in High Grade Serous Ovarian Cancer (HGSOC)**
  – Homologous recombination deficiency for PARP inhibitors: ready for prime-time?
    • BRCA mutations: germline and somatic, BRCA hypermethylation
    • Mutations or SCNAs in HR genes
    • HRD scar
  - When do we need this HRD information? Relapse?
  - Diagnosis?
  - Resistance mutations

• **Predictive biomarkers in other rare epithelial OC**

• **Diagnostic value** of genomic alterations in rare non-epithelial ovarian cancers

• **Endometrial Cancers**
**Molecular subtypes of epithelial ovarian cancer**

**High grade serous ovarian cancer (75%)**
- High Ki67
- Genomically unstable
- Chemosensitive but poor prognosis
- Universal TP53 mutation – few other mutations
- Associated with BRCA1/2 germline mutations (12-15%)
- Genomic homology with triple negative BC

**Rare subtypes (25-30%)**
- Histologic similarities with other primary tumors
- Chemotherapy resistance
- Frequent oncogenic mutations

**Subtypes of rare OC**

<table>
<thead>
<tr>
<th>Invasive histology</th>
<th>Histological similarities</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low grade serous</td>
<td></td>
</tr>
<tr>
<td>Mucinous</td>
<td>Colorectal cancer</td>
</tr>
<tr>
<td>Endometrioid</td>
<td>Endometrial cancer</td>
</tr>
<tr>
<td>Clear cell</td>
<td>Renal cell cancer</td>
</tr>
<tr>
<td>Transitional cell/Brenners</td>
<td>Urothelial tumors</td>
</tr>
</tbody>
</table>

12-15% of HGSOC have DNA repair deficiency via homologous recombination from germline inactivating mutation in BRCA

Results in genomic instability AND explains chemo-sensitivity

Reis-Filho et al 2011 Cell Cycle
High Grade Serous Ovarian Cancer (HGSOC): DNA repair deficiency

30% of HGSOC demonstrate alterations in BRCA1 or BRCA2

TCGA, Nature 2011

TCGA on >300 HGSOC
Concept of synthetic lethality of PARP inhibitors in BRCA mutated tumors

**Synthetic lethality:** HR defects renders BRCA mutated tumors ‘addicted’ or essentially dependent on other DNA repair pathways. Inhibition of PARP becomes synthetically lethal in the context of an inactivating BRCA mutation.

Iglehart, NEJM 2009

Dr. A. Leary 15 juin 2017 - Journée GFCO 2017
Olaparib Maintenance Therapy in Platinum-Sensitive Relapsed Ovarian Cancer

Greatest benefit of olaparib in BRCA mutated subset (germline or somatic)

![Graph showing progression-free survival of patients with BRCA mutation](image)

**NEJM, Ledermann et al 2012**

**Dr.A.Leary 15 juin 2017 - Journée GFCO 2017**
EMA approval for olaparib

- In patients with platinum-sensitive relapsed,
- High grade serous ovarian, fallopian tube or primary peritoneal cancer,
- Associated with a deleterious BRCA1 or 2 mutation (germline or somatic),
- Who are in response (PR or CR) to platinum based chemotherapy

Olaparib maintenance until progression or intolerance
EMA approval for olaparib

• In patients with platinum-sensitive relapsed,

• High grade serous ovarian, fallopian tube or primary peritoneal cancer,

• Associated with a deleterious BRCA1 or 2 mutation (germline or somatic),

• Who are in response (PR or CR) to platinum based chemotherapy

Olaparib maintenance until progression or intolerance

First targeted therapy associated with a genomic predictive predictive biomarker approved in gynecological cancers
PARP inhibitors beyond BRCA mutations in OC?
50% of HGSOC dysfunctional DNA repair homologous recombination (HR) pathway

30% BRCA loss
- via germline BRCA-M+
- somatic BRCA-M+
- BRCA hypermethylation

Another 20% with dysfunctional HR-mediated DNA repair:
- EMSY amp 5-17%
- RAD51 loss 3-5%
- ATM/ATR M+: 3%
  - Fanc M+: 5%
- PTEN loss 7%

TCGA, Nature 2011
Germline and somatic alterations in non-BRCA HR genes in OC

Identified in 1/3

Mainly M+ BRCA, other alterations are rare

Pennington et al

Dr.A.Leary 15 juin 2017 - Journée GFCO 2017
Identifying BRCA-WT OC with HR deficiency: One approach: SCNAs or mutations in HR genes

