TESIS DOCTORAL

UNIVERSIDAD DE GRANADA

FACULTAD DE FARMACIA

MARCADORES GENÉTICOS IMPLICADOS EN LA RESPUESTA Y TOXICIDAD DE LOS FÁRMACOS UTILIZADOS EN PACIENTES CON CÁNCER DE PULMÓN NO MICROCÍTICO

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Tesis presentada por Cristina Pérez Ramírez para optar al grado de Doctora.

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A mi compañero de viaje,

A mi hermano,

A mi madre,

A mi estrella del cielo.

Nada te Turbe;
Nada te espante;
Todo se pasa;
Dios no se muda;
La paciencia
Todo lo alcanza.
Quien a Dios tiene,
Nada le falta.
Sólo Dios basta.

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"Aunque estés lejos de mí, siempre estas en mi mente,

Nunca serás mi pasado y siempre serás mi presente"

RESUMEN

TITULO

Marcadores genéticos implicados en la respuesta y toxicidad de los fármacos utilizados en pacientes con cáncer de pulmón no microcítico.

INTRODUCCIÓN

Actualmente el cáncer de pulmón es un serio problema sanitario ya que es la principal causa de muerte relacionada con cáncer en el mundo. Es el tumor más frecuentemente diagnosticado, después del cáncer de próstata y el de mama. La incidencia es aproximadamente del 14% en ambos géneros., estimándose 117.920 y 106.470 nuevos casos en hombres y mujeres, respectivamente. Es un tumor cuya letalidad es muy elevada, siendo responsable del 27% de todas las muertes por cáncer. Según las últimas estadísticas de cáncer, se espera que más de 224.390 nuevos casos y 158.080 muertes se produzcan en los Estados Unidos en 2016.

Existen dos tipos principales de cáncer de pulmón: El carcinoma de células pequeñas o microcícito, (≈20%; SCLC, por sus siglas en inglés Small Cell Lung Cancer) y carcinoma no microcítico (≈80%; NSCLC, por sus siglas en inglés Non-Small Cell Lung Cancer). El NSCLC presenta diferentes subtipos: carcinoma escamoso/epidermoide, adenocarcinoma y carcinoma de células grandes.

El tratamiento estándar para el NSCLC en estadio temprano es la cirugía. Antes de someterse a cirugía, los pacientes suelen ser tratados con quimioterapia basada en platino para reducir el tamaño o la extensión del cáncer, lo que facilita el procedimiento y el éxito de la cirugía (quimioterapia neoadyuvante). Después de la cirugía, los pacientes con alto riesgo de recurrencia son tratados con quimioterapia basada en platino para maximizar la efectividad de la cirugía (quimioterapia adyuvante). La quimioterapia basada en platino sigue siendo el tratamiento de elección para el NSCLC en estadio avanzado. Este tratamiento se administra en pacientes sin mutaciones en EGFR (receptor del factor de crecimiento epidérmico) o translocaciones en ALK (cinasa del linfoma anaplásico) y en segunda línea en pacientes con EGFR mutado o ALK translocado. Los agentes anti-microtúbulos (taxanos y alcaloides de la vinca), agentes antifolato (pemetrexed), o antagonistas de pirimidinas (gemcitabina) se dan generalmente en combinación con cisplatino o carboplatino. Aunque la quimioterapia basada en platino mejora la supervivencia en comparación con el mejor tratamiento de soporte, la tasa de respuesta global es de aproximadamente 13 a 47,2%. En estadio precoz, la supervivencia libre de progresión y supervivencia global son alrededor de 46-75 meses y 21-48 meses,

respectivamente. Además, más del 75% de los pacientes están vivos cinco años después del diagnóstico. Sin embargo, este porcentaje se reduce bruscamente en estadio avanzado, siendo la tasa de supervivencia a cinco años del 16%. La mediana de la supervivencia libre de progresión y la supervivencia global también son pobres, con 4.3-7.9 meses y 10.4-24.3 meses respectivamente. Además, es un tratamiento muy agresivo con efectos secundarios graves de acuerdo con los criterios comunes de terminología para los eventos adversos (CTCAE) Versión 4, tales como astenia (44,0%), la toxicidad gastrointestinal (33,3%), la toxicidad hematológica (67,1%), neurotoxicidad (69,9%) y la nefrotoxicidad (20-30%).

Factores genéticos como polimorfismos de un solo nucleótido (SNPs) han demostrado estar asociados con diferencias interindividuales en la respuesta y la supervivencia en pacientes con NSCLC. Del mismo modo, el perfil de toxicidad varía de persona a persona y varios estudios han descrito que estas diferencias entre individuos pueden deberse a factores genéticos. Los antineoplásicos de platino, especialmente cisplatino y carboplatino, son complejos de metales pesados que ejercen sus efectos antiproliferativos mediante la inducción de daño en el ADN. Polimorfismos en genes implicados en la reparación del ADN y otros, tales como las vías PI3K/PTEN/AKT y TGF-β han demostrado estar asociados con la respuesta, la supervivencia y la toxicidad en pacientes con NSCLC tratados con quimioterapia basada en platino. Otros procesos celulares, como la metilación del ADN y la proliferación han mostrado tener influencia sobre con el resultado clínico de los regímenes de quimioterapia basada en platinos a través del metabolismo del folato y la señalización de citoquinas.

