Charles University

Faculty of Science

Study programme: Special Chemical and Biological Programmes

Branch of study: Molecular Biology and Biochemistry of Organisms

Jana Ivančinová

High grade serous ovarian carcinoma: molecular background and platinum-based chemotherapy challenges

High grade serózní karcinom ovaria: molekulární pozadí a výzvy chemoterapie založené na platině

Bachelor's thesis

Supervisor: doc. RNDr. Petr Heneberg, Ph.D.

Prague, 2022

Acknowledgement:

I wish to express my thanks and appreciation for the support, devoted time, advice and patience from my supervisor, doc. Petr Heneberg. My thesis would not have been possible without his selfless support. I would also like to thank my colleagues from university and friends that kept my sanity in check during my hard times.

Prohlášení:

Prohlašuji, že jsem závěrečnou práci zpracoval/a samostatně a že jsem uvedl/a všechny použité informační zdroje a literaturu. Tato práce ani její podstatná část nebyla předložena k získání jiného nebo stejného akademického titulu.

V Praze, 02.05.2022

Podpis

Abstract

Ovarian carcinoma (O.C.) represent a group of various disease entities derived from ovaries. The most common malignant gynaecological cancer is high-grade serous ovarian carcinoma (HGSOC). HGSOC is associated with a high mortality rate due to its aggressive behaviour and insufficient early-stage detection. The survival rate has not been significantly improved since 1970s. The most effective treatment of HGSOC patients is by cytoreductive surgery (for early stages I/II) and followed by platinum-based chemotherapy (HGSOC presented in advanced stage III/IV) combined with taxane or potentially with PARP inhibitors (for BRCA1/2 mutation carriers). Multiple factors affect the patient's outcome and prognosis. Chemoresistance, molecular mutational patterns, stage at presentation of HGSOC are one of the clinical challenges contributing to common relapses even though patients often initially respond well to the HGSOC chemotherapy. This thesis overviews the fundamental biology of HGSOC, the major obstacles in clinical management and its improvements by implementing of multitherapy approaches.

Key words: CA-125; platinum−based chemotherapy treatment; homologous recombination deficiency; ovarian carcinoma; resistance; *Tp53;* mortality; survival rate

Abstrakt

Karcinom ovaria (OC) je soubor různých rakovin pocházející z vaječníků. Nejčastějším zhubným gynekologickým nádorem je high-grade serózní karcinom vaječníků (HGSOC). HGSOC je spojen s vysokou úmrtností, a to z důvodu jeho agresivity a nedostatečné detekce v časném stadiu. Od 70. let 20. století se míra přežití výrazně nezlepšila. Nejúčinnější léčba pacientek s HGSOC je cytoredukční chirurgie (u časných stadií I/II) a následná chemoterapie na bázi platiny (HGSOC prezentovaný v pokročilém stadiu III/IV) v kombinaci s taxanem nebo případně s inhibitory PARP (u nosiček mutace BRCA1/2). Výsledek a prognózu pacienta ovlivňuje vícero faktorů. Chemorezistence, molekulární mutace, stadium při prezentaci HGSOC patří ku klinickým výzvám, které přispívají k častým relapsům, přestože pacientka původně dobře reagovala na léčbu HGSOC. Práce zabývá biologii HGSOC, hlavními překážkami v klinické léčbě a zlepšení zavedením multiterapeutických přístupů.

Klíčová slova: CA-125; terapie na bázi platiny; deficit homologní rekombinace*;* ovariální karcinom; *Tp53*; mortalita; míra přežiti

Contents

List of Abbreviations

ATM	Ataxia telangiectasia mutated
ATR	Ataxia telangiectasia and Rad3 related
Bcl2	B-cell lymphoma 2
BNIP3	*Bcl2 interacting protein 3*
BRCA	*Breast cancer*
CA-125	Cancer antigen 125
CA72-4	Carbohydrate antigen 72-4
CBDCA	Carboplatin
ChK1/2	Checkpoint kinase 1/2
CSMD3	*CUB and sushi multiple domain 3*
OCCC	Ovarian clear-cell carcinoma
CTLA−4	Cytotoxic T−lymphocyte associated antigen 4
CTR1	*Copper transport protein 1*
DACH	1,2−diaminocyclohexane
DDP	Cis−platin
DOXIL	Liposomal doxorubicin
DSB	Double-strand breaks
E.C.	Endometroid carcinoma
EOC	Epithelial ovarian carcinoma
FDA	U.S. Food and Drug Administration
FGFR2	*Fibroblast growth factor receptor 2*
FTSEC	Fallopian tube secretory epithelial cells
GLOBOCAN	Global Cancer Incidence, Mortality and Prevalence
GSH	Glutathion
HGSOC	High grade serous carcinoma
HMG-1	*High mobility group DNA binding protein-1*
hMLH	*Human mutL homolog*
HPf	High powered field
LPA	Lysophosphatidic acid
L−OHP	Oxaliplatin
LSGOC	Low grade serous ovarian carcinoma
MDM	Mouse double minute homolog
MMR	Mismatch repair
MOC	Mucinous ovarian carcinoma
MRP	multidrug resistance associated protein
MYC	**My**elo**c**ytomatosis
NAC	Neoadjuvant chemotherapy
NER	Nucleotide excision repair
NF1	*Neurofibromin 1*
O.C.	Ovarian carcinoma
OECD	Organisation for Economic Co-operation and Development
OSEC	Ovarian surface epithelium cells
PARP	Poly ADP-ribose
PAX8	*Paired box gene 8*
PD−L1	Programmed death ligand
PFS	Progression-free survival

PLD	Pegylated liposomal doxorubici
Pt	Platinum
PtC	Platinum based chemotherapy
PTEN	*Phosphate and tensin homolog on chromosome ten*
RB1	*Retinoblastoma protein 1*
RFI	Recurrence−free interval
Swi/SNF	Switch/Sucrose non-fermentable
TAG 72	tumour associated glycoprotein 72
TCGA	The cancer genome atlas
TNF	Tumor necrosis factor
Tp53	*Tumour protein p53*
ÚZIS ČR	Ústav zdravotnických informací a statistiky České republiky
WT	Wild−type

1. Introduction

The platinum−based chemotherapy (PtC) has remained the main post−surgery treatment since its introduction in the 1970s. Among the few treatment advancements is an introduction of PARP inhibitor treatment for patients with *BRCA* mutations and homologous recombination deficiency (HRD), and the introduction of a new generation of cis−platin (DDP) derivates [1]. The platin-based dosage is limited by the side effects. Many patients do not show any treatment responses. Others initially show good responses but later demonstrate relapses of the disease and consequently develop chemoresistance. Since the first use of the DDP, a variety of mechanisms of resistance has been discovered [2].

PtC is still used to treat high−grade serous ovarian carcinoma (HGSOC) despite manifesting strong chemoresistance. Patients with HGSOC are prevalently diagnosed in advanced stages due to the inefficient HGSOC screening in the early phases of the disease. The most widely used HGSOC marker is CA− 125. It is employed as a diagnostic tool in population screening and a clinical prognostic factor after surgery and first−line chemotherapy[3]. Alternative markers have been identified through the years, including osteopontin or lysophosphatidic acid (LPA)[4]. However, reliable techniques for detecting stage I/II HGSOC have not been developed yet.

It is essential to understand the aetiology, progression, molecular and physiological patterns of the cancer case to choose a successful treatment. Multiple variables affect the disease, these include origin, morphology, clinic-pathological characteristics, and cancer microenvironment. Accurate models are needed to detect patterns and variables of individual tumours and predict more precisely the response of the cancer cells to applied treatments. Although it is difficult to predict tumour's response, there has been an initiative to create more accurate in vitro and in vivo models [5].

This thesis aims to introduce HGSOC with its patterns, elucidate the molecular mechanism of HGSOC resistance, clarify the role of DDP in combatting ovarian carcinoma, and point towards anti−cancer drugs that could avert relapse and advance HGSOC treatment.

2. Epithelial ovarian carcinomas (EOC)

2.1 Cancer statistics

In 2018, overall cancer incidence in the Czech Republic was estimated by ÚZIS ČR to be 822 cases per 100 000 persons, and the OECD database estimated the mortality rate to be 215 cases per 100 000 persons (*Figure 1)* [6] [7]. Globally, there has been diagnosed 19 292 789 new cases of cancer, in 2020. Out of them, 313 959 cases have been diagnosed with ovarian carcinoma (O.C.) and 207 252 patients died because of ovarian carcinoma in the same year [8]. Therefore, epithelial ovarian carcinoma is the 14th most frequent cause of cancer death and the most frequent cause of death from gynaecological cancer globally [8]. In the Czech Republic, the O.C. incidence has been somewhat declining since 1996 [9](*Figure 2)*.

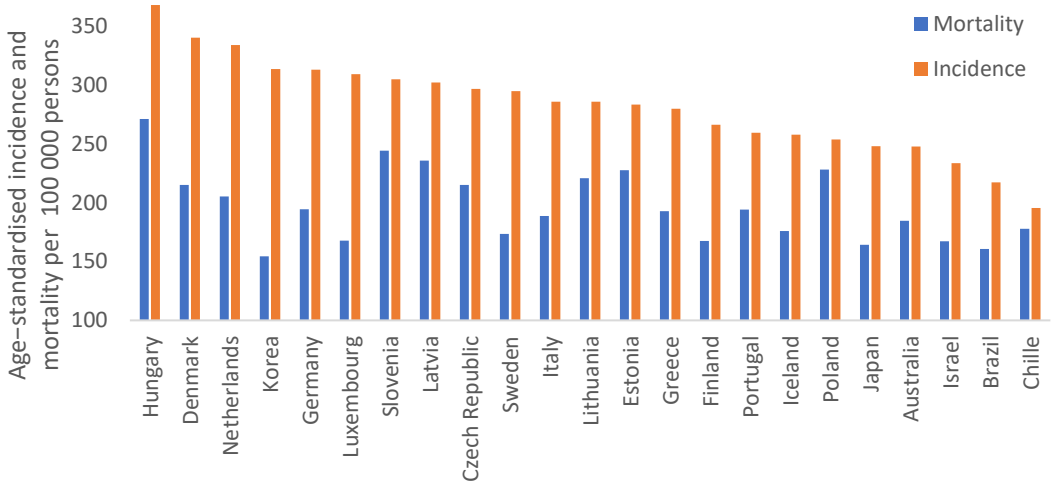

Figure 1: Country specific cancer incidence and mortality in 2018. The Y- axis shows the age-standardised incidence and mortality per 100 000 persons gained from different countries represented on the X-axis. Orange columns represent incidence and blue columns mortality. Source: OECD (2020); *OECD/European Union (2020))* [6] [7]

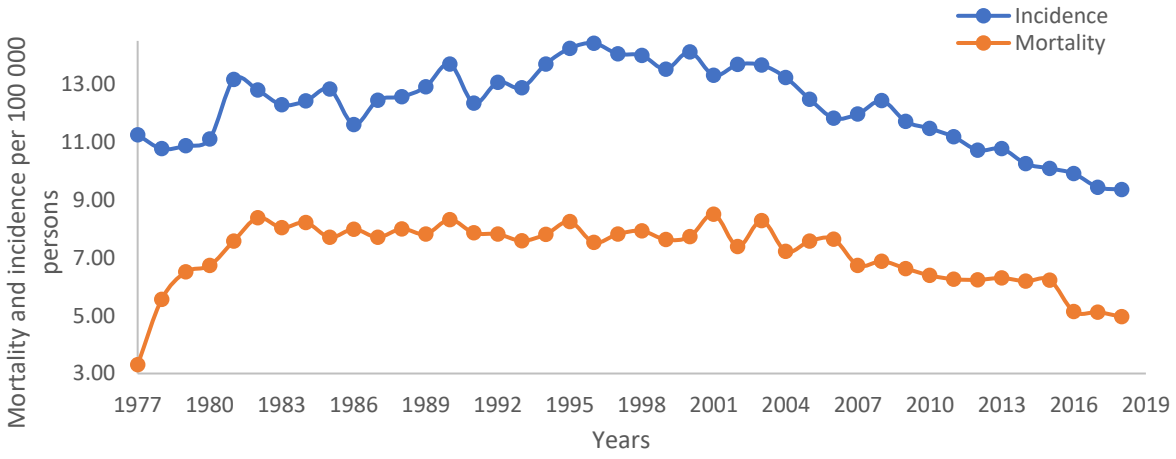

Figure 2: Progression of mortality and incidence of ovarian carcinoma (diagnosis C56) from 1977 to 2018 In the Czech Republic (the source data include the whole population). The X-axis represents the years when the data were gained, and Y-axis represents the number of cases per 100 000 persons. The orange trendline represents mortality and blue incidence. Source: ÚZIS ČR (2020) [8]

Unfortunately, the survival rates remain low for the advanced stage of HGSOC throughout decades, with only a slight improvement [10]. The improvement was by 11% in the 5-years survival rate in the last 20 years [11]. The survival rate depends on the stage and extent of the disease at the time of its detection and the number of tumour residuals after debulking surgery [10]. The relative 5-year survival rate in advanced phases of HGSOC was 35−40%. Comparably, the relative survival rate of Stage I/II presentation was 92% [12] [13]. Most ovarian carcinomas develop in postmenopausal women after losing the primary physiological role of the ovaries. Its anatomical location and lack of early symptoms impose a lot of difficulties in diagnosis. As a result of these factors, the diagnosis in stage I is rare. Therefore, the O.C. is usually diagnosed in advanced stages, when it reaches a large size, or disseminates. Most of the patients with stage I/II of O.C. had only one symptom, usually pelvic pain, and abdominal pain, or less often, increased girth or fullness [14]. In contrast, there is a wide variety of symptoms observed in stage III/IV of OC. These include abdominal bloating, already mentioned pelvic or abdominal pain, urinary symptoms and increased abdominal size [15].

2.2 O.C.subtypes

O.C. represent a group of various cancers that form in the ovaries[16]. Epithelial ovarian carcinomas (EOC) are the most common of gynaecological cancers, accounting for 90% of

cases[17]. There are four common histotypes of epithelial ovarian carcinoma based on their tumour cell morphology: serous; further subdivided into HGSOC (65−71% cases of O.C.) and less frequent low−grade ovarian carcinoma (LGSOC) (<5% cases of O.C.); endometrioid carcinoma (E.C.) (17% cases of O.C.), clear−cell carcinoma (OCCC) (10% cases of O.C.) and mucinous ovarian carcinoma (MOC) (5% cases of O.C.)[16]. Some O.C. types, like MOC, EC, OCCC and LGSOC, are more likely diagnosed in stage I. In contrast, HGSOC is almost exclusively diagnosed in stage III/IV (*Figure 3)* [11].