<table>
<thead>
<tr>
<th>Gene symbol</th>
<th>% mutated/deleted in HGSOC*</th>
<th>HR pathway subgroups</th>
</tr>
</thead>
<tbody>
<tr>
<td>BRCA1</td>
<td>13.4%</td>
<td>BRCA genes</td>
</tr>
<tr>
<td>BRCA2</td>
<td>10.5%</td>
<td>BRCA genes</td>
</tr>
<tr>
<td></td>
<td>23.9%</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Gene symbol</th>
<th>% mutated/deleted in HGSOC*</th>
<th>HR pathway subgroups</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATM</td>
<td>2.6%</td>
<td>DNA damage response genes</td>
</tr>
<tr>
<td>ATR</td>
<td>0.7%</td>
<td>DNA damage response genes</td>
</tr>
<tr>
<td>CHEK1</td>
<td>1.1%</td>
<td>DNA damage response genes</td>
</tr>
<tr>
<td>CHEK2</td>
<td>1.8%</td>
<td>DNA damage response genes</td>
</tr>
<tr>
<td>FANCA</td>
<td>5.0%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>FANCC</td>
<td>0.9%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>FANCD2</td>
<td>0.2%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>FANCE</td>
<td>0.2%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>FANCF</td>
<td>0.4%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>FANCG</td>
<td>0.2%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>FANCI</td>
<td>0.7%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>FANCL</td>
<td>0.4%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>FANCM</td>
<td>1.4%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>PALB2</td>
<td>1.1%</td>
<td>Fanconi Anemia genes</td>
</tr>
<tr>
<td>RAD50</td>
<td>1.5%</td>
<td>RAD genes</td>
</tr>
<tr>
<td>RAD51</td>
<td>3.5%</td>
<td>RAD genes</td>
</tr>
<tr>
<td>RAD51B</td>
<td>0.2%</td>
<td>RAD genes</td>
</tr>
<tr>
<td>RAD51C</td>
<td>0.0%</td>
<td>RAD genes</td>
</tr>
<tr>
<td>RAD51D</td>
<td>0.9%</td>
<td>RAD genes</td>
</tr>
<tr>
<td>RAD52</td>
<td>0.2%</td>
<td>RAD genes</td>
</tr>
<tr>
<td>RAD54L</td>
<td>0.9%</td>
<td>RAD genes</td>
</tr>
</tbody>
</table>

*Based on most up to date NGS data of 456 HGSOC tumors in TCGA (Apr 2013)

Note: a small subset of mutations and deletions co-occur, resulting in slightly lower cumulative frequencies for BRCA and nHRD
Are alterations in non-BRCA genes equally predictive of a response to PARPi?

Differential sensitivity to PARP inhibitor rucaparib (siRNA knockdown)

HRD Genotype Approach

siRNA knockdown of HR pathway genes renders differential sensitivity to rucaparib in ovarian cancer cell lines

- Rucaparib sensitivity was tested in ovarian cancer cell line OVCAR-3 with 29 homologous recombination (HR) pathway genes individually knocked down using siRNA
- Similar findings were observed in SK-OV-3, UWB1.289+BRCA1, and OAW-28 cell lines

Rucaparib IC50 fold change after knockdown in OVCAR-3

I McNeish, ESMO, 2014
BRCA hypermethylation does NOT have the same prognostic impact as BRCA mutation

BRCA hypermethylation does not have prognostic value in platinum treated OC

Valid surrogate for HRD?

TCGA, Nature 2011

Dr.A.Leary 15 juin 2017 - Journée GFCO 2017
Loss of heterozygosity (LOH) score: deficiency in homologous recombination (HRD)

BRCA\textsuperscript{mut}

HRD/BRCA-like

BRCA\textsuperscript{WT}

HR competent

High LOH score reflects HRD regardless of the cause BRCA-like \(\Rightarrow\) predict response to PARPi?
ARIEL studies*: Identify BRCA wild-type tumors with homologous recombination deficiency (HRD) that may benefit from PARP inhibitors

**ARIEL 2***

Metastatic Ovarian Cancer (mainly BRCA WT) → Biopsy → Tx PARPi

- **Responders**
- **Non-Responders**

**HRD genotype**
- Mutations in other genes in HR pathway

**HRD phenotype**
- 'Scarring' of the DNA suggesting HRD – LOH score

**HR deficiency signature** predictive of response to PARPi
### Do mutations in non-BRCA HR genes predict benefit from PARPi?