HIPÓTESIS

"Ciertos marcadores genéticos pueden predecir la respuesta y toxicidad del tratamiento con quimioterapia basada en platino en pacientes diagnosticados con cáncer de pulmón no microcítico".

OBJETIVOS

Objetivo principal

Evaluar la influencia de los marcadores genéticos como marcadores pronósticos y predictivos en pacientes diagnosticados con NSCLC y tratados con quimioterapia basada en platino.

Objetivos específicos

- Determinar la presencia de polimorfismos en genes implicados en la respuesta y la toxicidad en pacientes con NSCLC tratados con quimioterapia basada en platino.
- Evaluar la respuesta al tratamiento con quimioterapia basada en platino en pacientes diagnosticados con NSCLC.
- Evaluar la supervivencia global y la supervivencia libre de progresión en pacientes diagnosticados con NSCLC tratados con quimioterapia basada en platino.
- Evaluar el perfil de toxicidad en los pacientes diagnosticados de NSCLC tratados con quimioterapia basada en platino.
- Evaluar la influencia de marcadores genéticos en la eficacia del tratamiento.
- Evaluar la influencia de los marcadores genéticos de la toxicidad del tratamiento.

MATERIAL Y MÉTODOS

Se realizó un estudio ambispectivo de cohortes, incluyendo 141 pacientes con NSCLC. Se analizaron los siguientes polimorfismos: ERCC1 C118T (rs11615), ERCC1 C8092A (rs3212986), ERCC2 Lys751Gln (rs13181), ERCC2 Asp312Asn (rs1799793), ERCC2 (rs50872), ERCC2 (rs238416), ERCC5 His46His (rs1047768), ERCC5 Asp1104His (rs17655), XRCC1 Arg194Trp (rs1799782), XRCC1 Gln399Arg (rs25487), MDM2 (rs1470383), MDM2 (rs1690924), ABCB1 C3435T (rs1045642), ABCB1 C1236T (rs1128503), ABCB1 Ala893Ser/Thr (rs2032582), MTHFR A1298C (rs1801131), MTHFR C677T (rs1801133), MTR (rs1805087), SLC19A1 Arg27His (rs1051266), IL1B (rs12621220), IL1B (rs1143623), IL1B (rs16944), IL1B (rs1143627), IL6 (rs1800795), IL16 (rs7170924). Se evaluó el efecto de estos polimorfismos sobre la respuesta, supervivencia y toxicidad de la quimioterapia basada en platinos.

Los polimorfismos se analizaron mediante PCR en tiempo real con sondas TaqMan, secuenciación y reacción en cadena de la polimerasa con análisis del polimorfismo de los fragmentos de restricción (PCR-RFLP).

RESULTADOS

Los pacientes con el genotipo GG para ERCC1 C8092A (p=0.0268, OR=2.50; $IC_{95\%}$ =1.12-5.69) y el genotipo GG para XRCC1 Gln399Arg-GG (p=0.0161; OR=2.99; $IC_{95\%}$=1.26-7.62) mostraron una

significativa mayor tasa de respuesta global a la quimioterapia basada en platinos (Tabla 1). El análisis de supervivencia de Cox reveló que los pacientes que portaban el genotipo GG para MDM2 rs1690924 (p=0.0345; HR=1.99; $IC_{95\%}$=1.05-3.80) presentaron mayor riesgo de muerte (Tabla 2). Por otra parte, los portadores del alelo A para IL1B rs16944 (p=0.0141; HR=1.68; $CI_{95\%}$=1.11-2.54), el alelo A para MTR rs1805087 (p=0.0016; HR=12.19; $CI_{95\%}$=2.57-57.71), el genotipo AA para SLC19A1 Arg27His (p=0.0116; HR=1.75; $CI_{95\%}$=1.13-2.70) mostraron mayor riesgo de progresión (Tabla 3). No se encontró influencia de los polimorfismos ERCC1 C118T, ERCC2 Lys751Gln, ERCC2 Asp312Asn, ERCC2 rs50872, ERCC2 rs238416, ERCC5 Asp1104His, ERCC5 His46His, XRCC1 Arg194Trp, MDM2 rs1470383, ABCB1 C3435T, ABCB1 C1236T, ABCB1 Ale893Ser/Ther, MTHFR A1298C, MTHFR C677T, IL1B rs12621220, IL1B rs1143623, IL1B rs1143627, IL6 rs1800795, IL16 rs7170924 sobre los resultados clínicos de la quimioterapia basada en platinos

Tabla 1. Influencia de las características clínicas y polimorfismos genéticos sobre la respuesta.

	Respuesta	
	OR ($CI_{95\%}$)	**p-value**
Cirugía (Sí)	25.38 (4.94-467.06)	0.0021
ERCC1 C8092A-GG	2.50 (1.12-5.69)	0.0268
XRCC1 Gln399Arg-GG	2.99 (1.26-7.62)	0.0161

OR: odds ratio; $CI_{95\%}$: 95% intervalo de confianza.

Tabla 2. Influencia de las características clínicas y polimorfismos genéticos sobre la supervivencia global.

	Supervivencia Global	
	HR ($CI_{95\%}$)	**p-value**
Género (Varón)	3.21 (1.72-6.00)	0.0003
Histología (Adenocarcinoma)	2.21 (1.17-4.15)	0.0142
Agentes de quimioterapia (Gemcitabina and Pemetrexed)	2.35 (1.36-4.08)	0.0023
Cirugía (No)	5.93 (2.79-12.59)	<0.001
MDM2 rs1690924-GG	1.99 (1.05-3.80)	0.0345

HR: hazard ratio; $CI_{95\%}$: 95% intervalo de confianza.