	HGSOC	LSGOC	MOC	EC	OCCC
Risk factors	BRCA1/2, menopausal hormonal therapy	?	Smoking, oral contraceptive use	HNPCC, BMI ≤30 kg/m^2	?
Precursor lesions	tubal intraepithelial carcinoma	serous borderline tumour	cystadenoma/ borderline tumour	endometriosis	endometriosis
Genes alteration	*Tp53, BRCA1/2, PTEN*	*KRAS, BRAF, NRAS*	*KRAS, BRAF, Tp53, HER2*	*FGFR2, PTEN, ARID1A*	*ARID1A, ZNF217, PIK3CA, KRAS*
Mean age at diagnosis	*62*	*55*	*63*	*61*	*59*
Frequency	71%	<5%	5%	17%	10%
5−year survival rate	35.3%	55%	63%	83%	62%
First−line treatment	Resection surgery, PtC with taxane and possibly the usage of PARP inhibitors as maintenance therapy	Resection surgery, PtC with taxane	Bilateral salpingo-oophorectomy surgery, followed by carboplatin (CBDCA)/paclitaxel	Surgical approach consisting of hysterectomy and bilateral salpingo−oophorectomy, debulking; followed by anthracyclines, Pt compounds and taxane	Surgery followed by standard chemotherapy of paclitaxel and CBDCA

Table 1: Characteristics of each subtype of EOC. Sources: J. Prat (2009) [18]*;* Coleman et al. (2011) [19]*;* Yang et al. (2012) [20]*;* Colombo et al. (2013) [21]; Fotopoulou et al. (2017b)[21]*;* Kang et al. (2020) [23]*;* Matsuo et al. (2020) [11]; Gaitskell et al. (2022)*[23].*

Serous ovarian carcinomas have been classified historically into three groups: low− grade, intermediate−grade and high−grade. The MD Anderson Cancer Centre's two−tier (binary) grading system was developed and based on a mitotic index[25]. The mitotic index is a ratio of cells that were detected in mitosis to the total number of cells [26]. Tumours with a lower rate of the mitotic index are classified as low−grade tumours, with high mitotic index are high−grade tumours. A high rate of mitotic index is considered if >12 mitoses per 10 high−powered fields (HPFs) occur. The grading system MD Anderson Cancer Centre binary system, in comparison with other grading systems, Shimizu/Silverberg and FIGO grades have

shown a significant predictive ability[25]. Furthermore, the tumours with lower mitotic index are less aggressive and consist of the 5−year survival rate of LGSOC (55%)[27]. Whereas more aggressive HGSOC has a 5−year survival rate of 35.3% [11].

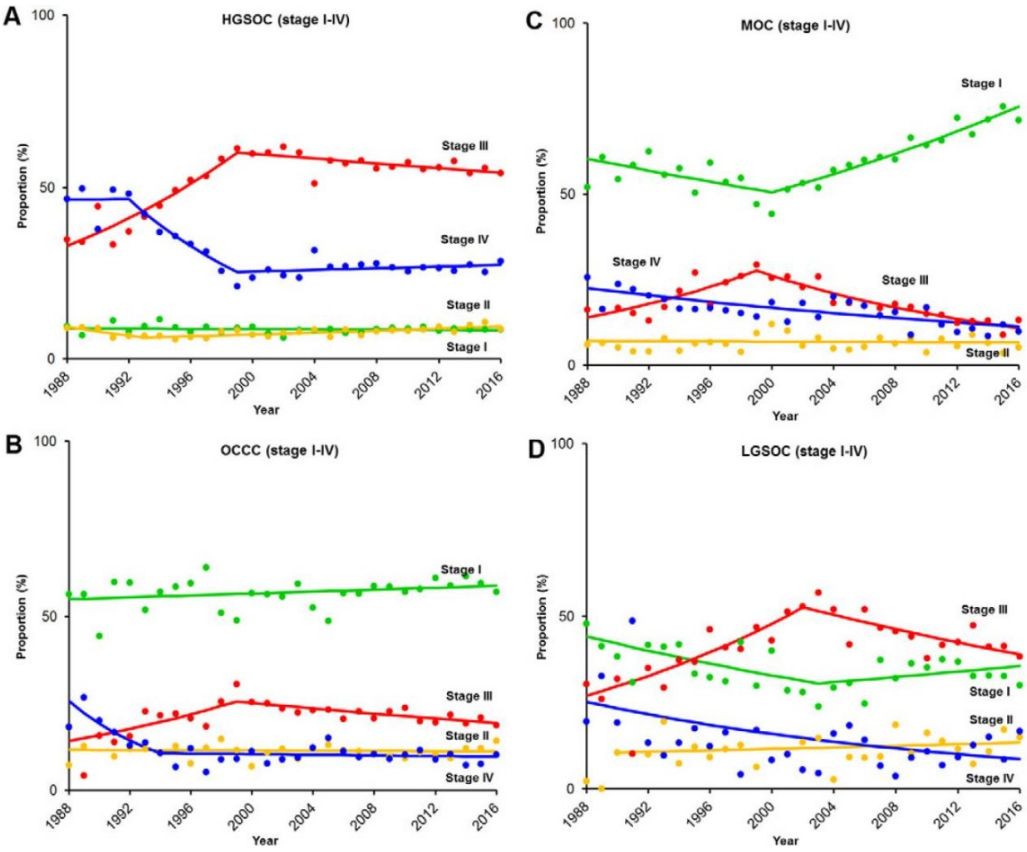

Figure 3: Trend in cancer stages of four EOC histotypes. The data are shown 1988 to 2016; the datasets are based on the U.S. population. X-axis shows the proportion of individual cancer stages. The green trendline corresponds to Stage I, yellow to stage II, red to stage III and blue to stage IV. A: HGSOC, B: OCCC, C: MOC, D: LGSOC. Source: Matsuo et al. (2020)*[11]*

3. HGSOC

HGSOC is the most lethal form of O.C., particularly because its aggressive and chemoresistant nature. HGSOC is responsible for 70−80% of EOC related deaths [11]. This cancer type shows heterogeneous histopathological patterns and could be papillary, solid or growing with a slit−like glandular lumen[28]. The nuclei of the HGSOC cells have a high mitotic index, greater than 12/10 HPFs [25].

3.2 Aetiology of HGSOC

Although much research has been published concerning HGSOC's origin, its origin remains controversial. Currently, there are two major theories: tubal and ovarian theories. Initially, it has been suggested that HGSOC arises from the ovarian surface epithelial cells (OSEC) that invaginate in the underlying stroma and form a cyst that transforms into a malignancy[29] [30]. However, recent studies suggest fallopian tube secretory epithelial cells (FTSEC) as possible precursor cells [31]. Detection of precancerous or early cancerous lesions in the fallopian tube of patients suffering from HGSOC, expression of the secretory cell lineage markers, presence of Müllerian marker and *PAX8* expression supports the FTSEC theory [32]. The high expression of calretinin, mesenchymal marker vimentin, *GATA4* and *NR5A1* in proliferative HGSOC support the OSEC theory [30] [32]. The formation of HGSOC could be possibly caused by the implantation of epithelial cells from the tube or possibly exfoliated cells from serous tubal intraepithelial carcinoma in the ovary (*Figure 4)*. It starts with the development of cortical inclusion cysts and cystadenomas, eventually leading to the development of a carcinoma. [32] [34].

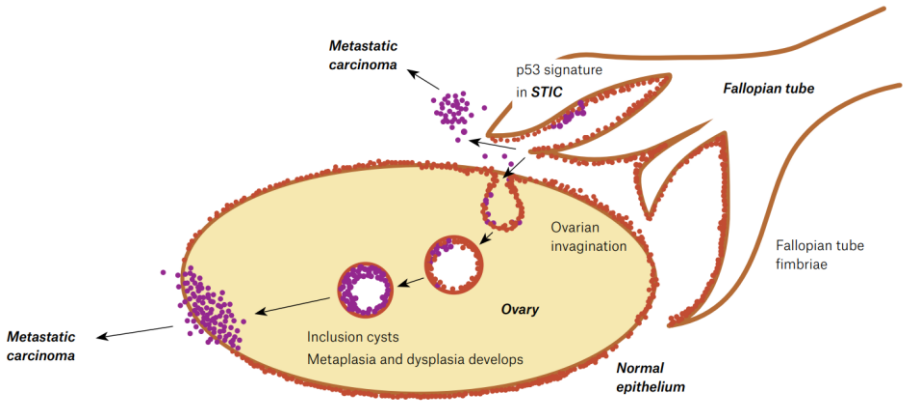

Figure 4: The current view of the HGSOC aetiology from fallopian tube cells that originate from fallopian tube distal end. STIC, serous tubal intraepithelial carcinoma. Source: Neesham et al. (2020) [35]

FTSEC as the origin of a substantial percentage of HGSOC has been confirmed by lineage tracing specifically tracing *Pax8rTA*, which selective expression was localised in FTSEC [36]. However, studies do not exclude the possibility of establishing HGSOC from either cell type. For instance, by tracing marker *Lgr5*, which is present in OSEC but not in FTSEC. *Lgr5* also drove HGSOC formation in adult mice combining of Rb family inactivation or *Tp53* mutation. Experiments with OSEC organoids confirmed the studies made on adult mice, and gave rise to HGSOC with the same genetic abnormalities[34]. These different properties of each cell−of−origin contribute to the inter−tumour heterogeneity and influence the behaviour of HGSOC [34].

Different cells-of-origin of HGSOC seem to drive a diversity in behaviour, growth pattern and metastasis. FTSEC−derived tumours had a bigger inclination to disseminate, whereas OSEC−derived tumours formed large and solitary lesions [34].Some oncogenic genetic profiles and molecular patterns are cell−of−origin−specific, supporting the theory of dualistic tissue origin of HGSOC. Gene expression and methylation profiling analyses were consistent that either hypothesis, OSEC or FTESEC/fimbrial cells could serve as the origin of EOC [38 39].The HGSOC could be classified into two subtypes based on dualistic tissue origin theory. Predictive genes for a favourable prognosis are expressed more in fallopian tube cells, whereas those associated with poor prognosis have lower expression in fallopian tube cells. Laboratory findings correlated with more favourable outcomes for fallopian tube-derived tumours than for OSEC-derived tumours [32] . The cell-of-origin max contribute to different tumour's sensitivity to chemotherapy. For example, the fallopian tube-derived tumorigenic organoids were more sensitive to CBDCA than the OSEC-derived organoids.[34]

3.2 Screening and diagnosis

Only a few screening tools are available for the early detection of EOC. Throughout the last decades, numerous potential biomarkers have been discovered, but few have reached clinical use[40]. Most of the markers have limited sensitivity for stage I/II of EOC. Identifying an adequate marker for population screening remains challenging [41 42].

3.2.1 CA-125

Altogether currently, the most clinically used O.C. marker is CA-125, which is suitable for diagnosis and monitoring the response to treatment due to its best sensitivity for EOC [43]. CA-125 is a heavily glycosylated mucin protein, uncovered in 1983 as an O.C. antigen [44]. The expression of CA-125 was detected using the murine monoclonal antibody clone OC125. OC125 determined the expression of antigen in tissue derived from the coelomic and Mullerian epithelium [45]. Although the surface of ovaries is formed by invagination of coelomic epithelium, it does not

normally express CA-125, except in cases of the presence of abnormal tissue such as neoplastic lesions, inclusion cyst, metaplasia areas or papillary excrescences [46]. In cases of tumorous tissue, the CA-125 could be detected in the ovarian surface epithelium. It is the most common method together with ultrasound for the detection of the majority of histotypes of EOC.

The threshold value of CA-125 in serum is ≥ 35 *U·mL−1* which could indicate different cancer risks. The cut−off value for detection could be lowered to 16 *U·mL−1* for significantly improved detection. It is the selected upper limit value of the normal reference range for postmenopausal women [46]. Although decrement of threshold value improves detection simultaneously, it imposes a risk of increment in false−positive patients. The increased level of serum CA-125 has been associated with various diseases [46]. Later, there has been developed a CA-125 II assay, which uses a different monoclonal antibody, termed M11. CA-125 II assay showed the same sensitivity and specificity as the older CA-125 I assay, but it showed good reproducibility even at lower concentrations of antigen, was a better predictor of recurrence and could be superior to the CA-125 I assay. A big benefit of the CA-125 II assay is an improvement in detecting of all histotypes of EOC, including MOC, which is difficult to detect using CA-125 assay I. The false-positive rate of CA-125 II when tested on 240 subjects with normal values resulted in 5.4% (14/240 cases in the control group) when chosen cut−off value was 35 $U \cdot mL^{-1}$. In the control group of 46 patients with O.C., 45 of them had levels of CA-125 II ≥ 35 $U \cdot mL^{-1}$ whereas CA-125 I assay showed only 91.3% positivity [47]. Raised levels of CA-125 were detected in 50% of patients diagnosed with O.C. of stage I and 90% of patients with EOC of stage II−IV [48].

3.2.2 Other molecular markers of O.C.

Carbohydrate antigen 72−4 (CA72−4) maker was detected by the TM−CA72−4 quantitative immunosorbent assay that was commercialized by DRG [49]. Tumour−associated glycoprotein 72 (TAG−72) showed more sensitivity in MOC than CA-125, according to Kobayashi (1989)[50]. Despite it's the positive results in studies, it has never gained FDA approval. TAG 72 is associated with O.C. and colon or gastric cancers, especially more elevated in mucinous tumours, where is CA-125 less efficient[51].

Another potential O.C. marker is a phospholipid with mitogenic activity, lysophosphatidic acid (LPA). It plays a key role in cell proliferation [52]. The levels of LPA were elevated in 9 of 10 patients in stage I of O.C. and in all 24 patients with more advanced stages the disease[53]. The healthy control group had elevated levels of LPA in five cases out of a total of 48 cases. LPA assay showed better sensitivity in stage I than the CA-125 assay. As the levels of

LPA were higher in eight samples out of nine examined samples form patients with stage I of O.C., whereas CA-125 showed only 2 out of 9 cases were elevated. The plasma LPA levels are not significantly influenced by histological subtype and grade of serous OC, but the plasma levels correlated with the extent of disease, and the assay offers a good prognostic and diagnostic tool even in stage I/II [54]. LPA was detected with significantly higher levels compared to healthy group only in E.C., the LPA predicted the EC in 5 women (83%), while CA-125 was detected only in one (16%) of women [55].

A serine protease prostasin is a marker of O.C. that was identified by immunostaining and microarray analysis of RNA expressed from human ovarian surface epithelial cells and normal human ovarian epithelium. This biomarker is secreted in a healthy organism by the prostate gland. The mean level of prostasin in the serum is 13.7 $\mu g \cdot mL^{-1}$ in patients with OC, and 7.5 $\mu g \cdot mL^{-1}$ in controls [56]. Combined with the CA-125 marker was achieved sensitivity equal to 92% (34/37 patients with non−mucinous OC) and a specificity of 94% (94/100 control subjects). The combination of CA-125 and prostasin showed better sensitivity compared to the CA-125 assay (24/37) alone or prostasin alone (19/37)[56].

More potential O.C. biomarkers have been isolated by ultracentrifugation from plasma and ascites. Their clinical significance in diagnosis and prognosis is under evaluation. For instance extracellular nanovesicles have shown their therapeutical potential whereas in diagnostic, as a therapeutical target or diagnostic biomarker [57]. Other potential biomarkers are claudin−4 extracted from HGSOC patients, sE−cad, Epithelial cell adhesion molecule, and Human-epididymis protein 4[43] [58].

3.2.3 Perspective of annual CA-125 screening

CA-125 and imagining remain the main clinical diagnostic tools, despite the variety of protentional O.C. markers. CA-125 is considered insufficient for the detection of stage I/II. Buys et al. (2011) screened a population of 78 216 women of the general U.S. population between the age of 55 to 74 years using CA-125 with a cut−off value of 35 $U \cdot mL^{-1}$ and transvaginal ultrasound [59]. Among these participants, O.C. was diagnosed in 388 women, mostly in stage III/IV. For 218 women, O.C. was the main cause of death. The false positivity of the CA-125 marker imposed a problem. In total, 3285 women were diagnosed with false-positive results.