<table>
<thead>
<tr>
<th>HR-pathway gene</th>
<th>Genetic alteration type</th>
<th>Germline/somatic inference</th>
<th>HRD molecular subgroup</th>
<th>RECIST response</th>
<th>CA-125 response</th>
</tr>
</thead>
<tbody>
<tr>
<td>NBN</td>
<td>Truncation</td>
<td>Germline</td>
<td>Biomarker-negative</td>
<td>Partial Response</td>
<td>Yes</td>
</tr>
<tr>
<td>RAD51C</td>
<td>Truncation</td>
<td>Germline</td>
<td>BRCA-like</td>
<td>Partial Response</td>
<td>Yes</td>
</tr>
<tr>
<td>RAD51C</td>
<td>Homozygous Del</td>
<td>Somatic</td>
<td>BRCA-like</td>
<td>Partial Response</td>
<td>Yes</td>
</tr>
<tr>
<td>RAD51C</td>
<td>Splice</td>
<td>Germline</td>
<td>BRCA-like</td>
<td>Partial Response</td>
<td>Yes</td>
</tr>
<tr>
<td>RAD51C</td>
<td>Splice</td>
<td>Germline</td>
<td>BRCA-like</td>
<td>Stable Disease</td>
<td>Yes</td>
</tr>
<tr>
<td>ATM</td>
<td>Homozygous Del</td>
<td>Somatic</td>
<td>Indeterminate</td>
<td>Stable Disease</td>
<td>Yes</td>
</tr>
<tr>
<td>RAD51L3</td>
<td>Truncation</td>
<td>Indeterminate</td>
<td>BRCA-like</td>
<td>Stable Disease</td>
<td>Yes</td>
</tr>
<tr>
<td>BRIP1</td>
<td>Splice</td>
<td>Germline</td>
<td>Biomarker-negative</td>
<td>Stable Disease</td>
<td>No</td>
</tr>
<tr>
<td>BRIP1</td>
<td>Truncation</td>
<td>Germline</td>
<td>Biomarker-negative</td>
<td>Stable Disease</td>
<td>No</td>
</tr>
<tr>
<td>CHEK2</td>
<td>Splice</td>
<td>Indeterminate</td>
<td>Biomarker-negative</td>
<td>Stable Disease</td>
<td>No</td>
</tr>
<tr>
<td>CHEK2</td>
<td>Truncation</td>
<td>Germline</td>
<td>BRCA-like</td>
<td>Stable Disease</td>
<td>No</td>
</tr>
<tr>
<td>RAD51L1</td>
<td>Truncation</td>
<td>Indeterminate</td>
<td>Biomarker-negative</td>
<td>Stable Disease</td>
<td>No</td>
</tr>
<tr>
<td>NBN</td>
<td>Truncation</td>
<td>Germline</td>
<td>Indeterminate</td>
<td>Stable Disease</td>
<td>Not evaluable</td>
</tr>
<tr>
<td>RAD54L</td>
<td>Truncation</td>
<td>Somatic (subclonal)</td>
<td>Biomarker-negative</td>
<td>Stable Disease</td>
<td>Not evaluable</td>
</tr>
<tr>
<td>FANCA</td>
<td>Homozygous Del</td>
<td>Somatic</td>
<td>BRCA-like</td>
<td>Stable Disease</td>
<td>Not evaluable</td>
</tr>
<tr>
<td>FANCI</td>
<td>Truncation</td>
<td>Germline</td>
<td>Biomarker-negative</td>
<td>Progressive Disease</td>
<td>No</td>
</tr>
<tr>
<td>ATM</td>
<td>Truncation</td>
<td>Somatic</td>
<td>Indeterminate</td>
<td>Not evaluable</td>
<td>Not evaluable</td>
</tr>
</tbody>
</table>
DOES THE HRD SCAR OR LOH SCORE PREDICT BENEFIT FROM PARPI?
Rucaparib in relapsed, platinum-sensitive high-grade ovarian carcinoma (ARIEL2 Part 1): an international, multicentre, open-label, phase 2 trial

Elizabeth M Swisher*, Kevin K Lin*, Arrait M Ozs, Clare L Scott, Heidi Giordano, James Sun, Gottfried E Konecny, Robert L Coleman, Anna V Tinker, David M O’Malley, Rebecca S Kristeleit, Ling Ma, Katherine M Bell-McGuira, James D Brenton, Janiel M Craig, Ana Ouknine, Isabelle Ray-Coquard, Maria Harrell, Uchina Mann, Scott H Kaufmann, Anne Hoque, Alexandra Leary, Thomas C Harding, Sandra Goble, Lara Maione, Jeff Isaacson, Andrew R Allen, Lindsey Rolfe, Roman Yelensky, Milch Raponi, Jain A McNeish*

Non-BRCA mutated HRD tumors (high LOH) respond to PARPi

Although less so than BRCA mutated OC

Dr. A. Leary 15 juin 2017 - Journée GFCO 2017
Niraparib Maintenance Therapy in Platinum-Sensitive, Recurrent Ovarian Cancer