Table 3. Influencia de las características clínicas y polimorfismos genéticos sobre la supervivencia libre de enfermedad.

	Supervivencia libre de enfermedad	
	HR ($CI_{95\%}$)	**p-value**
Cirugía (No)	11.99 (5.83-24.66)	<0.001
Radioterapia (No)	3.06 (1.88-4.97)	<0.001
IL1B rs16944- A alelo	1.68 (1.11-2.54)	0.0141
MTR rs1805087-A alelo	12.19 (2.57-57.71)	0.0016
SLC19A1 Arg27His-AA	1.75 (1.13-2.70)	0.0116

HR: hazard ratio; $CI_{95\%}$: 95% intervalo de confianza.

Los pacientes con el alelo T para ERCC1 C118T (p=0.00345; RR=26.05; 95% CI=4.33-515.77) y el genotipo CC para ERCC2 rs50872 (p=0.00291; RR=4.06; 95% CI=1.66-10.65) tenían un riesgo más alto de presentar toxicidad general a la quimioterapia basada en platino (Tabla 4). El alelo G de ERCC2 Asp312Asn, el genotipo TT de ABCB1 C1236T y los genotipos CT/TT de IL1B rs12621220 confieren un mayor riesgo de presentar múltiples eventos adversos (Tabla 5). El análisis de subtipos de toxicidad también reveló que el genotipo CC para ERCC2 rs5087 (p=0.01562; OR=3.23; 95% CI=1.29-8.82) y el alelo T para IL16 rs7170924 (p=0.01007; OR=3.19; 95% CI=1.35-7.97) se asociaron con toxicidad hematológica grado 3-4 (Tabla 6). No se encontró asociación de los polimorfismos ERCC1 C8092A, ERCC2 Lys751Gln, ERCC2 Asp312Asn, ERCC5 Asp1104His, XRCC1 Arg194Trp, MDM2 rs1690924, ABCB1 C3435T, ABCB1 Ala893Ser/Thr, MTHFR A1298C, MTHFR C677T, IL1B rs1143623, IL1B rs16944, y rs1143627 IL1B con la toxicidad de la quimioterapia basada en platinos.

Table 4 Influencia de las características clínicas y polimorfismos genéticos sobre la toxicidad general.

	General Toxicity	
	OR ($CI_{95\%}$)	**p-value**
Agentes de quimioterapia		
Gemcitabine (Referencia)		
Paclitaxel	0.13 (0.02-0.52)	0.00587
Pemetrexed	0.10 (0.02-0.38)	0.00147
Vinorelbina	0.27 (0.07-0.91)	0.04098
Historia personal de cáncer (No)	11.57 (2.09-217.62)	0.02245
ERCC1 C118T-CT/TT	26.05 (4.33-515.77)	0.00345
ERCC2 rs50872-CC	4.06 (1.66-10.65)	0.00291

OR: odds ratio; $CI_{95\%}$: 95% intervalo de confianza.

Table 5. Influencia de los polimorfismos genéticos sobre el numero de reacciones adversas.

	Número de reacciones adversas	
	OR ($CI_{95\%}$)	**p-value**
	>1 reacción adversa	
ERCC2 Asp312Asn		
AA	0.23 (0.07-0.71)	0.01140
(Referencia) AG		
GG	0.31 (0.13-0.73)	0.00891
IL1B rs12621220-CT/TT	3.37 (1.55-7.69)	0.00280
	>2 reacciones adversas	
ERCC2 Asp312Asn-AG/GG	5.92 (1.52-40.18)	0.026040
ABCB1 C1236T-TT	4.35 (1.67-11.76)	0.002842

OR: odds ratio; $CI_{95\%}$: 95% intervalo de confianza.

Table 6. Influencia de las características clínicas y polimorfismos genéticos sobre la toxicidad hematológica.

	Toxicidad Hematológica	
	OR ($CI_{95\%}$)	**p-value**
Agentes de quimioterapia		
Gemcitabine (Referencia)		
Paclitaxel	0.15 (0.03, 0.54)	0.00559
Pemetrexed	0.11 (0.02, 0.42)	0.00218
Vinorelbine	0.36 (0.11, 1.10)	0.07525
ERCC2 rs50872-CC	3.23 (1.29, 8.82)	0.01562
IL16 rs7170924- GT/TT	3.19 (1.36, 7.97)	0.01007

OR: odds ratio; $CI_{95\%}$: 95% intervalo de confianza.

CONCLUSIONES

I. Los genotipos GG para ERCC1 C8092A y XRCC1 Gln399Arg se asociaron con una mejor tasa de respuesta global en pacientes con NSCLC.

II. Pacientes con NSCLC que portan el genotipo GG para MDM2 rs1690924 estaban en mayor riesgo de muerte.

III. Los alelos A para IL1B rs1694 y MTR rs1805087 y el genotipo AA rs1051266 SLC19A1 se asociaron con mayor riesgo de progresión en el NSCLC.