In conclusion the annual screening of the US population for O.C. had not shown significant reduction in disease−specific mortality in women with an average risk of OC. These findings were proved from more screening trials and state that currently used biomarkers are

not enough for early diagnosis. However, the combination of biomarkers could be key for detection improvement, as they perform significantly better than each individually [59] [60].

3.3 Molecular characteristics of HGSOC

HGSOCs are characterised by somatic mutations in *Tp53* (95-100% of patients), *PTEN (*36% of cases), germline mutations of *BRCA1/2* (8-20% of patients), while only 3% of patients showed somatic mutation of *BRCA1* or *BRCA2* [61]. Other recurrently mutated genes are negative regulator of cellular proliferation *RB1,* kinase regulating RNA splicing *CDK12,* cell growth regulator *NF1, CSMD3*, *FAT3* and *GABRA6* for GABA receptor [61] [62]. The mutation in genes *BRAF, PIK3CA, KRAS* and *NRAS* are rarely present in HGSOC. Another molecular abnormality regarding HGSOC is recurrent genetic amplification of *CCNE1,* which codes for G1/S specific cyclin E1 [61]. *CCNE1* was common for primary chemotherapy resistant HGSOC. Structural variation and inactivation of tumour suppressors *RB1, NF1* and *PTEN* contributed to acquired chemotherapy resistance [63]. Out of 489 cases, 319 had signs of CCNE*1* alterations, 106 of them had amplification and 165 of them had copy number gain. However, only 46−54 % of *CCNE1* amplified HGSOC cases also had high expression of protein cyclin E1 [64].

Nelson et al. (2020) have performed time-lapse microscopy and whole-genome sequencing on established cell lines from *ex vivo* patient's culture in pursuit of characterisation of the genomic abnormalities and chromosomal instability of tumour cells. The mitotic dysfunction was due to the highly heterogenous abnormal mitoses associated with karyotype heterogeneity and the abnormal poles number of the spindle was revealed by time−lapse microscopy. HGSOC has also been characterised by *MYC* amplifications, which drives proliferation and biogenesis*, PTK2* for focal adhesion-associated protein kinase*, CCNE1* and loss of *PTEN* [65] [66].

Many mutations contribute to chromosomal instability, these include particularly the *Tp53* and *BRCA1/*2 mutations. The chromosomal instability could be enhanced by overexpression of some proteins such as cyclin E1 or by amplification of loci, like the amplification of 8q24 locus, which includes the *MYC* gene. As a result of chromosomal instability and a high rate of abnormal mitoses, as the O.C. tumour cells exhibited 52 % of abnormal mitosis, the HGSOC has developed highly deviant karyotypes. Whereby genomes were enriched just by specific genes to whole-genome duplication or chromosomes rearrangements. Despite a high number of catastrophic mitoses, there were a sufficient amount of survived daughter cells[66].

Analyses of updated the cancer genome atlas (TCGA) dataset of HSOC samples indicated a high chromosomal copy number of 8q24 involving *MYC* loci (*Figure 5)* [61]. *MYC* expression targets glutaminase and MYC-regulated genes. The MYC-regulated genes are part of mitochondrial solute transporter, glutathione synthesis, urea cycle, nucleotide synthesis, NADPH generation and impact redox balance during hypoxia [67]. However, there has not been found a correlation between HGSOC prognosis, cell dependencies on *MYC* and copy number of gene *MYC* [68].

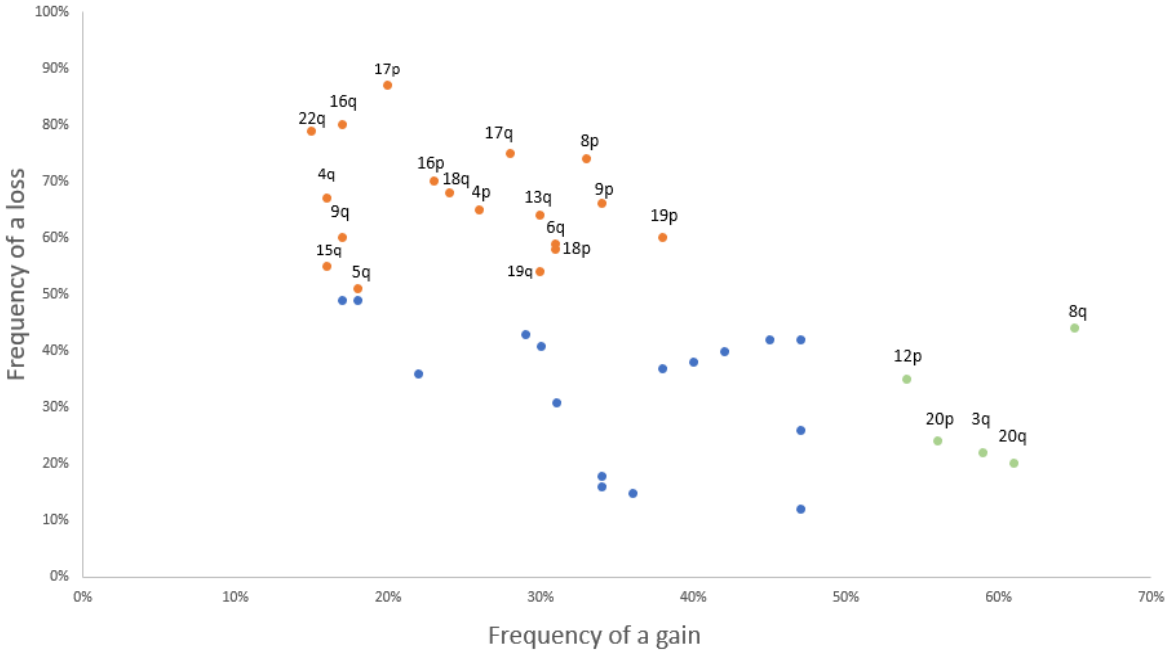

Figure 5: Frequency of loss or gain of genes on each arm of the 22 human autosomes. The X axis represents the frequency of gain of genes, and the Y-axis represents the frequency of loss. Orange points indicate arms that were lost in more than 50% of cases. Green points define the arms of chromosomes that were gained in more than 50% of cases. Blue points indicate arms that were gained or lost in less than 50% of cases. Source: TGCA (2011)[62]

3.3.1 *Tp53*

Tp53 is located at the small arm of chromosome 17, which is frequently lost in HGSOC *(Figure 4)* [61]. The transcriptional factor p53 is considered a gatekeeper of cell division and apoptosis. p53 regulates permanent cell cycle arrest, enhancing autophagy and apoptosis as a response to various cellular stress signals, DNA repair and modulation of reactive oxygen species levels [69]. p53 is phosphorylated by ATM and ATR kinases in response to the stress signals from the microenvironment. CHK1 and CHK2, promote p53 stability and activation when displaced from their negative regulator MDM2 and MDM4, which have E3 ubiquitin ligase activity

promoting proteasome−mediated degradation of p53[70]. The role of *Tp53* in cancer has been highlighted and studied over the decades. Mutations in p53 are common also in breast cancer, small cell lung cancer and oesophageal cancer. The mutations in *Tp53* are almost exclusively somatic; exceptions include Li Fraumeni syndrome [71].

Performing WGS was identified 116 different structural types of mutations in O.C. associated genes, and interestingly, the nonsense mutations of *Tp53* were found exclusively in patients treated by neoadjuvant chemotherapy (NAC). On the other hand, missense mutations have been typically harboured in patients, untreated by NAC[72]. However, the mutation rates are not constant, and even in the individual type of tumour, the mutation may differ by stage and between cell clones [73].

The *Tp53* mutations are mostly located in its DNA−binding domain. Mutated p53 can lose its tumour suppressive properties [74]. p53 associates with transcriptional factors, such as p63, p73, SP1 or chromatin regulating SWi/SNF. These transcriptional factors are important for cancer progression, metastasis and, in the end, supporting tumorigenesis and resistance to chemotherapy. The structural mutation *of Tp53* has limited effect on survival rate, although it has been noted that the mutation's function makes a slight difference. In the case of gain−of−function, the *Tp53* demonstrated a survival median of 44 months, while patients with HGSOC with loss−of−function in *Tp53* had a median of 50 months [71]. Furthermore, the mutations of *Tp53* are associated with Pt sensitivity [75].

Tp53 could be the main driver of HGSOC in the early development of tubal intraepithelial carcinoma. The cause of the chromosomal instability could be the downregulation of p53 and its negative regulators, such as *MDM2*, or targets downstream effectors of the pathway like *CDKN1A* encoding CDK inhibitor p21. P21 negatively regulates cyclin E and CDK2, which leads to the suppression of S−phase entry. When p21, a negative regulation is absent, cyclin E and CDK2 phosphorylate RB1 more rapidly, it permits de−sequestration of E2F, proving an essential role of p21 [76]. The importance of *Tp53* in the development of HGSOC was confirmed by experiments on genetically edited FNE1 cell lines of fallopian tube epithelium, as the loss of p53 was sufficient to drive the cell cycle dysregulation that induced chromosomal instability. The chromosomal instability could be exacerbated by manipulation with BRCA1 and overexpression of *MYC* [75].

3.3.2 *BRCA1/2*

HGSOC shares some genomic similarities with breast tumours due to the prevalence of BRCA1 and BRCA2 mutations. Defects in *BRCA1/BRCA2* lead to the disruption of homologous recombination and repair of DNA double−strand−breakage (DSB) machinery [66].

The germ−line mutations of BRCA*1/2* within O.C. are almost exclusively limited to HGSOC. There might be a connection between mutations of *BRCA1/2* function and *Tp53,* as an unrepaired DSB could be for cells lethal and trigger the p53 pathway leading to an apoptotic response [77]. Mutations of *BRCA* should be taken of particular interest because of their purpose in cells and their frequency in HGSOC patients. *BRCA* mutation are the second most common mutations in HGSOC, accounting for approximately 16%− 22% of germ−line mutations. Only minority of HGSOC, up to 3%, harbour somatic mutation in *BRCA1/2* [61].

Determination of the mutation status of the *BRCA* gene in HGSOC patients could be used for HGSOC patient's triage and as a useful prognostic tool because it improves the systematic selection of appropriate treatment approaches. Gained dataset showed that patients with germ-line mutation of *BRCA1/2* in stage III/IV have a better prognosis as they had slightly longer progression-free survival (23 months) than those patients lacking these mutations (17 months) (Kim et al., 2019). Mutations of *BRCA* are favourable prognostic sign for Pt sensitivity. The functional reversion of the mutations in *BRCA* genes by secondary mutation can lead to restoration of the open−reading frame and contribute to chemotherapy resistance [79]

3.3.3 *PTEN*

PTEN encodes a tumour suppressor, a phosphatase induced by the PIK3/Akt signalling pathway. Consequently, it inhibits cell proliferation, especially by negative pathway regulation. Besides its well−known effects in the cytoplasm, PTEN is also found in the nucleus, proving, that cells deficient in nuclear PTEN are susceptible to DNA damage as its loss leads to centromere breakage. *PTEN* in the nucleus may provide a role in maintaining of chromosomal stability and its integrity [80]. Moreover, loss of *PTEN* expression results in defect double−strand−breaks repairs as it regulates Rad51, an essential component of homology- directed DSB repair [81].

The cancer genome atlas research identified *PTEN* alterations in 7 % of HGSOC tumours and in another 6% of the tumours, homozygous deletion of allele occurred [61]. The study by Martins et al. (2014) updated the statistics on the frequency of *PTEN* alteration in HGSOC. The loss of a single *PTEN* allele was observed in 36% of samples (*N*=174). Even with a loss of one allele, the gene expression was remarkably lower [82]. They also discovered that its expression is independent of *Tp53.* The observation and analyses of the patient's overall survival and prognosis based on the expression level of *PTEN* demonstrated a worse survival rate if the expression of *PTEN* was reduced. HGSOC could be categorised into two subgroups based on *PTEN* expression profile; higher expression of *PTEN* is associated with a differentiated subgroup.

The second subgroup of HGSOC is defined by low or no expression as a proliferative group. The second group is associated with a poor prognosis [82].

3.4 Therapeutic approaches to HGSOC utilising PtC

Most the adequate care for HGSOC patients is cytoreductive surgery followed by Pt−paclitaxel−based chemotherapy with the potential use of PARP inhibitors in the case of *BRCA1/2* mutation carriers. This approach was introduced in the 1970s and is shown to be insufficient in the population of European descent [83]. Even women with HSGOC, who responded very well to chemotherapy after surgery, develop relapses in 20%-30% of cases within the half-year after the cessation of primary therapy [12]. Chemoresistance is one of the main obstacles to primary treatment and full recovery from disease [83].

The key determinant for Pt sensitivity in HGSOC is the mutation in the homologous recombination pathway, which renders it even more error-prone[84]. The homologous recombination defective pathway is caused by *BRCA 1/2* mutations but even more troublesome were HGSOC with *CCNE1* amplification, which is associated with a worse prognosis[64].

The primary debulking surgery is usually the first step for freshly diagnosed HGSOC patients. The visionary clinician Meigs in 1934 has changed adopted the new surgical techniques for treating O.C., decades before the discovery of adjuvant chemotherapy[85]. A number of postoperative residuals from disease could be a prognosis marker as there is a correlation between the percentage of maximal cytoreduction and survival rate. The increment by 10 % in maximal cytoreductive surgery has only a minor improvement in overall survival. Analyses showed a clear correlation between de-logged median survival rate and percentage of maximal cytoreductive surgery performed on patients with stage III/IV of OC. Cohorts with ≤25% of maximal cytoreductive surgery had estimated the mean survival rate 22 months, whereas cohorts with >75% maximal cytoreductive surgery had evaluated a median survival rate 33 months [86].

3.4.1 DDP

The standard approach followed the surgery in a case of advanced EOC was the use of cis-platin. Nowadays the approach is six cycles of 3−weekly paclitaxel/CBDCA. Throughout decades there has been synthesised a variety of Pt containing compounds, although active in clinical anticancer therapy and are especially three of them; DDP, CBDCA, L-OHP. DDP treat many type of cancers, including lung cancer, testicular cancer and more[87]. It interferes, with a DNA repair mechanism, in HRD cells deficient in functional *Tp53*. Consequently, DDP interacts with DNA synthesis and causes impaired cell division and cell−arrest in the prolonged G2 phase.

The DDP promotes nuclear and mitochondrial DNA damage and higher production of reactive oxygen species releasing pro−survival and pro−apoptotic signals [88]. DDP activates cell apoptosis also through the endoplasmic reticulum stress pathway through unfolded protein response[89]. The released signal molecules activate mitochondrial and non−mitochondrial pathways of apoptosis.

The mitochondrion is the primary target of DDP induced oxidative stress. Therefore, mitochondria exhibit increased metabolic activity and malfunction. The cancer cells exhibit greater reactive oxygen species concentrations than the healthy tissues. Oxidative stress is one of the mechanisms involved with DDP toxicity, one of the mechanisms triggering cell death[87]. The important mediators of DDP−induced cell death are p21, TNF receptor, Fas, and p53. DDP−induced cancer cell death is mediated by FasL, Bax/Bcl2 leading to activation of molecules inducing apoptosis, such as caspase−9/caspase−3 and caspase−8/caspase−3 [90 91].