M.R. Mirza, B.J. Monk, J. Herrstedt, A.M. Oza, S. Mahner, A. Redondo,

Platinum-Sensitive Recurrent High Grade Serous Ovarian Cancer

Treat until Progression of Disease

Niraparib 300 mg once daily

Placebo

Non-gBRCAmut

Treat until Progression of Disease

Placebo

Response to Platinum Treatment

2:1 Randomization

Niraparib 300 mg once daily

gBRCAmut

2:1 Randomization

Platinum-Sensitive Recurrent High Grade Serous Ovarian Cancer

Treatment with 4-6 Cycles of Platinum-based Therapy

Response to Platinum Treatment

2:1 Randomization

Niraparib 300 mg once daily
Progression-free Survival: gBRCAmut

<table>
<thead>
<tr>
<th>Treatment</th>
<th>PFS Median (95% CI)</th>
<th>Hazard Ratio (95% CI)</th>
<th>p-value</th>
<th>% of Patients without Progression or Death</th>
</tr>
</thead>
<tbody>
<tr>
<td>Niraparib (N=138)</td>
<td>21.0 (12.9, NR)</td>
<td>0.27 (0.173, 0.410)</td>
<td>&lt;0.0001</td>
<td>62% 50%</td>
</tr>
<tr>
<td>Placebo (N=65)</td>
<td>5.5 (3.8, 7.2)</td>
<td></td>
<td>p&lt;0.0001</td>
<td>16% 16%</td>
</tr>
</tbody>
</table>

Bénéfice de > 15 mois

Mirza et al 2016

Dr. A. Leary 15 juin 2017 - Journée GFCO 2017
### Progression-free Survival: non-gBRCAmut

<table>
<thead>
<tr>
<th>Treatment</th>
<th>PFS Median (95% CI) (Months)</th>
<th>Hazard Ratio (95% CI)</th>
<th>p-value</th>
<th>% of Patients without Progression or Death</th>
</tr>
</thead>
<tbody>
<tr>
<td>Niraparib (N=234)</td>
<td>9.3 (7.2, 11.2)</td>
<td>0.45 (0.338, 0.607)</td>
<td>$p&lt;0.0001$</td>
<td>41% 30%</td>
</tr>
<tr>
<td>Placebo (N=116)</td>
<td>3.9 (3.7, 5.5)</td>
<td></td>
<td></td>
<td>14% 12%</td>
</tr>
</tbody>
</table>

Can we identify the subset of BRCA WT OC that benefits the most from PARPi?

Mirza et al 2016
**Exploratory Analysis: PFS in Subgroups of Non-gBRCAmut Cohort**

### HRD-positive

<table>
<thead>
<tr>
<th></th>
<th>Treatme nt</th>
<th>PFS Median (95% CI) (Months)</th>
<th>Hazard Ratio (95% CI)</th>
<th>% of Patients without Progression or Death</th>
<th>12 mo</th>
<th>18 mo</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>sBRCAmut</strong></td>
<td>Niraparib  (N=35)</td>
<td>20.9 (9.7, NR)</td>
<td>0.27 (0.081, 0.903)</td>
<td>p=0.0248</td>
<td>62%</td>
<td>52%</td>
</tr>
<tr>
<td></td>
<td>Placebo   (N=12)</td>
<td>11.0 (2.0, NR)</td>
<td>p=0.00248</td>
<td></td>
<td>19%</td>
<td>19%</td>
</tr>
<tr>
<td><strong>BRCAwt</strong></td>
<td>Niraparib  (N=71)</td>
<td>9.3 (5.8, 15.4)</td>
<td>0.38 (0.231, 0.628)</td>
<td>p=0.0001</td>
<td>45%</td>
<td>27%</td>
</tr>
<tr>
<td></td>
<td>Placebo   (N=44)</td>
<td>3.7 (3.3, 5.6)</td>
<td>p=0.0001</td>
<td></td>
<td>11%</td>
<td>6%</td>
</tr>
</tbody>
</table>

### HRD-negative

<table>
<thead>
<tr>
<th></th>
<th>Treatme nt</th>
<th>PFS Median (95% CI) (Months)</th>
<th>Hazard Ratio (95% CI)</th>
<th>% of Patients without Progression or Death</th>
<th>12 mo</th>
<th>18 mo</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>sBRCAmut</strong></td>
<td>Niraparib  (N=92)</td>
<td>6.9 (5.6, 9.6)</td>
<td>0.58 (0.361, 0.922)</td>
<td>p=0.0226</td>
<td>27%</td>
<td>19%</td>
</tr>
<tr>
<td></td>
<td>Placebo   (N=42)</td>
<td>3.8 (3.7, 5.6)</td>
<td>p=0.0226</td>
<td></td>
<td>7%</td>
<td>7%</td>
</tr>
</tbody>
</table>

**Mirza et al 2016**
Authors Conclusions

- Niraparib significantly improved PFS in patients with platinum-sensitive recurrent ovarian cancer, regardless of *BRCA* mutation or HRD status
  - gBRCAmut: HR 0.27
  - Non-gBRCAmut: HR 0.45
  - Non-gBRCAmut HRD-positive: HR 0.38
  - Non-gBRCAmut HRD-negative (exploratory): HR 0.58