IV. No se encontró asociación en nuestros pacientes entre ERCC1 C118T, ERCC2 Lys751Gln, ERCC2 Asp312Asn, ERCC2 rs50872, ERCC2 rs238416, ERCC5 Asp1104His, ERCC5 His46His, XRCC1 Arg194Trp, MDM2 rs1470383, ABCB1 C3435T, ABCB1 C1236T, ABCB1 Ale893Ser/Ther, MTHFR A1298C, MTHFR C677T, IL1B rs12621220, IL1B rs1143623, IL1B

rs1143627, IL6 rs1800795, IL16 rs7170924 y los resultados clínicos de la quimioterapia basada en platino.

V. Los pacientes con el alelo T para ERCC1 C118T y el genotipo CC ERCC2 rs50872 tenían mayor riesgo de toxicidad general para la quimioterapia basada en platinos.

VI. El alelo G para ERCC2 Asp312Asn, el genotipo GG para ABCB1 C1236T y los genotipos CT/TT para IL1B rs12621220 confirieron un mayor riesgo de presentar múltiples eventos adversos en el CPNM.

VII. El genotipo CC para ERCC2 rs50872 y el alelo T para IL16-rs7170924 se asociaron con toxicidad hematológica grado 3-4 en el NSCLC.

VIII. No se encontró asociación en nuestros pacientes entre ERCC1 C8092A, ERCC2 Lys751Gln, ERCC5 Asp1104His, XRCC1 Arg194Trp, rs1690924 MDM2, ABCB1 C3435T, ABCB1 Ala893Ser / Thr, A1298C MTHFR, MTHFR C677T, rs1143623 IL1B, rs16944 IL1B, y rs1143627 IL1B y la toxicidad de la quimioterapia basada en platinos.

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ABBREVIATION LIST

ABCB1: ATP-binding cassette, sub-family B (MDR/TAP), member 1

AJCC: American Joint Committee on Cancer

AKT: v-akt murine thymoma viral oncogene

ALK: Anaplastic lymphoma kinase

APE1: Apurinic/apyimidinic endonuclease 1

BER: Base Excision Repair

BP: Physical position (base-pair)

BRCA1: Breast cancer 1

BRCA2: Breast cancer 2

CBDCA: Carboplatin

CDDP: Cisplatin

CHR: Chromosome

CHUG: Complejo Hospitalario Universitario de Granada

$CI_{95\%}$: 95% Confidence Interval

CT: Computed Tomography

CTCAE: Common Terminology Criteria for Adverse Events

CR: Complete Response

DSB: Double-Strand Break repair

DTX: Docetaxel

EBUS-TBNA: Endobronchial Ultrasound-guided Transbronchial Needle Aspiration

EGFR: Epidermal Growth Factor Receptor

ERCC1: Excision Repair Cross-Complementing group 1

ERCC2: Excision Repair Cross-Complementation group 2

ERCC4: Excision Repair Cross-Complementation group 4

ERCC5: Excision Repair Cross-Complementation group 5

EUS-FNA: Endoscopic Ultrasound-guided Fine Needle Aspiration

FEV1: Forced Expiratory Volume in One Second

GMZ: Gemcitabine

GSTPs: Glutathione S-transferase

GSTP1: Glutathione S-transferase Pi 1

HR: Hazard Ratio

ILs: Interleukins

MDM2: MDM2 proto-oncogene, E3 ubiquitin protein ligase

MMR: Mismatch Repair

MRE11: MRE11 homolog A, double strand break repair nuclease

MST: Median Survival Time (months)

MTHFR: Methylenetetrahydrofolate reductase

MTR: Methionine synthase

NBN: Nibrin

NER: Nucleotide Excision Repair

NR: Not Reached

NSCLC: Non-Small Cell Lung Cancer

PFS: Progression-Free Survival

OR: Odds Ratio

ORR: Overall Response Rate

OS: Overall Survival

PARP1: Poly (ADP ribose) polymerase 1

PARP2: Poly (ADP ribose) polymerase 2

PCR: Polymerase Chain Reaction

PCR-RFLP: PCR plus Restriction Fragment Length Polymorphism

PD: Progressive Disease

PET: Positron Emission Tomography

PI3K: Phosphatidylinositol 3-kinase

PMX: Pemetrexed

PR: Partial Response

PTEN: Phosphatase and tensin homolog

PTX: Paclitaxel

RAD50: RAD50 double strand break repair protein

RAD51: RAD51 recombinase

RECIST: Response Evaluation Criteria in Solid Tumors

RR: Relative Risk

SCLC: Small Cell Lung Cancer

SD: Stable Disease

SLC19A1: Solute carrier family 19 (folate transporter), members 1

SNPs: Single Nucleotide Polymorphisms

TGF-β: Transforming Growth Factor beta

TNM: Tumor-Node-Metastasis

VNR: Vinorelbine

VP16: Etopósido.

XRCC1: X-ray repair complementing defective repair in Chinese hamster cells 1

XRCC3: X-ray repair complementing defective repair in Chinese hamster cells 3

INTRODUCTION

1. INTRODUCTION

1.1 Lung cancer epidemiology

Lung cancer is the most common diagnosed type of cancer, being the second tumor in incidence (after prostate in men and breast cancer in woman). Estimated cases in 2016 are 117.920 in men and 106.470 in women, with an approximate incidence of 14% [1]. This tumor is the highest mortal cancer among both genders, being responsible for 27% of all cancer deaths [1]. According to the latest cancer statistics, over 224.390 new cases and 158.080 deaths are expected to occur in the United States in 2016.