BNIP3 is a BH3 protein, a member of the Bcl2 family, a factor which promotes tumour aggressiveness by influencing the proliferation and migration of cancer cells. *BINP3* expression correlated with cis-platin cytotoxicity in OC. Based on analyses of *BINP3* in O.C. Jie et al. (2020) hypothesised that there could be a dependency between DDP-induced apoptosis and OCs' *BINP3* levels to some extent. The depletion of this factor alleviates the cytotoxic effect. BNIP3 is more expressed in ovarian tumour tissue compared to healthy tissue under hypoxia, and it was confirmed that is BNIP3 silenced, there would be a remarkable decrement in the sensitivity to DDP [92].

DDP enters the cell at a relatively slow rate than other anti−cancer drugs, influx depends on many factors, and one of these is the presence of CTR1 transporter, which has been proven as substantial in DDP influx[93]. It is a Pt coordination compound of square planar geometry. Charged Pt ion is surrounded by two amine ligands and two chloride ligands. Chloride ligands, also called leaving groups, are initially activated intracellularly by the aquation of one of the two ligands, promoting the covalent binding of Pt with DNA [94]. Pt is coordinated in square-planar mode as the DDP bonds with N7 atoms of dinucleotides of DNA, closing a chelate ring. The amine ligands are hydrogen bond donors and could create a hydrogen bond in *cis* position with O6 of guanine [95]. The reaction products are inter−(minor product) and intra−strand crosslink (major product), like CpG cross−links, G−G crosslinks, monofunctional Pt−DNA adducts and at low rate also DNA−protein crosslinks, these DNA adducts force DNA in distortions, including unwinding and bending [96]. Bending is recognised by structure−specific proteins, especially by HMG−1

(Figure 6). These proteins are capable of bending DNA by themselves. As a result, they could act as DDP's cytotoxicity enhancer and affect DNA repair processes [97].

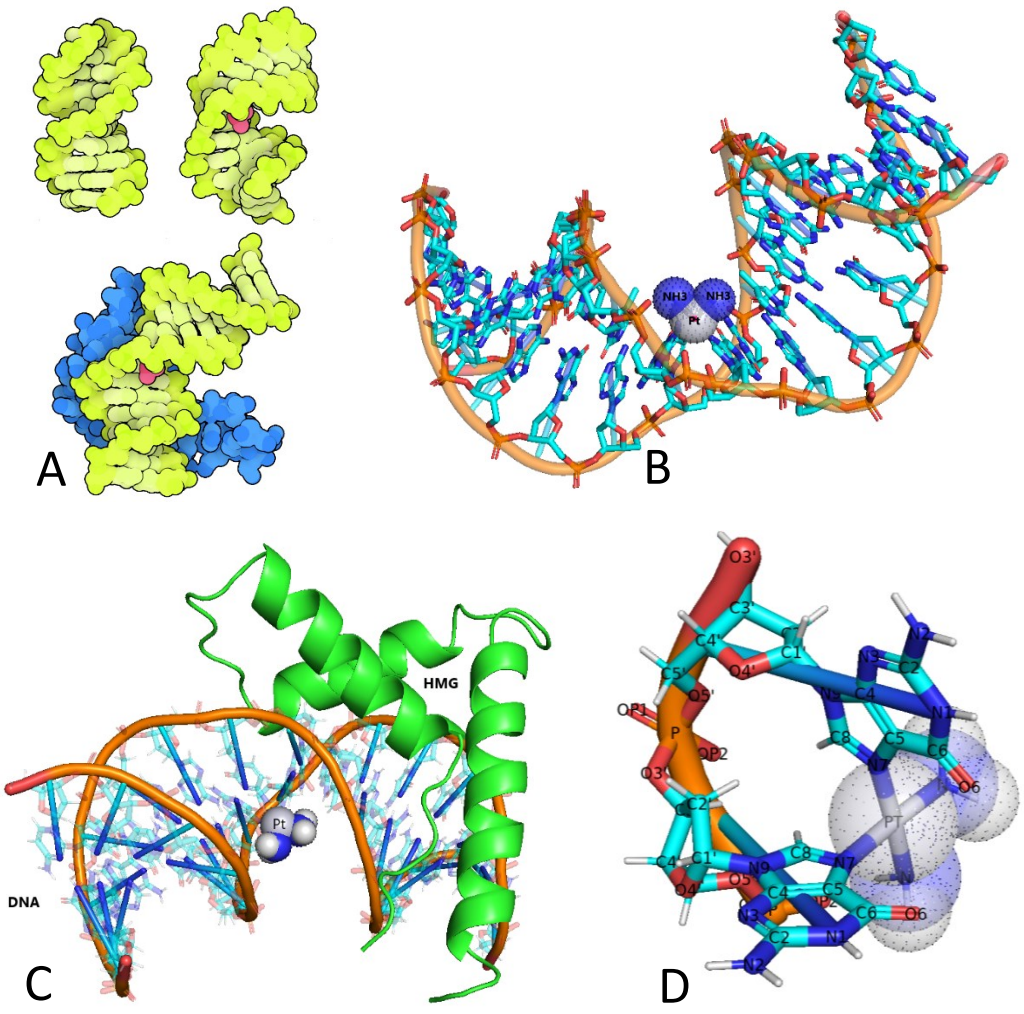

Figure 6: the Interaction of DDP with DNA. DDP forces DNA to bend, and the interaction is recognised by HMG-1 that can bend the DNA by itself. A: DNA without DDP, DNA with DDP, and DNA with DDP and protein. Yellow – DNA, blue – protein HMG-1 and red – DDP. B: Interaction of Pt atom of DDP and helix. C: The bending of DNA forced by HMG-1 and DDP. Green – protein, orange – DNA helix, white – Pt atom of DDP. D: detailed display of Pt interaction with N7 atoms of two guanines. Source: pdb-101: Cisplatin and DNA. Accessed form URL : <https://pdb101.rcsb.org/motm/255> [accessed on 2.5.2022][98]*. Edited in Pymol.*

M. Peyrone carried out DDP compound synthesis in 1844. The protentional use of DDP has not been elucidated until 1965, when Rosenberg et al. (1965) observed on *Escherichia coli* that the product of Pt mesh electrodes is capable of arresting the cell division and discovered its

therapeutics effects by accident [99]. Furthermore, based on the property of arresting cell cycle and promoting cancer cell death, DDP was the first FDA approved anticancer Pt drug [100].

The disadvantages of DDP arose from subsequent numerous safety issues associated with its cytotoxicity, such as causing severe kidney problems [91]. DDP induced cytotoxicity was noticed in 30−40% of patients. Consequently, DDP decreases immune system response to infections and haemorrhage [101]. A comparison study concluded that only 2 of 13 dose−intensity regimes of DDP had increased survival [83]. A combination of 600 mg/m^2 cyclophosphamide and cis−platin with dose intensity between 120−60 mg/m^2 led to an increase in 3−year survival to 60% at the cost of increased renal toxicity and neurotoxicity. In the case of 750 mg/m^2 cyclophosphamide with cis−platin, 50−100 mg/m^2 has been noted increased survival rate of 32,4% versus 26,6% and higher toxicity[83].

3.4.2 CBDCA

The severe side effects, especially nephrotoxicity, have driven the further the development, which led in the 1980s to the introduction of CBDCA. Analyses asserted that carboplatin is equally effective as DDP with a similar spectrum of clinical response but with less inconvenient side effects. The more attractive profile of CBDCA is based on the more stable leaving group [102]. CBDCA received approval from FDA in 1989 for clinical purpose against ovarian carcinoma.

The dosing regimen of carboplatin combined with 80 mg/m^2 of paclitaxel for ≥ 6 cycles had increased survival and (PFS). The overall response rate of Pt resistant patients was 58%, and the PFS median was estimated at 12 months[103]. Similarly to the case of DDP CBDCA's major product, counting 65% according to Bradley et al. (1993), is an intra−strand crosslink between adjacent guanines GpG at the N7 position [104]. The structure of CBDCA is like DDP except for the substitution of chloride ligands with carboxylate compounds, bidentate dicarboxylate, which is the leaving group [105].

Even though CBDCA is particularly effective in the fight against OC, its toxicity and mutagenic effect in cell culture and animal models remain the main limitation. The limiting dosage factors are not nephrotoxicity and ototoxicity but myelosuppression and thrombocytopenia [106].

3.4.3 L-OHP

L-OHP, (Trans−R, R)1,2−diaminocyclohexaneoxalatoplatinum (III) is the third generation of Pt complexes synthesised by Kindai in 1978 [107]. Charged ion of Pt surrounded by oxalate ligand

leaving group and (trans− R, R)1,2− diamino−cyclohexane carrier ligand, which form Pt−DNA adduct[108]. L-OHP shows higher hydrophobicity and greater size due to its ligand diaminocyclohexan, leading to stress induction in cells [109]. Thanks to these properties of L-OHP the modification of DNA could be more significant. L− OHP exerts effects by binding to DNA and forming an obstacle to cellular proteins and interferes with RNA synthesis[109].

It was licensed in 1999 by European Union and later in 2002 in the U.S., initially as a treatment for advanced colorectal cancer [110]. L-OHP creates a higher number of DNA double breaks (DBS) has been than in DDP exposed cells. The effectiveness of L-OHP in DDP resistant cells lies in distinct repair and damage recognition processes. The formation of DNA−Pt adduct lead to activating the signalling pathway that ends with apoptosis or with futile cycling [111].

The mismatch repair (MMR) in DDP is the main damage recognition repair process. Binding of the MMR complex, the cytotoxicity is increased, especially in the case of DDP adducts. L-OHP's adducts are not recognised by MMR, such as hMSH2, PMS2 and MutSα, which bind with greater affinity to DDP adducts than to L-OHP. Cells with defective hMLH1 or hMSH6 have increased chemoresistance and remarkably increased replicative bypass [111]. L-OHP has proven to be very efficient in DDP−resistant cells, but its main advantage is in significantly decreased nephrotoxicity. On the other hand, in most patients, new side effect arose, especially peripheral neurotoxicity, in most patient.

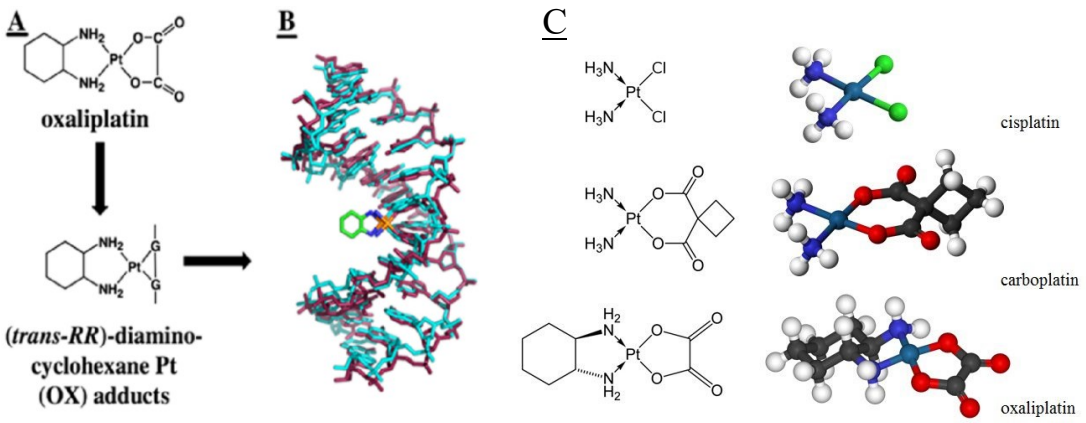

Figure 7: Structure of Pt compounds. A: Molecule of oxaliplatin and its DNA binding, creating GpG adduct. B: Bending of DNA helix after reacting with oxaliplatin. C: Structure of DDP, CBDCA and oxaliplatin. Source: Rageh et al. (2019) [112]

3.4.2 Paclitaxel

Paclitaxel is the mitogenic inhibitor, a diterpenoid compound that binds preferentially to microtubules and stabilises them. The antineoplastic effect of paclitaxel has the ability to

arrest G2/M−phase transition and modify pathways that induce apoptosis and senescence [113]. Paclitaxel consists of apoptosis through downstream effectors that include PI3K/RAC−α, AKT pathway, epidermal growth factor receptor and MAPK pathway [114] (*Figure 8)*.

Further development in clinical research for higher PFS and increased overall survival led to Pt combination therapy, which has shown to be highly effective especially in Pt resistant diseases. The tested overall response rate of DDP/paclitaxel therapy in the patients, untreated before, was 94% in 17 evaluable patients. In the case of patients with recurrent disease, the response was evaluated at 84% in 25 evaluable patients [115]. A test done on in vitro HGSOC culture treated by CBDCA/paclitaxel showed arrest in the G2/M phase of the cell cycle. Furthermore, there has been notably upregulated expression of p16, p21 and p53 proteins [113]. Altogether these changes promoted drug−induced premature senescence and displayed decreased expression of cyclin B1. During the senescence, there were changes in protein levels of STAT3, which engages cellular senescence. On the other hand, CBDCA/paclitaxel may help the tumour to develop a promoting phenotype by provoking their premature senescence [116].

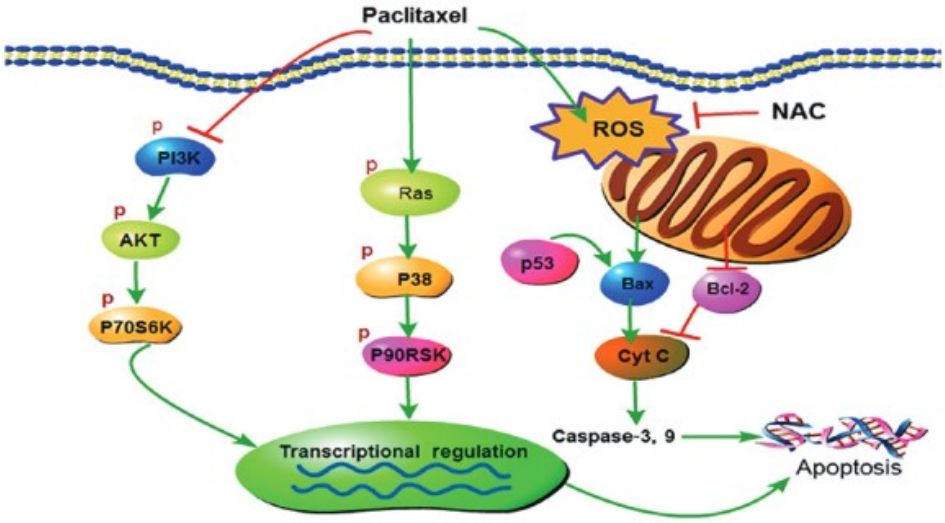

Figure 8: Mechanisms induced by paclitaxel. This compound targets multiple pathways, such as the AKT/MAPK signalling pathway, modulates mitochondrial membrane potential and inhibit PI3K/AKT pathway. Together, the paclitaxel action lead to apoptosis and premature senescence. Source: Ren et al. (2018)[113]

4. Exhibiting drug resistance to PtC

Patients with a good initial response to chemotherapy drugs in stage III/IV have a high risk of recurrence. Of all patents in stage III/IV, 70% relapse within 5-years due to developed chemoresistance, which is associated with poor outcome[117]. Reduction of a cytotoxic drug accumulation correlated with increased cytoplasmic detoxification, increased DNA repair activity, repair of inter−strand crosslinks or tolerance to Pt adducts are the main mechanisms of resistance development of HGSOC [118] [102]. The exhibiting of resistance is multifactorial but could be associated with specific pathways [118] (*Figure 9)*.