- These landmark results warrant niraparib maintenance therapy to whole study population

Perhaps expensive genomic characterization is not needed, platinum sensitivity is valid surrogate predictive marker for PARP inhibitor sensitivity?
Genomic profiling of HGSOC

Homologous recombination deficiency (HRD) in 50% of HGSOC:

→ First targeted therapy in HGSOC associated with a genomic biomarker – germline or somatic BRCA mutations (RR to PARPi=80%)

→ Can we identify the subset of BRCA WT patients that could benefit from PARP inhibitors?
  • High LOH score?
  • Mutations in non-BRCA HR genes?
  • Platinum sensitivity?
Homologous recombination deficiency (HRD) in 50% of HGSOC:

→ First targeted therapy in HGSOC associated with a genomic biomarker – germline or somatic BRCA mutations (RR to PARPi=80%)

→ Can we identify the subset of BRCA WT patients that could benefit from PARP inhibitors?
  • High LOH score?
  • Mutations in non-BRCA HR genes?
  • Platinum sensitivity?

When do we need information on BRCA mutations or HRD?
  At relapse?
  At diagnosis?
PAOLA1

Platine, Avastin and Olaparib in first line advanced high grade ovarian carcinoma patients

- Phase III randomized, placebo-controlled, double-blind, multicenter
- Olaparib tablets administered at 300 mg daily for up to 15 cycles.

First-line surgery and chemotherapy (allowed: dose-dense, IP, neo adjuvant) + bevacizumab*

If not PD

Randomize

Bevacizumab 15 mg/kg + olaparib 15 cycles

Observation (to PD)

Survival follow-up (post-PD)

Bevacizumab 15 mg/kg + Placebo 15 cycles

*At least 3 cycles

Estimated median months from diagnosis to randomization (7 months)
RESISTANCE MUTATIONS TO PARP INHIBITORS?
Secondary Somatic Mutations Restoring BRCA1/2 Predict Chemotherapy Resistance in Hereditary Ovarian Carcinomas
Barbara Norquist, Kaitlyn A. Wurz, Christopher C. Pennil, Rochelle Garcia, Jenny Gross, Wataru Sakai, Beth Y. Karlan, Toshiyasu Taniguchi, and Elizabeth M. Swisher

45 ptes BRCA mutated and relapse biopsied → To identify reversion mutations restoring BRCA WT function

45% of patients biopsied for a platinum resistant relapse showed acquired reversion mutation restoring BRCA wild-type function
Acquired reversion BRCA mutations associated with platinum and PARP inhibitor resistance

Whole-genome characterization of chemoresistant ovarian cancer

Matched analysis of primary tumor and relapse or post-mortem biopsy

5 BRCA mutated patients showed a reversion mutation restoring WT BRCA at relapse

One post mortem analysis:

14 different reversion BRCA mutations!

Was resistant to platinum and PARP inhibitor
HGSOC: 50% genomic alterations associated with HRD, how about the other half?

• Mutually exclusive from HRD
• Associated with platinum resistance?

CCNE1 amplification poor prognostic marker
Genomic profiling of HGSOC

Homologous recombination deficiency (HRD) in 50% of HGSOC:

→ First targeted therapy in HGSOC associated with a genomic biomarker – germline or somatic BRCA mutations (RR to PARPi=80%)

→ Can we identify the subset of BRCA WT patients that could benefit from PARP inhibitors?
  • High LOH score?
  • Mutations in non-BRCA HR genes?
  • Platinum sensitivity?

→ May soon need information regarding HRD status sooner, at diagnosis

→ Need to identify markers showing restoration of HR with resistance

→ Useful molecular markers in the non-HRD subset (50% of HGSOC)
Molecular subtypes of epithelial ovarian cancer

High grade serous ovarian cancer (75%)
- High Ki67
- Genomically unstable
- Chemosensitive but poor prognosis
- Universal TP53 mutation – few other mutations
- Associated with BRCA1/2 germline mutations (12-15%)
- Genomic homology with triple negative BC

Rare subtypes (25-30%)
- Histologic similarities with other primary tumors
- Chemotherapy resistance
- Frequent oncogenic mutations

Subtypes of rare OC

<table>
<thead>
<tr>
<th>Invasive histology</th>
<th>Histological similarities</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low grade serous</td>
<td></td>
</tr>
<tr>
<td>Mucinous</td>
<td>Colorectal cancer</td>
</tr>
<tr>
<td>Endometrioid</td>
<td>Endometrial cancer</td>
</tr>
<tr>
<td>Clear cell</td>
<td>Renal cell cancer</td>
</tr>
<tr>
<td>Transitional cell/Brenners</td>
<td>Urothelial tumors</td>
</tr>
</tbody>
</table>
Recurrent genomic alterations in rare subtypes of epithelial ovarian cancer