1.2 Risk factors

Advanced age is the main risk factor for the majority of cancers. Other risk factors for lung cancer include:

- Smoking: smokers have 10 times greater risk of developing lung cancer than non-smokers, and this probability increases with the quantity of cigarettes, duration of smoking and starting age [2]. Although quit smoking, former smokers continue to have an elevated risk for lung cancer for years [2]. A cause of lung cancer in non-smokers is passive smoking. Several studies have reported a significantly increased risk among those exposed to passive smoke [3].

- Occupational exposure to asbestos, arsenic, chromium, beryllium, nickel [4].

- Living in an area with air pollution [5-7].

- Exposure to radon gas: radon is a radioactive gas produced by the natural breakdown of uranium in soil, rock and water. This gas is in atmospheric air at low concentration and may accumulate in any building, including homes [8]. However, exposure to domestic radon depends how houses are constructed and ventilated. Moreover, as an occupational risk factor, is especially important for miners because they are usually exposed to high concentrations of this gas.

- Alcohol: alcohol consumption of at least 30 g/day increases the risk lung cancer [9].

- Diet: fruits and vegetables provide a great quantity of vitamins and others micronutrients such as carotenoids. Several studies have reported the benefit of these

substances because they decrease the risk of cancer, but excessive consumption may be harmful [10,11].

- Family history of cancer: People with a parent, sibling or child with lung cancer have an increased risk of the disease [12].

- Other diseases: patients with idiopathic pulmonary fibrosis have a risk of lung cancer 14 times greater. Similarly, in patients with chronic obstructive pulmonary disease, the risk of lung cancer increase 4 times [13,14].

1.3 Sign and symptoms

Lung cancer may present with symptoms or be found incidentally on chest imaging. Symptoms and signs may result from the location of the primary local invasion or compression of adjacent thoracic structures, distant metastases, or paraneoplastic phenomena [15]. Non-specific symptoms may include loss of appetite, weight loss and fatigue, whereas the specific symptoms are the following:

- Respiratory symptoms: coughing, coughing up blood, wheezing, or shortness of breath.

- Systemic symptoms: weight loss, weakness, fever, or clubbing of the fingernails.

- Symptoms due to the cancer mass pressing on adjacent structures: chest pain, bone pain, superior vena cava obstruction, or difficulty swallowing.

At the time of diagnostic, the majority of the patients have metastatic disease and the symptoms include neurological defect or personality change from brain metastases or pain from bone metastases [15]. The most common sites of metastases for lung cancer are the other lung, adrenal glands, bones, brain, liver, pericardium and kidneys [15].

1.4 Diagnosis

The procedures used to determine the presence of cancer include the following [15]:

- **Clinical examination:** although the diagnosis of lung cancer may not be based on the findings of respiratory clinical examination, this examination should always be part of the diagnostic studies of a patient if respiratory symptoms are reported or abnormal findings are detected in radiological tests. Respiratory clinical examination includes inspection, palpation, percussion and chest auscultation. Lung auscultation should

carefully be interpreted, put in context with medical history and other clinical findings of the patient. Clinical examination should include physical palpation of the superficial neck lymph nodes groups, as well as those located above the collarbone (supraclavicular).

- **Imaging test:** Radiological tests are crucial to confirm a diagnosis of lung cancer and to define its extension. They include:

  - Chest X-ray: usually taken as a first test for diagnostic studies of a patient.

  - Computed tomography (CT) of the chest and upper abdomen: a radiological test that is necessary for the correct staging of NSCLC. This technique allows an accurate assessment of the extent of the tumor.

  - Brain computed tomography: It is necessary to exclude the presence of metastases in the brain.

  - Positron emission tomography (PET): A test nuclear medicine imaging which it allows examine the morphology and metabolic activity of the tumor. It is recommended

  - Bone scan: is an imaging test of nuclear medicine that is performed to check if lung cancer has metastasized to the bones.

- **Histopathological examination:** This involves laboratory analysis of the cells obtained from a tumor tissue sample (biopsy). The most common techniques used to obtain tumor biopsies are the following:

  - Bronchoscopy: A technique used to visualize the inside of the airway with an instrument inserted through the mouth or nose. It allows examining the patient's airway to check for anomalies.

  - Pulmonary needle biopsy guided by computed tomography: this technique is used when considered bronchoscopy is not useful to obtain a biopsy. A needle is inserted into the chest using computed tomography guidance.

  - Endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA): it is a technique that allows confirmation of the invasion of the lymph regional nodes. An ultrasound tube is used to identify any suspicious node that may be close to the

airways. The tube also contains a very fine needle. This needle is used to take samples of body tissue by pushing through the bronchus to the tissue on the other side.

- Endoscopic ultrasound-guided fine needle aspiration (EBUS-FNA): it is similar to the EBUS; this technique is useful to know the invasion of lymph regional nodes. Unlike EBUS, the instrument is inserted by the esophagus.

- Mediastinoscopy: it is an invasive procedure that allows visualization of the mediastinum with a laparoscope inserted through an incision of approximately 1 cm above the union of the sternum to the clavicle.

- Biopsy from metastatic lesions: if the disease has spread to other parts of the body, a biopsy from metastatic lesion may be obtained.

- **Cytological examination**: unlike histopathological examination, which is performed in a tumor tissue sample, cytological examination is the laboratory examination of the cancer cells spontaneously separate from the tumor. The main methods for cytological exploration are below:

  - Bronchoscopy: during bronchoscopy is usually performed bronchial washings and collection of secretions for checking the presence of cancer cells.