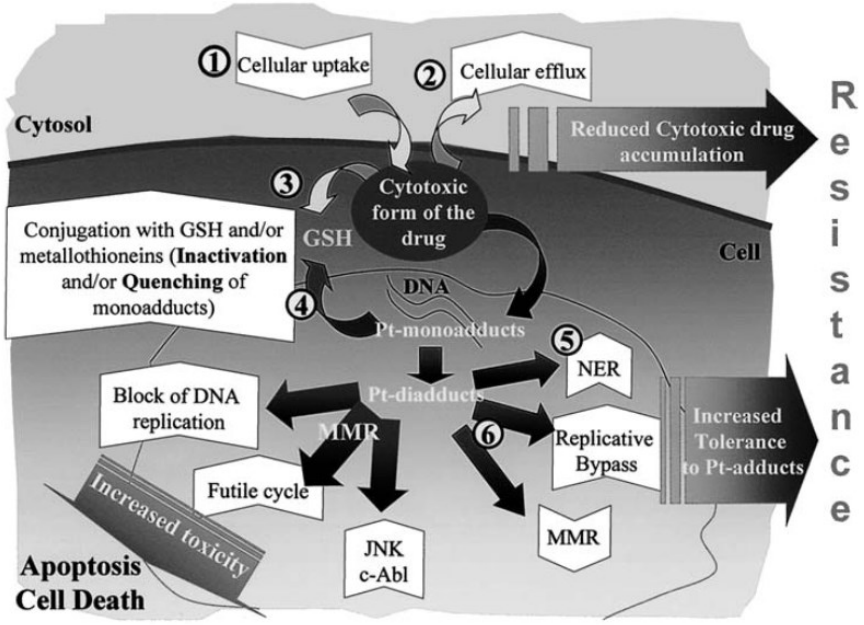

Figure 9: The schematic view of DDP resistance or increased toxicity mechanisms in cancer cells. 1) Cellular uptake of DDP is regulated by copper transporters. 2) Cellular efflux of DDP is mediated by multiple transporters 3) Inactivation of the DDP by conjunction with GSH, or anothermetallothioneins.4) Quenching of monoadducts. 5) Decreasing in MMR processes through nuclear excision repair (NER) that may enhance the tolerance to the action of DDP. Source: Di Francesco et al. (2002)[118]

4.1 Reduction of an accumulation of DDP in cells

One of the resistance mechanisms to DDP, CBDCA is to restrain the influx into the cell by losing substantial transporter CTR1. Loss of *hCTR1* was triggered by cells exposure to >2 mol/L of DDP; this level of DDP caused the almost complete disappearance of the transporter from the cell membrane. This loss could be blocked by inhibitors of endosomal pathways, including amiloride, cytochalasin D, nystatin and methyl−β−cyclodextrin. The amiloride block specifically the pathway of micropinocytosis. The effects of membrane transporters CTR1 correlate with the uptake of copper. CBDCA and copper accumulation were tested on *CTR1* −/− cells, and each

compound was individually measured and compared to *CTR1* wt cells. *CTR1* -/- cells exposed to 2µM copper during 1h has accumulated only 5.7 % of the amount that have accumulated wt cells. On behalf of CBDCA *CTR1-/-* cells accumulated only 35% to 36% of *CTR1 +/+.* The uptake of L−OHP is CTR1−independent according to the testing and could be clinically utilised on CTR1 $^{-/-}$ DDP & CBDCA resistant cells as a chemotherapy treatment [119].

The intracellular concentration is also reduced by the efflux of DDP from the cell. Proteins, ATPase ATP7A and ATP7B, mediators of the copper efflux, are involved in the modulation of the DDP export. Even a slight increment in the expression of one of the P−ATPases is sufficient to cause DDP and CBDCA and L−OHP resistance. Transfected ovarian carcinoma cells with ATP7B cDNA expression vector accumulated only 61% of CBDCA compared to the control group, thus exhibiting resistance. A study of Samimi et al. (2004) confirmed that ATP7A function for sequestration of the Pt drug into intracellular vesicles in the cell system therefore, limiting the possibility of the interaction of Pt drug with nucleus and mitochondrion [120]. In some resistant cell lines such as AMD473 the resistance is due to reduced formation of a glutathione-drug complex and efflux of complexes through GS-X pump, a glutathione complex dependent transporter [121](*Figure 10*).

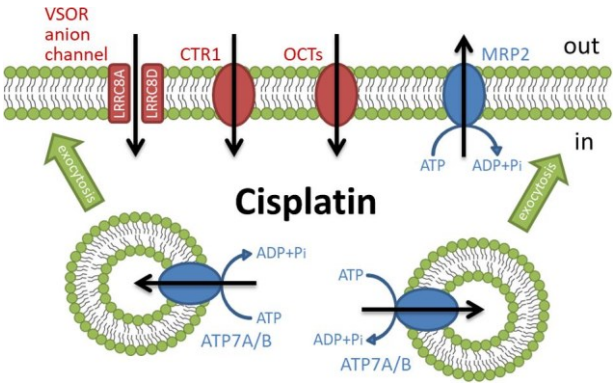

Figure 10: Cellular membrane with transporters that contribute to the DDP resistance by the regulation of DDP accumulation. ATP7A sequestrates Pt to vesicles. CTR1 and OCT regulate the uptake of DDP and CBDCA. MRP2 mediate the efflux of conjugated Pt compounds. Source: Shimizu et al. (2020)[122]

Some of the resistant cells had increased levels of thiol−containing molecules. Compounds with the thiol group contain sulphur that helps the detoxification as the sulphur binds Pt compounds. The levels of glutathione or metallo-thio compounds significantly correlated with resistance of O.C. cell lines, mouse cells and human O.C.s. However, in vitro studies by Kikuchi et al. (1998) has been stated that increased resistance is also associated with rising levels of glutathione−S−transferase activity, which leads to decreasing accumulation of DDP[123] (*Figure 11)*. The effect of reduced glutathione (GSH) is well noted 5 mM reduced GSH reduces the levels of platinated DNA approximately by 50% [121]. The conjunction of DDP and glutathione is promoted by Glutathione−S−transferase, and the efflux of these conjugates is mediated mainly by MRP family, especially MRP1 and MRP2 [124].

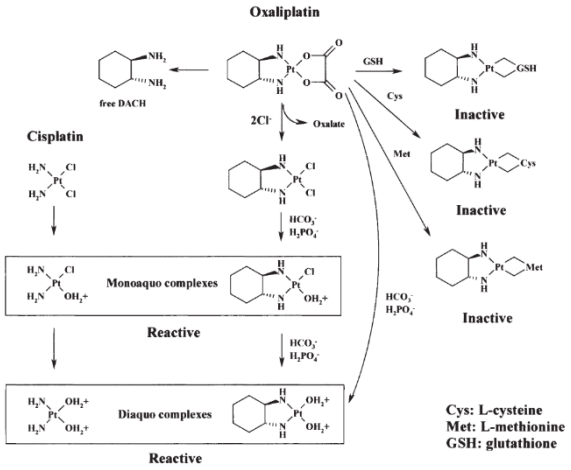

Figure 11: Major oxaliplatin and DDP metabolites. Shown on the pathways responsible for increased resistance to DDP and oxaliplatin in exposed cells. Conjunction of GSH, Methionine and Cystein to Pt inactivate the Pt cytotoxic activity. Source: Di Francesco et al. (2002)[118]

DDP could also undergo a side reaction with sulphur donors, that inactivates the DPP, or been trapped by the sulfhydryl group of GSH before the Pt cytotoxic action. In the end, the trapping and side reaction inhibits the DDP potency. GSH−mediated DDP resistance is due to the alteration of its biosynthetic route. DDP elevates mRNA levels of enzymatic activity of γ−glutamyl transpeptidase at a lower degree than L-OHP[118].

4.2 Modification of repair systems

Some cells have circumvented the effect of DDP through increased repair of DNA−adducts primarly through nuclear excision repair (NER) [118]. Resistance to DDP and CBDCA as they act in the same manner, form the same type of adducts, and are recognised by the same repair complexes specifically by MMR complexes. The loss of functional DNA MMR, either by mutation or hypermethylation of the *hMLH1* promoter leading the O.C. cells gain a resistance

[125]. There is a correlation between defective *hMLH1* and *hMSH6* genes (subunits of hMutSα) of MMR mechanism and the DDP resistance that has been increased 1,5−4,8 fold together with 2,5−6 fold increased replicative bypass [126]. The studied cell lines A2780/CP70 derived from human O.C. have shown deficiency due to the loss of *MLH1*, contributing to an increase of cell replicative bypass of DDP-DNA adducts [127]. The instability of microsatellites and loss of *hMLHI* in A2780 displayed resistance to DDP and CBDCA but no significant resistance to L-OHP as the DDP adducts are recognised by hMutSα/hMutLα proteins, which do not recognise DACH-Pt-DNA adducts [128].

NER pathway is enhanced in DDP resistant cells and plays a critical role as protective mechanism against Pt toxicity. Whereas NER does not discriminate between DDP−and DACH adducts, this mechanism of resistance could also work on L-OHP exposed cells [129]. The transcription-coupled NER subpathway is initiated by RNA polymerase 2, when lesion-stalling RNA polymerase 2 finds a DNA distortion or an inappropriate base it recruits UVSSA, CSA and CSB, which are essential for an assembly of downstream machinery [130]. The XPA, RPA and multisubunit protein complex TFIIH (especially core subunits XPD and XPB) are factors of NER pathway responsible for forming of a damage recognition subunits [118* 125]. The formed damage recognition subunit recruit endonucleases XPG and XPF−ERCC1 that are responsible for the excision of damage containing DNA. Testing of different factors in NER, base-excision repair, homologous recombination and translation DNA synthesis pathways discovered that gene *CSA* of transcription−coupled NER, *ERCC1/XPF, polymerase B/polymerase L* strongly interfere with the sensitivity of cells to L-OHP [130 131]. Furthermore, there is a possibility that base-excision repair and transcription−coupled NER act together to protect DNA against sustaining distortion and damage [130].

Exposure to DDP and LOH−P contributes to an immobilisation of XRCC1 which is involved in BER pathway and if exposed to L-OHP the XRCC1 engages in glycosylase-dependent BER pathway, where OGG1 could possibly recognise Pt-DNA crosslinks and subsequently remove it [130]. Using inhibitors on NER pathway, defecting NER capacity could lead in cells to contradict the DDP resistance [125] .

4.3 Tolerance of DNA damage

This process is also referred to as replication bypass or translation synthesis. Replicative bypass refers to an ability to functionally replicate DNA without disruption by DNA damage/crosslinks or by distortion/bending. Thus, there is no reparation of bulky adducts that usually blocks the complex during synthesis, but the replication goes past it. In some cases of

O.C. there has been noted a correlation between the increment of the replicative bypass past Pt−adducts in cells and resistance to DPP, or to a lesser extent to LOH−P [118] .

Cell ability to tolerate adducts is dependent on the nature of adducts, thus cells might be able to discriminate between cis−Pt-adducts and DACH-adducts due to their different geometrical and chemical properties. The defects in MLH1 and MSH6 proteins (components of MMR) are believed to collaborate with translation synthesis [132]. The replicative bypass is contributed by the activity of DNA polymerases, such as polymerase η, ι, κ, ζ and DNA polymerase β[133].

On the other hand, DNA polymerases α, ε, and δ have demonstrated no part in replicative bypass as GpG adducts completely block them. Defects in REV3, the catalytic subunit of polymerase ζ, and POLH have been connected to increased sensitivity to DDP[133]. The ability of replicative bypass of DNA polymerase β could be prior to its capacity to reinitiate DNA synthesis at the opposite side to cis−Pt-adduct as it frequently dissociates and reassociate with DNA. Furthermore, β polymerase could also elongate products of the arrested replication[134]. DNA polymerases β and η are well known for efficiently bypassing the adducts. Comparing β polymerase and η polymerase, polymerase η catalysed with at least 2−fold higher frequency of (−)1 frameshift deletion than polymerase β, approximately 50% of β polymerase's products contained single based deletion [135]. Furthermore, bypass products of η polymerases in cells exposed to L-OHP have shown multiple replication errors, such as mis-insertion and frameshifts[135]. Wu and colleagues (2021) have studied DNA lesion bypass using a chromatin immunoprecipitation−based assay and found elevated levels of ub−proliferating cell nuclear antigen and polymerase η in O.C. stem cells, suggesting enhanced trans-lesion DNA synthesis activity in O.C. stem cells [136].

5. New HGSOC treatments

Due to the grim 5-year survival rate of patients with HGSOC, there has been an intense global effort to find a new, more effective methods or drug combinations for treating HGSOC better than with the widely used PtC. After the discovering CBDCA, the development of the new generation of Pt compounds has not retarded and the research has brought new generations of Pt compounds. The satraplatin (bis−aceto−ammine−dichloro−cyclohexylamine) and picoplatin (2-methylpyridine azane) are representatives of the most recent generation of Pt compounds, they are utilised as the orally active version of CBDCA. However, they failed to either overcome the DDP resistance or did not extend the overall survival.

As has been mentioned before most of the HGSOC harvest somatic *Tp53* mutations. Their occurrence and oncogenic properties render them a good target for treatment. One of the drugs targeting *Tp53* is APR−246. APR-246 restores a transcriptional activity of p53, the toxicity profile was very tolerable, pharmacokinetics were favourable and it showed cell cycle arrest and increased apoptosis [137].

PTEN is mutated on minor occasions in HGSOC. As the deficit of PTEN causes HRD and makes the tumour cells susceptible to inhibitors of DNA repair enzyme poly (ADP−ribose) polymerase, PARP inhibitors [138]. Besides PTEN deficient tumour cells PARP inhibitors have been already demonstrated on *BRCA* deficient tumour cells using of olaparib, a high−potency PARP inhibitor[81]. Olaparib is currently in phase II of clinical trials for recurrent HGSOC. PARP is responsible for recruiting BER factors [139140]. *BRCA* mutant patients have increased formation of DSBs thanks to defects in DNA repair of homologous recombination. PARP inhibitors target HRD cells by increasing the formation of DBS. PARP inhibitors render homologous recognition pathway more ineffective. Niraparib is a highly selective inhibitor PARP1 and PARP2 and is used as maintenance therapy in patients with recurrent O.C., who had responded to PtC. Niraparib an orally administrated compound. The positive effect of niraparib on PFS has been proven regardless of alteration of *BRCA* genes and is especially effective in HRD−positive groups, but benefits have also been noted in 20% of patients from HRD−negative subgroup. Dose-limiting factor of PARP inhibitor was myelotoxicity and haematological abnormalities, which has been commonly demonstrated in 93 patients [141]. Niraparib prolonged the progression-free survival response in patients with complete response after Pt-chemotherapy[142].