Among rare subtypes of epithelial OC:
- Genomic alterations are **histology-specific**, and
- Frequently actionable

**Endometrioid:**
- PTEN M+ or loss (40%)
- PIK3CA M+ (20%)
- MSI (19%)
- ARID1A M+ (40%)

**Mucinous:**
- KRAS M+ (40-60%)
- HER2 Amp (18%)
- MSI (17%)

**Clear cell:**
- PTEN loss (20%)
- Met Amp (12%)
- HER2 Amp (15%)
- PIK3CA M+ (30%)
- ARID1A (60%)
- MSI (12%)

**Low grade serous:**
- KRAS M+ 40%
- BRAF M+ 5%

**High-grade serous**
ARID1A mutations common in Clear cell and endometrioid ovarian cancers

- Early event (found in precursor lesions)
- **ARID1A** = component of the SWI/SNF chromatin remodelling complex
- Tumor suppressor - regulation of
  - Lineage-specific differentiation,
  - Proliferation
  - Migration…

---

Table 1. Tumors with ARID1A mutations (13-32).

<table>
<thead>
<tr>
<th>Tumor</th>
<th>Rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ovarian clear cell carcinoma</td>
<td>46-57</td>
</tr>
<tr>
<td>Ovarian endometrioid carcinoma</td>
<td>30</td>
</tr>
<tr>
<td>Low-grade endometrioid carcinoma</td>
<td>40</td>
</tr>
<tr>
<td>Renal clear cell carcinoma</td>
<td>34</td>
</tr>
<tr>
<td>Gastric carcinoma</td>
<td>8-27</td>
</tr>
<tr>
<td>Transitional cell carcinomas of the bladder</td>
<td>13</td>
</tr>
<tr>
<td>Esophageal adenocarcinoma</td>
<td>9.1-15</td>
</tr>
<tr>
<td>Hepatocellular carcinoma</td>
<td>10-13</td>
</tr>
<tr>
<td>Esophageal carcinoma</td>
<td>9</td>
</tr>
<tr>
<td>Pulmonary adenocarcinoma</td>
<td>8</td>
</tr>
<tr>
<td>Prostate carcinoma</td>
<td>8</td>
</tr>
<tr>
<td>Pancreatic carcinoma</td>
<td>2-8</td>
</tr>
<tr>
<td>Breast carcinoma</td>
<td>2-4</td>
</tr>
<tr>
<td>Burkitt lymphoma</td>
<td>14-17</td>
</tr>
<tr>
<td>Neuroblastoma</td>
<td>6</td>
</tr>
<tr>
<td>Medulloblastoma</td>
<td>2</td>
</tr>
</tbody>
</table>

Takeda, 2016

---

ARID1A mutation: Synthetic lethality with ATR inhibition

ARID1A required for localisation of TOP2A to DNA

TOP2A causes DSBs in DNA and facilitates transport of one double helix through another

ATR activity normally invoked when TOP2A defective

ARID1A defect = TOP2A defect = activation of ATR signaling cascade = delayed progression through S and G2

ARID1A defect plus ATRi = cells fail to adequately respond to TOP2A defect = premature cytokinesis in the face of unresolved DNA structures
ARID1A mutant tumour cells are sensitive to small molecule ATR inhibitors, both \textit{in vitro} and \textit{in vivo}

\textbf{ARID1A mutations are frequent:}
50\% of clear cell ovarian cancers
30\%-40\% of endometrioid ovarian cancers

May become useful predictive biomarker in the future

**ARID1A WT xenografts**

**ARID1A DEFICIENT xenografts**

Dr. A. Leary 15 juin 2017 - Journée GFCO 2017
Microsatellite instability (MSI) frequent in non-serous ovarian cancer

MSI:
- Mutations (or promoter hypermethylation) in mismatch repair (MMR) genes
- Initially described as germline (Lynch)
- Somatic MMR mutations also described
- leads to hypermutated tumor → sensitivity to immunotherapies

1234 OC cases in 22 studies

<table>
<thead>
<tr>
<th>Histology</th>
<th>% MSI</th>
<th>N (tumours tested)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ALL</strong> epithelial OC</td>
<td>10%</td>
<td>Over 1000</td>
</tr>
<tr>
<td>Serous</td>
<td>8%</td>
<td>564</td>
</tr>
<tr>
<td>Endometrioid</td>
<td>19%</td>
<td>314</td>
</tr>
<tr>
<td>Clear cell</td>
<td>12%</td>
<td>117</td>
</tr>
<tr>
<td>Mucinous</td>
<td>17%</td>
<td>101</td>
</tr>
</tbody>
</table>

Useful predictive biomarker for Immune therapies in MSI ovarian cancer?