  - Thoracentesis/pleural drainage: These techniques allow the aspiration of liquids from the pleural cavity in case of pleural effusion.

  - Pericardiocentesis/pericardial drainage: These techniques allow aspiration fluid from the pericardial cavity in case of pericardial effusion.

1.5 Classification of lung cancer

The most important types of lung cancer are small cell lung cancer (SCLC) and non-small cell lung cancer (NSCLC). NSCLC accounts for approximately 80-85% of all lung cancer cases and is divided into different subtypes: squamous cell carcinoma, adenocarcinoma and large cell carcinoma. In accordance with the American Joint Committee on Cancer (AJCC), the majority of the patients are catalogued as advanced stage (IIIB-IV) at the time of diagnosis [16-18].

1.6 Non-small cell lung cancer staging

The international classification or international staging system used in NSCLC is developed and maintained by the AJCC, being the latest version the 7th edition, that was published in 2009. The descriptors of the internationally used TNM (Tumor-Node-Metastasis) classification include the size of and the degree of loco regional invasion by the primary tumor (T), the extent of regional lymph node involvement (N), and the presence or absence of intrathoracic or distant metastases (M) [18]. The goal of such a classification system is to assist clinicians in planning treatment, determining prognosis, evaluating treatment results, and facilitating information exchange between multiple centers. The general outline for the TNM classification is below and Table 1 shows the TNM staging groups.

Primary tumor (T):

TX: Primary tumor cannot be assessed, or the tumor is proven by the presence of malignant cells in sputum or bronchial washing but is not visualized by imaging or bronchoscopy.

T0: No evidence of primary tumor.

Tis: Carcinoma in situ

T1: Tumor ≤ 3 cm in greatest dimension, surrounded by lung or visceral pleura, no bronchoscopic evidence of invasion more proximal than the lobar bronchus (not in the main bronchus); superficial spreading of tumor in the central airways (confined to the bronchial wall)

T1a: Tumor ≤ 2 cm in the greatest dimension.

T1b: Tumor > 2 cm but ≤ 3 cm in the greatest dimension.

T2: Tumor > 3 cm but ≤ 7 cm or tumor with any of the following:

- Invades visceral pleura.
- Involves the main bronchus ≥ 2 cm distal to the carina.
- Associated with atelectasis/obstructive pneumonitis extending to hilar region but not involving the entire lung.

T2a: Tumor > 3 cm but ≤ 5 cm in the greatest dimension.

T2b: Tumor > 5 cm but ≤ 7 cm in the greatest dimension.

T3: Tumor more than 7 cm or one that directly invades any of the following: parietal pleural (PL3), chest wall (including superior sulcus tumors), diaphragm, phrenic nerve, mediastinal pleura, parietal pericardium; or tumor in the main bronchus less than 2 cm distal to the carina1 but without involvement of the carina; or associated atelectasis or obstructive pneumonitis of the entire lung or separate tumor nodule(s) in the same lobe.

T4: Tumor of any size that invades any of the following: mediastinum, heart, great vessels, trachea, recurrent laryngeal nerve, esophagus, vertebral body, carina, separate tumor nodule(s) in a different ipsilateral lobe.

Regional Nodes (N)

NX: Regional lymph nodes cannot be assessed.

N0: No regional node metastasis.

N1: Metastasis in ipsilateral peribronchial and/or ipsilateral hilar lymph nodes and intrapulmonary nodes, including involvement by direct extension.

N2: Metastasis in the ipsilateral mediastinal and/or subcarinal lymph node(s).

N3: Metastasis in the contralateral mediastinal, contralateral hilar, ipsilateral or contralateral scalene, or supraclavicular lymph nodes.

Distant metastasis (M)

MX: Distant metastasis cannot be assessed.

M0: No distant metastasis.

M1: Distant metastasis.

M1a: Separate tumor nodule(s) in a contralateral lobe; tumor with pleural nodules or malignant pleural (or pericardial) effusion.

M1b: Distant metastasis.

Table 1. TNM stage system

Anatomic stage/prognostic groups			
Occult carcinoma	TX	N0	M0
Stage 0	Tis	N0	M0
Stage IA	T1a	N0	M0
	T1b	N0	M0
Stage IB	T2a	N0	M0
Stage IIA	T2b	N0	M0
	T1a	N1	M0
	T1b	N1	M0
	T2a	N1	M0
Stage IIB	T2a	N1	M0
	T3	N0	M0
Stage IIIA	T1a	N2	M0
	T1b	N2	M0
	T2a	N2	M0
	T2b	N2	M0
	T3	N1	M0
	T3	N2	M0
	T4	N0	M0
	T4	N1	M0
Stage IIIB	T1a	N3	M0
	T1b	N3	M0
	T2a	N3	M0
	T2b	N3	M0
	T3	N3	M0
	T4	N2	M0
	T4	N3	M0
Stage IV	Any T	Any T	M1a
	Any T	Any T	M1b

1.7 Non-small cell lung cancer treatment

Generally, the treatment of NSCLC includes several options: surgery, chemotherapy, radiotherapy and targeted therapy. The selection of the therapy will depend on stage of the disease and certain characteristics of patients, such as age or general condition. The treatment is not only focus on lung primary tumor; it is also directed to metastases that may appear in other part of the body.