The bevacizumab an anti-angiogenic agent, humanised monoclonal antibody that binds to all circulating, soluble VEGF−A. Bevacizumab inhibits the activation of VEGF signalling pathway by binding to soluble VEGF-A, which is important for tumour proliferation, invasiveness

and immune system suppression [143]. Alike cediranib, bevacizumab was introduced in clinical use more than 17 years ago [144]. Its use is limited due to safety concerns of increased incidence of hypertension. The use of maintenance PARP inhibitors and anti−angiogenic agents allows for chemotherapy-free combination regimes circumventing PtC. PARP inhibitors and bevacizumab could collaborate and improve clinical outcomes as bevacizumab induces hypoxia that could accelerate the production of DNA damage and genetic instability enhancing HRD and raising sensitivity to PARP inhibitors [139]. In the end, hypoxic cancer is more vulnerable to toxic effects. A combination of cediranib and olaparib, has shown remarkably longer PFS than using each agent alone (18 months vs. 9 months HR 0,42)[145]. Side effects of combination therapy were increased incidence of proteinuria and almost doubling incidence of all−grade hypertension [145].

There are currently lot of drugs in an examination, in clinical trials, for instance namely veliparib (oral PARP inhibitor in phase III of clinical trials in combination with CBDCA and paclitaxel), rucaparib (small inhibitor of PARP), talazoparib (PARP inhibitor), cyclophosphamide (immune system suppressor, in phase II in combination with veliparib), ipilimumab (anti−CTLA−4 antigen blocking cytotoxic inhibition), nivolumab (blocking PD−L1 binding of cytotoxic lymphocyte (*TABLE 2)*. [146] [147] [148] [149]. There is also ongoing research regarding the improvement of early detection of HGSOC. One of the candidates are miRNAs [150].

Drugs	Control	Aim	HR	CI	Reference
i.v. CBDCA (AUC 6) and paclitaxel 175 mg/m2 Q 21 Days x 3 Courses Plus Low Dose Paclitaxel 40 mg/m2/week	*i.v.* CBDCA (AUC 6) and paclitaxel 175 mg/m2 Q 21 Days x 3 Courses	Progression−free 5− year survival	0.807	0.565−1.150	NCT00003644, [151]
combination of trabectedin (1.1 mg/m^2/3weeks) with DOXIL (30mg/m^2/3weeks) and dexamethasone (20 mg)	DOXIL (50 mg/m^2/4weeks) monotherapy in patients with OC	PFS (3 year)	0.680	0.550−0.840	NCT00113607, [152]
lurbinectedin (PMO1183) (3.2 mg/m^2/3weeks)	Pogylated liposomal doxorubicin (PLD) (50 mg/m^2/4weeks) or topotecan (1.50 mg/m^2/3weeks) in Pt resistant OC	PFS (3 years)	1.057	0.854−1.309	NCT02421588, [153]

paclitaxe+ pertuzumab+ topotecan/ gemcitabine+ paclitaxel+ pertuzumab+ topotecan/ paclitaxe+ pertuzumab+ placebo/ paclitaxel+ pertuzumab + topotecan	gemcitabine (1000 mg/m^2/3weeks), paclitaxel (80 mg/m^2/3weeks), placebo, topotecan (1.25 mg/m^2/3weeks)	Overall survival rate	0.900	0.61−1.32	NCT01684878, [154]
niraparib (300mg/m^2/once daily in 28−day cycle)	placebo	PFS	*(BRCAm)=0.27; (HRD+)=0.38; (wt BRCA)=0.45*	(m*BRCA)=0.170−0.410*; (HRD+)=0.240−0.590; (wt *BRCA)=* 0.340−0.610	NCT01847274, [155]
avastin 15mg/kg/3weeks+ DOXIL(40mg/m^2/4weeks)+ paclitaxel (80mg/m^2/4x in 4weeks) + topotecan (4mg/m^2/3x in 4weeks)	DOXIL (40mg/m^2/4weeks)+ paclitaxel (80mg/m^2/4x in 4weeks)+ topotecan (4 mg/m^2/3x in4 weeks)	PFS	0.480	0.380−0.600	NCT00976911,[156]
tamoxifen (once daily 1−28 day)	thalidomide (twice daily on days 1−28)	PFS (3 years)	1.310	0.930−1.850	NCT00041080, [157]
paclitaxel (80mg/m^2/weekly), saracatinib (175 mg/m^2/once daily)	paclitaxel (80 mg/m2/weekly), Placebo	PFS (6 months)	1.000	0.650−1.540	NCT01196741, [158]
nintedanib (200mg), paclitaxel (175mg/m^2), CBDCA (5 mg/mL per min)	placebo, paclitaxel, CBDCA	PFS (29 months)	0.840	0.720−0.980	NCT01015118, [159]
mirvetuximab soravtnsine (6 mg/kg body weight)	paclitaxel (80 mg/m2/4x in 4weeks), PLD (40 mg/m2/once 4 weeks), topotecan (4mg/m2/3x in 4 weeks)	PFS (62.9−86.9 weeks)	0.980	0.730−1.310	NCT02631876, [160]
CBDCA (4mg/mL/min)+ gemcitabine (1000mg/m2) + bevacizumab (15mg/kg)	CBDCA (4mg/mL/minute)+ gemcitabine (1000mg/m2)+ Placebo	PFS (3 years, 5 months)	0.484	0.388−0.605	NCT00434642, [161]
pazopanib (800mg tablet daily for 24 months)	placebo	PFS (15 −19.7 months)	0.720	0.690−0.860	NCT00866697, [162]

paclitaxel (80 mg/m2/weekly)+ bevacizumab+ CBCDA i.v. // paclitaxel (80 mg/m2/weekly)+ CBDCA i.p.+ bevacizumab	paclitaxel (135 mg/m2/ once per 3 weeks) +bevacizumab (15 mg/kg/ every 3 weeks)+ DDP (75 mg/m2/day 2) i.p.	PFS (10 years)	(CBDCA)i.v.= 0.925; (Carboplatin) i.p.= 0.977	(CBDCA) i.v.= 0.802−1.07; (Carboplatin) i.p.= 0.847−1.130	NCT00951496, [163]
combination of trabectedin (1.1 mg/m2/3weeks) with DOXIL (30mg/m2/3weeks) and Dexamethasone (20 mg)	DOXIL (50 mg/m2/4weeks) monotherapy in patients with OC	Overall survival rate (4.3 years)	0.920	0.730−1.180	NCT01846611, [164]
paclitaxel, CBDCA, bevacizumab	placebo, paclitaxel, CBDCA	PFS (up to 6 years)	0.717	0.625−0.824	NCT00262847, [165]

TABLE 2: Completed clinical trials with results in phase III−IV of drugs for the O.C. treatment. Source: NLM, ClinicalTrials.gov: Studies of High grade serous ovarian carcinoma. Accessed from URL: <https://clinicaltrials.gov/ct2/results?cond=High+grade+serous+ovarian+carcinoma&term=&cntry=&state=&city=&dist=&recrs=a&recrs=d&recrs=e> [accessed on 12 April 2022]

6. Conclusion

HGSOC treatment is associated with slightly improved patient's PFS compared to previous decades. DDP are useful against many cancers, but their side effects and resistance surpass their profit. Therefore, the new generation of platinum compound as CBDCA, iproplatin, tetraplatin cycloplatam, L-OHP, satraplatin and nicoplatin, have been synthesised. Some DDP derivates failed in clinical trials. Those which passed, demonstrated lower toxicity and even prolonged overall survival. L-OHP and CBDCA were outstanding in clinical trials and are currently, especially CBDCA, a backbone of HGSOC treatment procedures. Trabectedin with PLD (trabectedine +PLD 18 months vs PLD 12 months), Niraparib (niraparib 22 months vs placebo six months) and nintedanib combined with CBDCA and paclitaxel (combination 17.2 vs 16.6 months placebo) showed somewhat longer PFS in clinical trials, but we are still awaiting a treatment that would shift HGSOC to the at least manageable disease. Even the new generation of Pt compounds does not obviate the resistance mechanisms. In order to avoid the resistance, it is necessary to elucidate each step of the resistance pathway. There has been made a great progress since the last century in understanding the resistance, but some mechanisms are still unclear [2].

The DDP use is, is insufficient for the treatment of patients with HGSOC and is substituted by other compounds. The future lies in chemo−free personalised treatment with new potential diagnostic and prognostic markers and technologies for diagnosis at stage I/II of HGSOC. As has been mentioned earlier, PDO could be a good tool for diagnostics patient’s malignancy’s molecular patterns but also as a guide for the optimal treatment combination as the PDO can foretell the response to each type of drug [166] [5]

List of literature

Secondary citations are marked with

1. Colombo, N. & Ledermann, J. A. Updated treatment recommendations for newly diagnosed epithelial ovarian carcinoma from the ESMO Clinical Practice Guidelines. *Ann. Oncol.* **32**, 1300–1303 (2021).
2. Kelland, L. R. New platinum antitumor complexes. *Crit. Rev. Oncol. Hematol.* **15**, 191–219 (1993)*.
3. Lee, M. *et al.* Clinical Significance of CA125 Level after the First Cycle of Chemotherapy on Survival of Patients with Advanced Ovarian Cancer. *Yonsei Med. J.* **57**, 580–587 (2016).
4. Kim, J.-H. *et al.* Osteopontin as a Potential Diagnostic Biomarker for Ovarian Cancer. *JAMA* **287**, 1671–1679 (2002).
5. Nero, C. *et al.* Patient-derived organoids and high grade serous ovarian cancer: from disease modeling to personalized medicine. *J. Exp. Clin. Cancer Res.* **40**, 116 (2021).
6. OECD. *Deaths from cancer*. https://www.oecd-ilibrary.org/social-issues-migration-health/deaths-from-cancer/indicator/english_8ea65c4b-en (2018).
7. Health status - Deaths from cancer - OECD Data. *theOECD* http://data.oecd.org/healthstat/deaths-from-cancer.htm.
8. Sung, H. *et al.* Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. *CA. Cancer J. Clin.* **71**, 209–249 (2021).
9. Den boje proti rakovině a statistiky ÚZIS ČR - Aktuality - ÚZIS ČR. https://www.uzis.cz/index.php?pg=aktuality&aid=8466.
10. Allemani, C. *et al.* Global surveillance of cancer survival 1995–2009: analysis of individual data for 25 676 887 patients from 279 population-based registries in 67 countries (CONCORD-2). *Lancet Lond. Engl.* **385**, 977–1010 (2015).
11. Matsuo, K. *et al.* Evolving population-based statistics for rare epithelial ovarian cancers. *Gynecol. Oncol.* **157**, 3–11 (2020)*.
12. Berns, E. M. J. J. & Bowtell, D. D. The Changing View of High-Grade Serous Ovarian Cancer. *Cancer Res.* **72**, 2701–2704 (2012)*.
13. Lheureux, S., Gourley, C., Vergote, I. & Oza, A. M. Epithelial ovarian cancer. *The Lancet* **393**, 1240–1253 (2019).
14. Chen, H.-H., Wang, Y.-C. & Fann, M.-J. Identification and Characterization of the CDK12/Cyclin L1 Complex Involved in Alternative Splicing Regulation. *Mol. Cell. Biol.* **26**, 2736–2745 (2006).
15. Goff, B. Symptoms Associated With Ovarian Cancer. *Clin. Obstet. Gynecol.* **55**, 36–42 (2012).
16. Peres, L. C. *et al.* Histotype classification of ovarian carcinoma: A comparison of approaches. *Gynecol. Oncol.* **151**, 53–60 (2018).
17. Torre, L. A. *et al.* Ovarian cancer statistics, 2018. *CA. Cancer J. Clin.* **68**, 284–296 (2018).
18. Prat, J. Ovarian carcinomas: five distinct diseases with different origins, genetic alterations, and clinicopathological features. *Virchows Arch.* **460**, 237–249 (2012)*.
19. Coleman, M. *et al.* Cancer survival in Australia, Canada, Denmark, Norway, Sweden, and the UK, 1995–2007 (the International Cancer Benchmarking Partnership): an analysis of population-based cancer registry data. *Lancet* **377**, 127–138 (2011).
20. Yang, H. P. *et al.* Ovarian cancer risk factors by histologic subtypes in the NIH-AARP diet and health study. *Int. J. Cancer* **131**, 938–948 (2012).

21. Colombo, N. *et al.* Endometrial cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up†. *Ann. Oncol.* **24**, vi33–vi38 (2013).
22. Fotopoulou, C. *et al.* British Gynaecological Cancer Society (BGCS) epithelial ovarian/fallopian tube/primary peritoneal cancer guidelines: recommendations for practice. *Eur. J. Obstet. Gynecol. Reprod. Biol.* **213**, 123–139 (2017).
23. Kang, J.-H. *et al.* Clinical factors associated with prognosis in low-grade serous ovarian carcinoma: experiences at two large academic institutions in Korea and Taiwan. *Sci. Rep.* **10**, 20012 (2020).
24. Gaitskell, K. *et al.* Ovarian cancer survival by stage, histotype, and pre-diagnostic lifestyle factors, in the prospective UK Million Women Study. *Cancer Epidemiol.* **76**, 102074 (2022).
25. Malpica, A. *et al.* Grading Ovarian Serous Carcinoma Using a Two-Tier System. *Am. J. Surg. Pathol.* **28**, 496–504 (2004).
26. Hoffman, J. G. Theory of the mitotic index and its application to tissue growth measurement. *Bull. Math. Biophys.* **11**, 139–144 (1949).
27. Smith Sehdev, A. E., Sehdev, P. S. & Kurman, R. J. Noninvasive and Invasive Micropapillary (Low-Grade) Serous Carcinoma of the Ovary: A Clinicopathologic Analysis of 135 Cases. *Am. J. Surg. Pathol.* **27**, 725–736 (2003).
28. Gilks, C. B. & Prat, J. Ovarian carcinoma pathology and genetics: recent advances. *Hum. Pathol.* **40**, 1213–1223 (2009)*.
29. Bell, D. A. & Scully, R. E. Early de novo ovarian carcinoma. A study of fourteen cases. *Cancer* **73**, 1859–1864 (1994).
30. Kurman, R. J. & Shih, I.-M. The Origin and Pathogenesis of Epithelial Ovarian Cancer- a Proposed Unifying Theory. *Am. J. Surg. Pathol.* **34**, 433–443 (2010).
31. Lawrenson, K. *et al.* Integrated Molecular Profiling Studies to Characterize the Cellular Origins of High-Grade Serous Ovarian Cancer. 330597 (2018) doi:10.1101/330597.
32. Hao, D. *et al.* Integrated Analysis Reveals Tubal- and Ovarian-Originated Serous Ovarian Cancer and Predicts Differential Therapeutic Responses. *Clin. Cancer Res.* **23**, 7400–7411 (2017).
33. Ozcan, A., Liles, N., Coffey, D., Shen, S. S. & Truong, L. D. PAX2 and PAX8 Expression in Primary and Metastatic Müllerian Epithelial Tumors: A Comprehensive Comparison. *Am. J. Surg. Pathol.* **35**, 1837–1847 (2011).
34. Zhang, S. *et al.* Both fallopian tube and ovarian surface epithelium are cells-of-origin for high-grade serous ovarian carcinoma. *Nat. Commun.* **10**, 5367 (2019).
35. Neesham, D., Richards, A. & McGauran, M. Advances in epithelial ovarian cancer. *Aust. J. Gen. Pract.* **49**, 665–669 (2020).
36. Perets, R. *et al.* Transformation of the Fallopian Tube Secretory Epithelium Leads to High-grade Serous Ovarian Cancer in Brca;Tp53;Pten Models. *Cancer Cell* **24**, 751–765 (2013).
37. Marquez, R. T. *et al.* Patterns of gene expression in different histotypes of epithelial ovarian cancer correlate with those in normal fallopian tube, endometrium, and colon. *Clin. Cancer Res. Off. J. Am. Assoc. Cancer Res.* **11**, 6116–6126 (2005).
38. Xiang, L., Rong, G., Zhao, J., Wang, Z. & Shi, F. Identification of candidate genes associated with tubal origin of high-grade serous ovarian cancer. *Oncol. Lett.* **15**, 7769–7775 (2018).
39. Lo Riso, P. *et al.* A cell-of-origin epigenetic tracer reveals clinically distinct subtypes of high-grade serous ovarian cancer. *Genome Med.* **12**, 94 (2020).
40. Menon, U., Griffin, M. & Gentry-Maharaj, A. Ovarian cancer screening—Current status, future directions. *Gynecol. Oncol.* **132**, 490–495 (2014).