Murphy 2011
Summary: Molecular biomarkers in epithelial ovarian cancer

- **In most frequent HGSOC:**
  - Homologous recombination DNA repair deficiency
  - What are the most useful molecular markers?
    - BRCA germline mutations
    - BRCA somatic mutations
    - Alterations in other HR genes
    - HRD score
    - Is platinum sensitivity enough

- **In non-serous epithelial OC:**
  - Actionable alterations frequent: ARID1A mutations, MSI, HER2 amplifications…
  - In the near future these genomic alterations may become relevant to offer ‘personalized’ treatment
Summary: Molecular biomarkers in epithelial ovarian cancer

- **In most frequent HGSOC:**
  - Homologous recombination DNA repair deficiency
  - What are the most useful molecular markers?
    - BRCA germline mutations ✓
    - BRCA somatic mutations
    - Alterations in other HR genes
    - HRD score
    - Is platinum sensitivity enough

- **In non-serous epithelial OC:**
  - Actionable alterations frequent: ARID1A mutations, MSI, HER2 amplifications…
  - In the near future these genomic alterations may become relevant to offer ‘personalized’ treatment
Summary: Molecular biomarkers in epithelial ovarian cancer

• **In most frequent HGSOC:**
  – Homologous recombination DNA repair deficiency
  – What are the most useful molecular markers?
    - BRCA germline mutations ✓
    - BRCA somatic mutations ✓
    - Alterations in other HR genes
    - HRD score
    - Is platinum sensitivity enough

• **In non-serous epithelial OC:**
  – Actionable alterations frequent: ARID1A mutations, MSI, HER2 amplifications…
  – In the near future these genomic alterations may become relevant to offer ‘personalized’ treatment
Summary: Molecular biomarkers in epithelial ovarian cancer

- **In most frequent HGSOC:**
  - Homologous recombination DNA repair deficiency
  - What are the most useful molecular markers?
    - BRCA germline mutations
    - BRCA somatic mutations
    - Alterations in other HR genes?
    - HRD score
    - Is platinum sensitivity enough

- **In non-serous epithelial OC:**
  - Actionable alterations frequent: ARID1A mutations, MSI, HER2 amplifications...
  - In the near future these genomic alterations may become relevant to offer ‘personalized’ treatment
• **In most frequent HGSOC:**
  – Homologous recombination DNA repair deficiency
  – What are the most useful molecular markers?
    • BRCA germline mutations ✓
    • BRCA somatic mutations ✓
    • Alterations in other HR genes ?
    • HRD score ?
    • Is platinum sensitivity enough

• **In non-serous epithelial OC:**
  – Actionable alterations frequent: ARID1A mutations, MSI, HER2 amplifications…
  – In the near future these genomic alterations may become relevant to offer ‘personalized’ treatment
Summary: Molecular biomarkers in epithelial ovarian cancer

- **In most frequent HGSOC:**
  - Homologous recombination DNA repair deficiency
  - What are the most useful molecular markers?
    - BRCA germline mutations ✓
    - BRCA somatic mutations ✓
    - Alterations in other HR genes ?
    - HRD score ?
    - Is platinum sensitivity enough Perhaps....

- **In non-serous epithelial OC:**
  - Actionable alterations frequent: ARID1A mutations, MSI, HER2 amplifications...
  - In the near future these genomic alterations may become relevant to offer ‘personalized’ treatment
Diagnostic value of genomic characterization of rare ovarian tumors?
Molecular markers in non-epithelial ovarian cancer

- Germ cell ovarian cancer
- Sex-cord tumors of the ovary
  - Granulosa cell tumors
  - Sertoli-Leidig tumors
- Small cell cancer of the ovary, hypercalcemic type (SCCOHT)...

- Rare
- Diagnostic challenge
- Diagnostic value of genomic characterization
Molecular markers in non-epithelial ovarian cancer

Mutation of FOXL2 in Granulosa-Cell Tumors of the Ovary

>90% adult granulosa tumors

Recurrent Somatic DICER1 Mutations in Nonepithelial Ovarian Cancers

DICER1: endoribonuclease involved in microRNA processing
Mutated in 50% of Sertoli-Leidig tumors

Molecular markers in non-epithelial ovarian cancer

Mutation of FOXL2 in Granulosa-Cell Tumors of the Ovary

>90% adult granulosa tumors

Screening for these genomic alterations has entered routine practice as diagnostic test

Recurrent Somatic DICER1 Mutations in Nonepithelial Ovarian Cancers

DICER1: endoribonuclease involved in microRNA processing
Mutated in 50% of Sertoli-Leidig tumors

EXTREMELY RARE
< 1% of ovarian cancers

Highly aggressive form of ovarian cancer

- Mean age of diagnosis: 23 years
- Poor Prognosis
- Frequently mis-diagnosed (confused with germ cell, round cell tumors, granulosa…)
- Diagnostic challenge: requires pathologist with expertise ‘rare ovarian tumors’
- Accurate diagnosis urgency needed because the right treatment needs to be started quickly