1.7.1 Resectable tumors

Based on NCCN version 4.2016 guideline, the standard treatment for resectable early-stage (I-IIIA) NSCLC is surgery (Figure 1). In this point, it is essential to determine which resection method is recommended in each case, determining the individual risks of each patient. If the expiratory volume in one second (FEV1, Forced Expiratory Volume in One Second) is higher than 80% and no evidence of dyspnea, the patient is a candidate for resection including pneumonectomy.

Before using radical treatment intervention, patients with locally advanced disease are usually treated with platinum-based chemotherapy to reduce the size or extent of the cancer, facilitating the procedure and success of the surgery (neoadjuvant chemotherapy). After surgery, patients with high risk of recurrence are treated with platinum-based chemotherapy to maximize surgery effectiveness (adjuvant chemotherapy). Patients with unresectable early-stage NSCLC are candidates for platinum-based chemotherapy and/or radiotherapy [19].

1.7.2 Unresectable tumors

Platinum-based chemotherapy remains the treatment of choice for advanced-stage (IIIB-IV) NSCLC (Figure 1) [19]. At this stage the treatment is palliative, not curative; it is used to alleviate symptoms, improving the quality of life and survival of patients. Cisplatin and carboplatin are the main platinum drugs used in NSCLC. These compounds interfere with DNA, generating adducts that induce cells to die in a programmed way. They are usually given in combination with a third generation drugs, which are the following [19]:

- Taxanes (paclitaxel/docetaxel) and vinca alkaloids (vinorelbine): the principal mechanism of action of these drugs is the disruption of microtubule function [20]. Microtubules are essential to cell division, so inhibition of these structures result in cell death [20].

- Pemetrexed: this drug is chemically similar to folic acid and is in the class of chemotherapy drugs called antifolate agents. This compound acts inhibiting the formation of precursor purine and pyrimidine nucleotides [21]. Thus, pemetrexed prevents the formation of DNA and RNA, which are required for the growth and survival of both normal cells and cancer cells [21].

- Gemcitabine: it is a pyrimidine analogous whose fundamental mechanism of action is the inhibition of DNA synthesis [22].

No significant differences have been found in any of the combination [23,24]. In clinical practice, carboplatin is an alternatively to cisplatin when there is a contraindication.

Selective patients with advanced NSCLC may benefit from targeted therapy (Figure 1) [19]. Patients with mutations in EGFR (epidermal growth factor receptor) are treated with an EGFR tyrosine kinase inhibitor, such as gefitinib, erlotinib or afatinib. Similarly, patients harboring

translocations in ALK (anaplastic lymphoma kinase) are treated with crizotinib, an ALK inhibitor. At second line, these patients are treated with platinum-based chemotherapy.

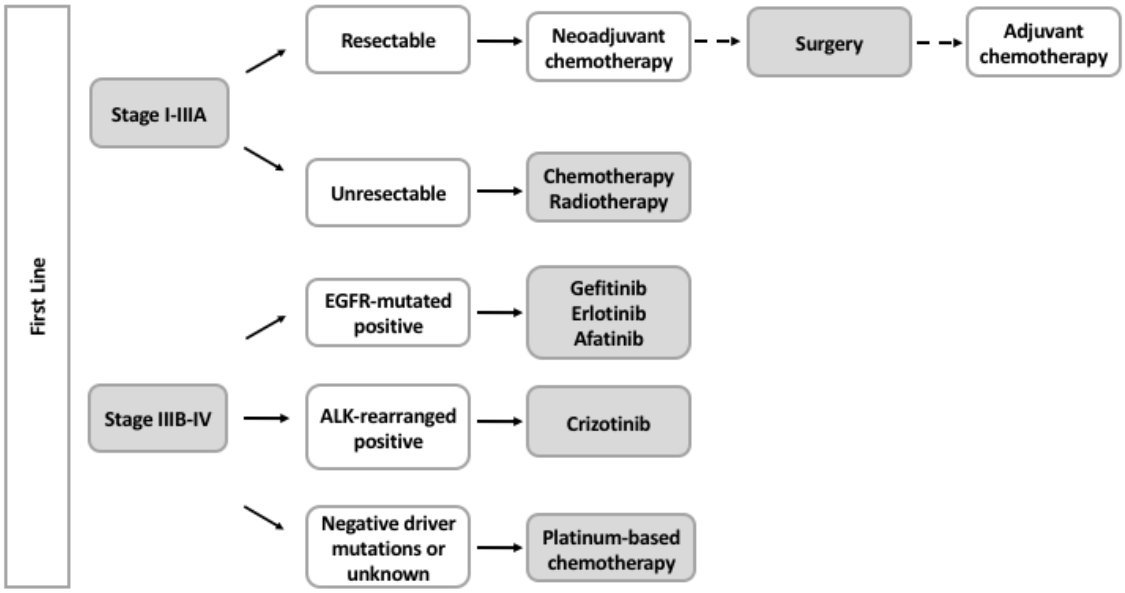

Figure 1. NSCLC treatment algorithm

1.8 Clinical outcomes and toxicity of platinum-based chemotherapy

Although platinum-based chemotherapy improves survival in comparison with best supportive care [25], overall response rate (ORR) is about 13-47.2% [26-42]. In early-stage, median progression-free survival (PFS) and overall survival (OS) are around 46-75 months and 21-48 months, respectively [29]. In addition, over 75% of the patients are alive five years after diagnosis [43]. However, this percentage is reduced abruptly at late-stages and survival five-year survival rate is around 16%. Median PFS and OS are also poor, with 4.3-7.9 months for PFS and 10.4-24.3 months for OS [44]. Furthermore, it is a very aggressive treatment with severe side effects according to the common terminology criteria for adverse events (CTCAE) Version 4, such as asthenia (44.0%), gastrointestinal toxicity (33.3%), hematological toxicity (67.1%), neurotoxicity (69.9%) and nephrotoxicity (20-30%) [45-47].