41. Jacobs, I. J. *et al.* Ovarian cancer screening and mortality in the UK Collaborative Trial of Ovarian Cancer Screening (UKCTOCS): a randomised controlled trial. *The Lancet* **387**, 945–956 (2016).
42. Jacobs, I. J. & Menon, U. Progress and Challenges in Screening for Early Detection of Ovarian Cancer *. *Mol. Cell. Proteomics* **3**, 355–366 (2004).
43. Cramer, D. W. *et al.* Ovarian Cancer Biomarker Performance in Prostate, Lung, Colorectal, and Ovarian Cancer Screening Trial Specimens. *Cancer Prev. Res. Phila. Pa* **4**, 365–374 (2011).
44. Kabawat, S. E. *et al.* Tissue Distribution of a Coelomic- Epithelium-Related Antigen Recognized by the Monoclonal Antibody OC125. *Int. J. Gynecol. Pathol.* **2**, 275–285 (1983).
45. Bast, R. C. *et al.* A radioimmunoassay using a monoclonal antibody to monitor the course of epithelial ovarian cancer. *N. Engl. J. Med.* **309**, 883–887 (1983).
46. Zhang, M., Zhang, Y., Fu, J. & Zhang, L. Chapter Thirteen - Serum CA125 levels are decreased in rectal cancer but increased in fibrosis-associated diseases and in most types of cancers. in *Progress in Molecular Biology and Translational Science* (ed. Zhang, L.) vol. 162 241–252 (Academic Press, 2019)*.
47. Kobayashi, H., Tamura, M., Satoh, T. & Terao, T. Clinical evaluation of new cancer-associated antigen CA125 II in epithelial ovarian cancers: Comparison with CA125. *Clin. Biochem.* **26**, 213–219 (1993).
48. Fritsche, H. A. & Bast, R. C. CA 125 in Ovarian Cancer: Advances and Controversy. *Clin. Chem.* **44**, 1379–1380 (1998).
49. Mariampillai, A. I. *et al.* Cancer Antigen 72-4 for the Monitoring of Advanced Tumors of the Gastrointestinal Tract, Lung, Breast and Ovaries. *Anticancer Res.* **37**, 3649–3656 (2017).
50. Kobayashi, H. [The clinical usefulness of serum CA72-4 analysis in patients with ovarian cancer]. *Nihon Sanka Fujinka Gakkai Zasshi* **41**, 585–589 (1989).
51. Ohuchi, N. *et al.* [Levels of circulating tumor-associated glycoprotein (TAG-72) in patients with carcinoma using a novel tumor marker, CA 72-4]. *Gan To Kagaku Ryoho* **15**, 2767–2772 (1988).
52. Zhao, P. *et al.* LPA3 is a precise therapeutic target and potential biomarker for ovarian cancer. *Med. Oncol.* **39**, 17 (2022).
53. Xu, Y. *et al.* Lysophosphatidic Acid as a Potential Biomarker for Ovarian and Other Gynecologic Cancers. *JAMA* **280**, 719–723 (1998).
54. Sedláková, I., Vávrová, J., Tošner, J. & Hanousek, L. Lysophosphatidic acid (LPA)—a perspective marker in ovarian cancer. *Tumour Biol. J. Int. Soc. Oncodevelopmental Biol. Med.* **32**, 311–316 (2011).
55. Minis, E. *et al.* Evaluation of lysophosphatidic acid in vaginal fluid as a biomarker for ovarian cancer: A pilot study. *Eur. J. Obstet. Gynecol. Reprod. Biol. X* **2**, 100012 (2019).
56. Mok, S. C. *et al.* Prostasin, a Potential Serum Marker for Ovarian Cancer: Identification Through Microarray Technology. *JNCI J. Natl. Cancer Inst.* **93**, 1458–1464 (2001).
57. Al-Dossary, A. A. *et al.* Engineered EV-Mimetic Nanoparticles as Therapeutic Delivery Vehicles for High-Grade Serous Ovarian Cancer. *Cancers* **13**, 3075 (2021).
58. Chang, L. *et al.* Liquid biopsy in ovarian cancer: recent advances in circulating extracellular vesicle detection for early diagnosis and monitoring progression. *Theranostics* **9**, 4130–4140 (2019).
59. Buys, S. S. *et al.* Effect of Screening on Ovarian Cancer Mortality: The Prostate, Lung, Colorectal and Ovarian (PLCO) Cancer Screening Randomized Controlled Trial. *JAMA* **305**, 2295–2303 (2011).

60. Terry, K. L. *et al.* A Prospective Evaluation of Early Detection Biomarkers for Ovarian Cancer in the European EPIC Cohort. *Clin. Cancer Res.* **22**, 4664–4675 (2016).
61. Integrated genomic analyses of ovarian carcinoma. *Nature* **474**, 609–615 (2011).
62. Song, H. *et al.* Common variants in RB1 gene and risk of invasive ovarian cancer. *Cancer Res.* **66**, 10220–10226 (2006).
63. Patch, A.-M. *et al.* Whole–genome characterization of chemoresistant ovarian cancer. *Nature* **521**, 489–494 (2015).
64. Karst, A. M. *et al.* Cyclin E1 deregulation occurs early in secretory cell transformation to promote formation of fallopian tube derived high-grade serous ovarian cancers. *Cancer Res.* **74**, 1141–1152 (2014).
65. Baker, V. V. *et al.* c-myc amplification in ovarian cancer. *Gynecol. Oncol.* **38**, 340–342 (1990).
66. Nelson, L. *et al.* A living biobank of ovarian cancer ex vivo models reveals profound mitotic heterogeneity. *Nat. Commun.* **11**, 822 (2020).
67. Shen, Y.-A. *et al.* Inhibition of the MYC-regulated glutaminase metabolic axis is an effective synthetic lethal approach for treating chemoresistant cancers. *Cancer Res.* **80**, 4514–4526 (2020).
68. Jung, M. *et al.* Clinical Importance of Myc Family Oncogene Aberrations in Epithelial Ovarian Cancer. *JNCI Cancer Spectr.* **2**, pky047 (2018).
69. Shieh, S.-Y., Ikeda, M., Taya, Y. & Prives, C. DNA Damage-Induced Phosphorylation of p53 Alleviates Inhibition by MDM2. *Cell* **91**, 325–334 (1997).
70. Leroy, B., Anderson, M. & Soussi, T. TP53 Mutations in Human Cancer: Database Reassessment and Prospects for the Next Decade. *Hum. Mutat.* **35**, 672–688 (2014).
71. Ghezelayagh, T. S. *et al.* Characterizing TP53 mutations in ovarian carcinomas with and without concurrent BRCA1 or BRCA2 mutations. *Gynecol. Oncol.* **160**, 786–792 (2021).*
72. Lee, S. *et al.* Molecular Analysis of Clinically Defined Subsets of High-Grade Serous Ovarian Cancer. *Cell Rep.* **31**, (2020).
73. Doherty, J. A. *et al.* Challenges and Opportunities in Studying the Epidemiology of Ovarian Cancer Subtypes. *Curr. Epidemiol. Rep.* **4**, 211–220 (2017).
74. Bieging, K. T., Mello, S. S. & Attardi, L. D. Unravelling mechanisms of p53-mediated tumour suppression. *Nat. Rev. Cancer* **14**, 359–370 (2014)*.
75. Bronder, D. *et al.* TP53 loss initiates chromosomal instability in fallopian tube epithelial cells. *Dis. Model. Mech.* **14**, dmm049001 (2021).
76. Sullivan, K. D., Galbraith, M. D., Andrysik, Z. & Espinosa, J. M. Mechanisms of transcriptional regulation by p53. *Cell Death Differ.* **25**, 133–143 (2018).
77. Bowtell, D. D. L. The genesis and evolution of high-grade serous ovarian cancer. *Nat. Rev. Cancer* **10**, 803–808 (2010)*.
78. Kim, S. I. *et al.* Effect of BRCA mutational status on survival outcome in advanced-stage high-grade serous ovarian cancer. *J. Ovarian Res.* **12**, 40 (2019).
79. Norquist, B. *et al.* Secondary Somatic Mutations Restoring BRCA1/2 Predict Chemotherapy Resistance in Hereditary Ovarian Carcinomas. *J. Clin. Oncol.* **29**, 3008–3015 (2011).
80. Shen, W. H. *et al.* Essential Role for Nuclear PTEN in Maintaining Chromosomal Integrity. *Cell* **128**, 157–170 (2007).
81. Mendes-Pereira, A. M. *et al.* Synthetic lethal targeting of PTEN mutant cells with PARP inhibitors. *EMBO Mol. Med.* **1**, 315–322 (2009).
82. Martins, F. C. *et al.* Combined image and genomic analysis of high-grade serous ovarian cancer reveals PTEN loss as a common driver event and prognostic classifier. *Genome Biol.* **15**, 526 (2014).

83. van der Burg, M. E. L., Boere, I. A. & Berns, P. M. J. J. Dose-dense therapy is of benefit in primary treatment of ovarian cancer: contra. *Ann. Oncol.* **22**, viii33–viii39 (2011).
84. Bowtell, D. D. *et al.* Rethinking ovarian cancer II: reducing mortality from high-grade serous ovarian cancer. *Nat. Rev. Cancer* **15**, 668–679 (2015).*
85. Schorge, J. O., Bregar, A. J., Durfee, J. & Berkowitz, R. S. Meigs to modern times: The evolution of debulking surgery in advanced ovarian cancer. *Gynecol. Oncol.* **149**, 447–454 (2018).*
86. Bristow, R. E., Tomacruz, R. S., Armstrong, D. K., Trimble, E. L. & Montz, F. J. Survival Effect of Maximal Cytoreductive Surgery for Advanced Ovarian Carcinoma During the Platinum Era: A Meta-Analysis. *J. Clin. Oncol.* (2016) doi:10.1200/JCO.2002.20.5.1248.
87. Dasari, S. & Tchounwou, P. B. Cisplatin in cancer therapy: molecular mechanisms of action. *Eur. J. Pharmacol.* **740**, 364–378 (2014).*
88. Brozovic, A., Ambriović-Ristov, A. & Osmak, M. The relationship between cisplatin-induced reactive oxygen species, glutathione, and BCL-2 and resistance to cisplatin. *Crit. Rev. Toxicol.* **40**, 347–359 (2010).
89. Rabik, C. A. & Dolan, M. E. Molecular mechanisms of resistance and toxicity associated with platinating agents. *Cancer Treat. Rev.* **33**, 9–23 (2007).
90. Del Bello, B., Valentini, M. A., Zunino, F., Comporti, M. & Maellaro, E. Cleavage of Bcl-2 in oxidant- and cisplatin-induced apoptosis of human melanoma cells. *Oncogene* **20**, 4591–4595 (2001).
91. Miller, R. P., Tadagavadi, R. K., Ramesh, G. & Reeves, W. B. Mechanisms of Cisplatin Nephrotoxicity. *Toxins* **2**, 2490–2518 (2010).
92. Jia, J. *et al.* BNIP3 contributes to cisplatin-induced apoptosis in ovarian cancer cells. *FEBS Open Bio* **10**, 1463–1473 (2020).
93. Ishida, S., Lee, J., Thiele, D. J. & Herskowitz, I. Uptake of the anticancer drug cisplatin mediated by the copper transporter Ctr1 in yeast and mammals. *Proc. Natl. Acad. Sci. U. S. A.* **99**, 14298–14302 (2002).
94. Cleare, M. J. & Hoeschele, J. D. Studies on the antitumor activity of group VIII transition metal complexes. Part I. Platinum (II) complexes. *Bioinorg. Chem.* **2**, 187–210 (1973).
95. Monnet, J. & Kozelka, J. Cisplatin GG-crosslinks within single-stranded DNA: Origin of the preference for left-handed helicity. *J. Inorg. Biochem.* **115**, 106–112 (2012).
96. Plooy, A. C. M., van Dijk, M. & Lohman, P. H. M. Induction and Repair of DMA Cross-Links in Chinese Hamster Ovary Cells Treated with Various Platinum Coordination Compounds in Relation to Platinum Binding to DMA, Cytotoxicity, Mutagenicity, and Antitumor Activity. **44**, 9 (1984).
97. Sherman, S. E., Gibson, Dan., Wang, A. H. J. & Lippard, S. J. Crystal and molecular structure of cis-[Pt(NH3)2[d(pGpG)]], the principal adduct formed by cis-diamminedichloroplatinum(II) with DNA. *J. Am. Chem. Soc.* **110**, 7368–7381 (1988).
98. PDB101: Molecule of the Month: Cisplatin and DNA. *RCSB: PDB-101* http://pdb101.rcsb.org/motm/255.
99. Rosenberg, B., Van Camp, L. & Krigas, T. Inhibition of Cell Division in Escherichia coli by Electrolysis Products from a Platinum Electrode. *Nature* **205**, 698–699 (1965).
100. Kelland, L. Broadening the clinical use of platinum drug-based chemotherapy with new analogues. Satraplatin and picoplatin. *Expert Opin. Investig. Drugs* **16**, 1009–1021 (2007)*.
101. Volarevic, V. *et al.* Molecular mechanisms of cisplatin-induced nephrotoxicity: a balance on the knife edge between renoprotection and tumor toxicity. *J. Biomed. Sci.* **26**, 25 (2019).*