SCCOHT: Small Cell Carcinoma of the Hypercalcemic Type, OC
SMARCA4 mutation universal in SCCOHT

Confirmed SMARCA4 mutations in SCCOHT

SMARCA4 mutations are ubiquitous in SCCOHT (>90%)

Screening for SMARCA4 mutation now routine diagnostic test in difficult to characterize aggressive tumor in a young woman

Dr. A. Leary 15 juin 2017 - Journée GFCO 2017
Summary: Molecular/genomic markers in rare non-epithelial ovarian cancer

- Genomic profiling of rare ovarian tumors has provided a new useful diagnostic tool
- FOXL2 mutations for granulosa tumors
- DICER1 mutations for sertoli leidig tumors
- SMARCA4 mutations for small cell cancer of the ovary of the hypercalcemic type
Genomic profiling of endometrial cancer
Not all Endometrial Cancers are the same...

TCGA identified 4 classes of endometrial cancer
Classified according to mutation load and copy number

Not all Endometrial Cancers are the same...

Genomic classification provides prognostic information:
- **POLE** mutated: best survival (could be spared toxic adjuvant treatment)
- **TP53** mutated: worst. Need adjuvant chemotherapy?

Does genomic profiling of endometrial cancer provide predictive information?
POLE mutated and MSI endometrial cancers: High mutation load and high TILs

From: Association of Polymerase e–Mutated and Microsatellite-Instable Endometrial Cancers With Neoantigen Load, Number of Tumor-Infiltrating Lymphocytes, and Expression of PD-1 and PD-L1
POLE/MSI tumors

- High mutation load
- More neo-antigens
- Increased TILS CD3+/CD8+
- Increased PD1/PDL1+ expression

Response to PD1/PDL1 inhibitors?

Response to PD1 inhibitor in MSI vs MSS tumors

<table>
<thead>
<tr>
<th></th>
<th>MSS CRC N=18</th>
<th>MSI CRC N=10</th>
<th>MSI tumors N=7</th>
</tr>
</thead>
<tbody>
<tr>
<td>RR</td>
<td>0</td>
<td>40%</td>
<td>70%</td>
</tr>
</tbody>
</table>

RR= 70% in MSI non colorectal cancers (including endometrial cancer)

Le D NEJM 2015
**POLE/MSI tumors**

- High mutation load
- More neo-antigens
- Increased TILS CD3+/CD8+

**MAY 2017**

**FDA Approves Pembrolizumab for Microsatellite Instability-High and Mismatch Repair Deficient Cancers**

Response to PD1 inhibitor in MSI vs MSS tumors

<table>
<thead>
<tr>
<th></th>
<th>MSS CRC N=18</th>
<th>MSI CRC N=10</th>
<th>MSI tumors N=7</th>
</tr>
</thead>
<tbody>
<tr>
<td>RR</td>
<td>0</td>
<td>40%</td>
<td>70%</td>
</tr>
</tbody>
</table>

**RR= 70% in MSI non colorectal cancers (including endometrial cancer)**

Le D NEJM 2015
Genomic biomarkers in Endometrial Cancer

Genomic stability

- Ultra-mutated POLE M+
- Hyper-mutated MSI
- Copy-number low MSS
- Copy number high TP53 M+

Prognosis

- Excellent prognosis
- Good prognosis
- Poor prognosis
  Need adjuvant chemotherapy?

Tailored therapy?

- Excellent prognosis
- Hyper-mutated
  Treatment: Anti-PDL1

Dr. A. Leary 15 juin 2017 - Journée GFCO 2017
Conclusions: Value of genomic profiling in endometrial cancers

Most EC are cured by local treatment alone

Greatest value of genomic profiling in refining prognostic groups:
- POLE: Excellent prognosis
- TP53: Poorest prognosis

Actionable alterations?
- MSI yes!
- TP53 mutations: need post-operative chemotherapy?
Biomarkers in gynecological tumors: Conclusion

**In HGSOC:** PARP inhibitors: 1st targeted therapy associated with a genomic biomarker – g or s BRCA mutations

→ Can we identify the subset of BRCA WT with HR deficiency?
  • High LOH score?
  • Mutations in non-BRCA HR genes?
  • Platinum sensitivity?

→ Will need this information earlier – At diagnosis
→ Other biomarkers? Reversion BRCA mutations, caracterizing the ‘non-HRD’

**In non-serous epithelial OC:** Number of candidate predictive biomarkers (ARID1A mutations, MSI, HER2 amplifications…)

**In non-epithelial OC:** Useful diagnostic tool: FOXL2 mutations, SMARCA4 mutations

**In endometrial cancers:** Prognostic (POLE or TP53 mutations) and predictive (MSI)