102. Kelland, L. The resurgence of platinum-based cancer chemotherapy. *Nat. Rev. Cancer* **7**, 573–584 (2007)*.
103. Cadron, I. *et al.* The “Leuven” dose-dense paclitaxel/carboplatin regimen in patients with recurrent ovarian cancer. *Gynecol. Oncol.* **106**, 354–361 (2007).
104. Bradley, L. J. N., Yarema, K. J., Lippard, S. J. & Essigmann, J. M. Mutagenicity and genotoxicity of the major DNA adduct of the antitumor drug cis-diamminedichloroplatinum(II). *Biochemistry* **32**, 982–988 (1993).
105. Knox, R. J., Friedlos, F., Lydall, D. A. & Roberts, J. J. Mechanism of Cytotoxicity of Anticancer Platinum Drugs: Evidence That cis-Diamminedichloroplatinum(II) and cis-Diammine-(1,1-cyclobutanedicarboxylato)platinum(II) Differ Only in the Kinetics of Their Interaction with DNA1. *Cancer Res.* **46**, 1972–1979 (1986).
106. Cheung-Ong, K., Giaever, G. & Nislow, C. DNA-Damaging Agents in Cancer Chemotherapy: Serendipity and Chemical Biology. *Chem. Biol.* **20**, 648–659 (2013).
107. Kidani, Y., Noji, M. & Tashiro, T. Antitumor activity of platinum(II) complexes of 1,2-diamino-cyclohexane isomers. *Gan* (1980) doi:10.20772/CANCERSCI1959.71.5_637.
108. Alian, O. M., Azmi, A. S. & Mohammad, R. M. Network insights on oxaliplatin anti-cancer mechanisms. *Clin. Transl. Med.* **1**, e26 (2012).
109. Sutton, E. C. *et al.* Nucleolar Stress Induction by Oxaliplatin and Derivatives. *J. Am. Chem. Soc.* **141**, 18411–18415 (2019).
110. Bhargava, A. & Vaishampayan, U. N. Satraplatin: leading the new generation of oral platinum agents. *Expert Opin. Investig. Drugs* **18**, 1787–1797 (2009).*
111. Vaisman, A. *et al.* The role of hMLH1, hMSH3, and hMSH6 defects in cisplatin and oxaliplatin resistance: correlation with replicative bypass of platinum-DNA adducts. *Cancer Res.* **58**, 3579–3585 (1998).
112. Rageh, M. M., Mohamad, E. A. & El-Garhy, M. R. Magnetic Field Improves the Efficiency of Low Dose Cis-Platin by Alteration the Oxidative Stress in Ehrlich Carcinoma-Bearing Mice. *Egypt. Acad. J. Biol. Sci. B Zool.* **11**, 107–112 (2019).
113. Ren, X. *et al.* Paclitaxel suppresses proliferation and induces apoptosis through regulation of ROS and the AKT/MAPK signaling pathway in canine mammary gland tumor cells. *Mol. Med. Rep.* **17**, 8289–8299 (2018).
114. Okano J, null & Rustgi, A. K. Paclitaxel induces prolonged activation of the Ras/MEK/ERK pathway independently of activating the programmed cell death machinery. *J. Biol. Chem.* **276**, 19555–19564 (2001).
115. de Jongh, F. E. *et al.* Dose-dense cisplatin/paclitaxel: a well-tolerated and highly effective chemotherapeutic regimen in patients with advanced ovarian cancer. *Eur. J. Cancer* **38**, 2005–2013 (2002).
116. Uruski, P. *et al.* Primary high-grade serous ovarian cancer cells are sensitive to senescence induced by carboplatin and paclitaxel in vitro. *Cell. Mol. Biol. Lett.* **26**, 44 (2021).
117. Kim, J. Y., Cho, C. H. & Song, H. S. Targeted therapy of ovarian cancer including immune check point inhibitor. *Korean J. Intern. Med.* **32**, 798–804 (2017).
118. Di Francesco, A. M., Ruggiero, A. & Riccardi, R. Cellular and molecular aspects of drugs of the future: Oxaliplatin. *Cell. Mol. Life Sci. CMLS* **59**, 1914–27 (2002).*
119. Petris, M. J., Smith, K., Lee, J. & Thiele, D. J. Copper-stimulated Endocytosis and Degradation of the Human Copper Transporter, hCtr1 *. *J. Biol. Chem.* **278**, 9639–9646 (2003).
120. Samimi, G. *et al.* Increased Expression of the Copper Efflux Transporter ATP7A Mediates Resistance to Cisplatin, Carboplatin, and Oxaliplatin in Ovarian Cancer Cells. *Clin. Cancer Res.* **10**, 4661–4669 (2004).

121. Holford, J., Beale, P. J., Boxall, F. E., Sharp, S. Y. & Kelland, L. R. Mechanisms of drug resistance to the platinum complex ZD0473 in ovarian cancer cell lines. *Eur. J. Cancer Oxf. Engl. 1990* **36**, 1984–1990 (2000).
122. Shimizu, T., Fujii, T. & Sakai, H. The Relationship Between Actin Cytoskeleton and Membrane Transporters in Cisplatin Resistance of Cancer Cells. *Front. Cell Dev. Biol.* **8**, (2020).
123. De Luca, A. *et al.* A structure-based mechanism of cisplatin resistance mediated by glutathione transferase P1-1. *Proc. Natl. Acad. Sci. U. S. A.* **116**, 13943–13951 (2019).
124. Nakayama, K. *et al.* Copper-transporting P-type adenosine triphosphatase (ATP7B) as a cisplatin based chemoresistance marker in ovarian carcinoma: Comparative analysis with expression of MDR1, MRP1, MRP2, LRP and BCRP. *Int. J. Cancer* **101**, 488–495 (2002).
125. Rocha, C. R. R., Silva, M. M., Quinet, A., Cabral-Neto, J. B. & Menck, C. F. M. DNA repair pathways and cisplatin resistance: an intimate relationship. *Clinics* **73**, e478s (2018).
126. Vaisman, A. *et al.* The Role of hMLH1, hMSH3, and hMSH6 Defects in Cisplatin and Oxaliplatin Resistance: Correlation with Replicative Bypass of Platinum-DNA Adducts1. *Cancer Res.* **58**, 3579–3585 (1998).
127. Drummond, J. T., Anthoney, A., Brown, R. & Modrich, P. Cisplatin and adriamycin resistance are associated with MutLalpha and mismatch repair deficiency in an ovarian tumor cell line. *J. Biol. Chem.* **271**, 19645–19648 (1996).
128. Fink, D. *et al.* The role of DNA mismatch repair in platinum drug resistance. *Cancer Res.* **56**, 4881–4886 (1996).
129. Reardon, J. T., Vaisman, A., Chaney, S. G. & Sancar, A. Efficient Nucleotide Excision Repair of Cisplatin, Oxaliplatin, and Bis-aceto-ammine-dichloro-cyclohexylamine-platinum(IV) (JM216) PlatinumIntrastrand DNA Diadducts1. *Cancer Res.* **59**, 3968–3971 (1999).
130. Slyskova, J. *et al.* Base and nucleotide excision repair facilitate resolution of platinum drugs-induced transcription blockage. *Nucleic Acids Res.* **46**, 9537–9549 (2018).
131. Duan, M., Ulibarri, J., Liu, K. J. & Mao, P. Role of Nucleotide Excision Repair in Cisplatin Resistance. *Int. J. Mol. Sci.* **21**, 9248 (2020).
132. Galluzzi, L. *et al.* Molecular mechanisms of cisplatin resistance. *Oncogene* **31**, 1869–1883 (2012).*
133. Shachar, S. *et al.* Two-polymerase mechanisms dictate error-free and error-prone translesion DNA synthesis in mammals. *EMBO J.* **28**, 383–393 (2009).
134. Hoffmann, J. S. *et al.* DNA polymerase beta bypasses in vitro a single d(GpG)-cisplatin adduct placed on codon 13 of the HRAS gene. *Proc. Natl. Acad. Sci.* **92**, 5356–5360 (1995).
135. Bassett, E. *et al.* Frameshifts and deletions during in vitro translesion synthesis past Pt–DNA adducts by DNA polymerases β and η. *DNA Repair* **1**, 1003–1016 (2002).
136. Wu, D. *et al.* Determination of DNA lesion bypass using a ChIP-based assay. *DNA Repair* **108**, 103230 (2021).
137. Lehmann, S. *et al.* Targeting p53 in Vivo: A First-in-Human Study With p53-Targeting Compound APR-246 in Refractory Hematologic Malignancies and Prostate Cancer. *J. Clin. Oncol.* **30**, 3633–3639 (2012).
138. Martins, F. C. *et al.* Combined image and genomic analysis of high-grade serous ovarian cancer reveals PTEN loss as a common driver event and prognostic classifier. *Genome Biol.* **15**, 526 (2014).
139. Hegan, D. C. *et al.* Inhibition of poly(ADP-ribose) polymerase down-regulates BRCA1 and RAD51 in a pathway mediated by E2F4 and p130. *Proc. Natl. Acad. Sci. U. S. A.* **107**, 2201–2206 (2010).

140. Matulonis, U. A. *et al.* Olaparib monotherapy in patients with advanced relapsed ovarian cancer and a germline BRCA1/2 mutation: a multistudy analysis of response rates and safety. *Ann. Oncol.* **27**, 1013–1019 (2016).
141. Mirza, M. R. *et al.* Niraparib Maintenance Therapy in Platinum-Sensitive, Recurrent Ovarian Cancer. *N. Engl. J. Med.* **375**, 2154–2164 (2016).
142. González-Martín, A. *et al.* Niraparib in Patients with Newly Diagnosed Advanced Ovarian Cancer. *N. Engl. J. Med.* **381**, 2391–2402 (2019).
143. Marchetti, C., Muzii, L., Romito, A. & Benedetti Panici, P. First-line treatment of women with advanced ovarian cancer: focus on bevacizumab. *OncoTargets Ther.* **12**, 1095–1103 (2019).
144. Garcia, J. *et al.* Bevacizumab (Avastin®) in cancer treatment: A review of 15 years of clinical experience and future outlook. *Cancer Treat. Rev.* **86**, (2020).
145. Liu, J. F. *et al.* Combination cediranib and olaparib versus olaparib alone for women with recurrent platinum-sensitive ovarian cancer: a randomised phase 2 study. *Lancet Oncol.* **15**, 1207–1214 (2014).
146. M.D, A. C. *Phase II Randomized Trial of ABT-888 in Combination With Metronomic Oral Cyclophosphamide in Refractory BRCA-Positive Ovarian, Primary Peritoneal, Ovarian High-Grade Serous Carcinoma, Fallopian Tube Cancer, or Triple-Negative Breast Cancer*. https://clinicaltrials.gov/ct2/show/NCT01306032 (2017).
147. Memorial Sloan Kettering Cancer Center. *A Pilot Study of Nivolumab With or Without Ipilimumab in Combination With Front-Line Neoadjuvant Dose Dense Paclitaxel and Carboplatin Chemotherapy and Post-Surgical Dose Dense Paclitaxel and Carboplatin Chemotherapy in Patients With High Grade Serous Ovarian, Fallopian Tube, or Primary Peritoneal Cancer*. https://clinicaltrials.gov/ct2/show/NCT03245892 (2021).
148. Clovis Oncology, Inc. *ARIEL4 (Assessment of Rucaparib In Ovarian CancEr TriaL): A Phase 3 Multicenter, Randomized Study of Rucaparib Versus Chemotherapy in Patients With Relapsed, BRCA Mutant, High Grade Epithelial Ovarian, Fallopian Tube, or Primary Peritoneal Cancer*. https://clinicaltrials.gov/ct2/show/NCT02855944 (2022).
149. Phase II Study of AZD2281 in Patients With Known BRCA Mutation Status or Recurrent High Grade Ovarian Cancer or Patients With Known BRCA Mutation Status/ Triple Neg Breast Cancer - Tabular View - ClinicalTrials.gov. https://clinicaltrials.gov/ct2/show/record/NCT00679783.
150. Perrone, A. M. *Analysis of Circulating miRNAs as Potential Biomarkers of Diagnosis and Prognosis in High Grade Serous Ovarian Cancer*. https://clinicaltrials.gov/ct2/show/NCT05146505 (2021).
151. Mannel, R. S. *et al.* A randomized phase III trial of IV carboplatin and paclitaxel × 3 courses followed by observation versus weekly maintenance low-dose paclitaxel in patients with early-stage ovarian carcinoma: A Gynecologic Oncology Group Study. *Gynecol. Oncol.* **122**, 89–94 (2011).
152. Krasner, C. N. *et al.* Patient-reported outcomes in relapsed ovarian cancer: results from a randomized Phase III study of trabectedin with pegylated liposomal doxorubicin (PLD) versus PLD alone. *Gynecol. Oncol.* **127**, 161–167 (2012).
153. Gaillard, S. *et al.* Lurbinectedin versus pegylated liposomal doxorubicin or topotecan in patients with platinum-resistant ovarian cancer: A multicenter, randomized, controlled, open-label phase 3 study (CORAIL). *Gynecol. Oncol.* **163**, 237–245 (2021).
154. Lorusso, D. *et al.* Patient-reported outcomes and final overall survival results from the randomized phase 3 PENELOPE trial evaluating pertuzumab in low tumor human

epidermal growth factor receptor 3 (HER3) mRNA-expressing platinum-resistant ovarian cancer. *Int. J. Gynecol. Cancer Off. J. Int. Gynecol. Cancer Soc.* **29**, 1141–1147 (2019).
155. Mirza, M. R. *et al.* Niraparib Maintenance Therapy in Platinum-Sensitive, Recurrent Ovarian Cancer. *N. Engl. J. Med.* **375**, 2154–2164 (2016).
156. Pujade-Lauraine, E. *et al.* Bevacizumab combined with chemotherapy for platinum-resistant recurrent ovarian cancer: The AURELIA open-label randomized phase III trial. *J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol.* **32**, 1302–1308 (2014).
157. Hurteau, J. A. *et al.* Randomized phase III trial of tamoxifen versus thalidomide in women with biochemical-recurrent-only epithelial ovarian, fallopian tube or primary peritoneal carcinoma after a complete response to first-line platinum/taxane chemotherapy with an evaluation of serum vascular endothelial growth factor (VEGF): A Gynecologic Oncology Group Study. *Gynecol. Oncol.* **119**, 444–450 (2010).
158. McNeish, I. A. *et al.* A randomised, placebo-controlled trial of weekly paclitaxel and saracatinib (AZD0530) in platinum-resistant ovarian, fallopian tube or primary peritoneal cancer†. *Ann. Oncol.* **25**, 1988–1995 (2014).
159. du Bois, A. *et al.* Standard first-line chemotherapy with or without nintedanib for advanced ovarian cancer (AGO-OVAR 12): a randomised, double-blind, placebo-controlled phase 3 trial. *Lancet Oncol.* **17**, 78–89 (2016).
160. Moore, K. N. *et al.* Phase III, randomized trial of mirvetuximab soravtansine versus chemotherapy in patients with platinum-resistant ovarian cancer: primary analysis of FORWARD I. *Ann. Oncol.* **32**, 757–765 (2021).
161. Aghajanian, C. *et al.* Final overall survival and safety analysis of OCEANS, a phase 3 trial of chemotherapy with or without bevacizumab in patients with platinum-sensitive recurrent ovarian cancer. *Gynecol. Oncol.* **139**, 10–16 (2015).
162. Friedlander, M. *et al.* Quality of life in patients with advanced epithelial ovarian cancer (EOC) randomized to maintenance pazopanib or placebo after first-line chemotherapy in the AGO-OVAR 16 trial. Measuring what matters—patient-centered end points in trials of maintenance therapy. *Ann. Oncol.* **29**, 737–743 (2018).
163. Walker, J. L. *et al.* Randomized Trial of Intravenous Versus Intraperitoneal Chemotherapy Plus Bevacizumab in Advanced Ovarian Carcinoma: An NRG Oncology/Gynecologic Oncology Group Study. *J. Clin. Oncol.* **37**, 1380–1390 (2019).
164. Monk, B. J. *et al.* A phase 3 randomized, open-label, multicenter trial for safety and efficacy of combined trabectedin and pegylated liposomal doxorubicin therapy for recurrent ovarian cancer. *Gynecol. Oncol.* **156**, 535–544 (2020).
165. Monk, B. J. *et al.* Patient reported outcomes of a randomized, placebo-controlled trial of bevacizumab in the front-line treatment of ovarian cancer: a Gynecologic Oncology Group Study. *Gynecol. Oncol.* **128**, 573–578 (2013).
166. Cojocaru, E., Parkinson, C. A. & Brenton, J. D. Personalising Treatment for High-Grade Serous Ovarian Carcinoma. *Clin. Oncol.* **30**, 515–524 (2018).