1.9 Prognostic factors of platinum-based chemotherapy

The knowledge of prognostic factors may improve clinical outcomes of NSCLC therapy, by stratifying patients into subgroups that could be managed differently. The crucial factors for NSCLC prognosis are presence of pulmonary symptoms, large tumor size (>3 cm), non-squamous cell type (histology), degree of spread (stage) and metastases to multiple lymph nodes, and

vascular invasion [48-56]. However, a significantly variability in terms of response and survival has been described among patients with the same clinical characteristics, suggesting that other factors may play a role on NSCLC prognosis [17,57]. Interestingly, genetic factors such as single nucleotide polymorphisms (SNPs) have demonstrated to be associated with inter-individual differences in response and survival in NSCLC patients [58-73]. Similarly, the toxicity profile varies from person to person and various studies have reported that these inter-individual differences may be due to genetic factors, which are involved in platinum pharmacodynamics, metabolism and mechanism of action [65,67-81].

1.10 Pharmacogenetics of platinum-based chemotherapy

Platinum drugs, particularly cisplatin and carboplatin, are heavy metal complexes that exert their antiproliferative effects by inducing DNA damage. They bind covalently to two different sites, either within the same DNA molecule or between two different DNA molecules, generating adducts that inhibit DNA synthesis and transcription and are responsible for severe toxicity of these drugs [82,83]. Mammalian cells have different DNA repair pathways to repair DNA damage, including nucleotide-excision repair (NER), base excision repair (BER) and double-strand break repair (DSB) (Figure 2). Several proteins of these pathways, such as excision repair cross-complementing group 1 (*ERCC1*), excision repair cross-complementation group 2 (*ERCC2*, also known as *XPD*), excision repair cross-complementation group 5 (*ERCC5*), X-ray repair complementing defective repair in Chinese hamster cells 1 (*XRCC1*) and X-ray repair complementing defective repair in Chinese hamster cells 3 (*XRCC3*), are involved in the detection and repair of the adducts and cross-links induced by platinum activity [84,85]. The p53 pathway also plays an essential role in DNA repair, together with cell cycle control and apoptosis initiation in response to DNA damage [86]. The *MDM2* proto-oncogene, E3 ubiquitin protein ligase (MDM2) modulates the activity of the pathway through directing binding, ubiquitination and degradation of p53 [87]. Numerous studies have reported that SNPs in any of these genes may regulate the DNA repair functions in the normal and tumor cells, contributing to individual variation in the response and toxicity to platinum-based chemotherapy [58-65,88,89].

Although DNA repair pathways are the key players in platinum toxicity and response, other pathways and proteins are also involved, including the phosphatidylinositol 3-kinase/ phosphatase and tensin homolog /v-akt murine thymoma viral oncogene (PI3K/PTEN/AKT) pathway, the transforming growth factor beta (TGF-β) and cytokine signaling pathways, drug transporters, detoxification systems and folate metabolism enzymes. Genetic alterations in genes of the PI3K/PTEN/AKT and TGF-β pathways may modify signaling and have an impact in

the development of toxicity or disease progression to platinum-based therapies [90-94]. Polymorphisms in ATP-binding cassette, sub-family B (MDR/TAP), member 1 (*ABCB1*, also called *MDR1*) have also been suggested as predictive markers of response and side effects to platinum therapy [95-97]. *ABCB1* gene is an cell membrane transporter [98], involved in the ATP dependent export of chemicals out of cell [99-101] that modulates response and toxicity by impairing the intracellular retention of multiple anticancer drugs [102,103]. Detoxification of platinum compounds is mediated by glutathione S-transferase Pi 1 (*GSTP1*); gene polymorphisms in this enzyme have been correlated with clinical outcome and toxicity of platinum therapy [75,104-110]. Methylenetetrahydrofolate reductase (*MTHFR*), methionine synthase (*MTR*) and solute carrier family 19 (folate transporter), members 1 (*SLC19A1*) are involved in folate metabolism [111-115]. Genetic alterations in these genes disturb methylation of DNA, which may influence the effectiveness and toxicity of platinum-based chemotherapy [66-68,71]. Cytokine signaling regulates tumor progression by promoting angiogenesis, cell growth and differentiation of tumor cells [116]. Finally, gene polymorphisms in interleukins 1B (*IL1B*), 6 (*IL6*), 12A (*IL12A*), 13 (*IL13*), 16 (*IL16*) have also been associated with survival to platinum-based chemotherapy [77,78].

1.10.1 DNA repair pathways

Genetic alterations in DNA repair genes can modify individual responses and toxic side effects to platinum-based chemotherapy. Four main DNA repair pathways are utilized by the cells: NER, BER, DBS and MMR (mismatch repair). NER, BER, and DBS are the main repair pathways in the removal of DNA lesions produced by platinum compounds [84,85]. MMR is not directly involved in repair of platinum adducts. It recognizes DNA adducts and activates apoptosis [117]. All of them are regulated by p53, which can trigger cell cycle arrest and DNA repair or apoptosis (Figure 2) [118-121].

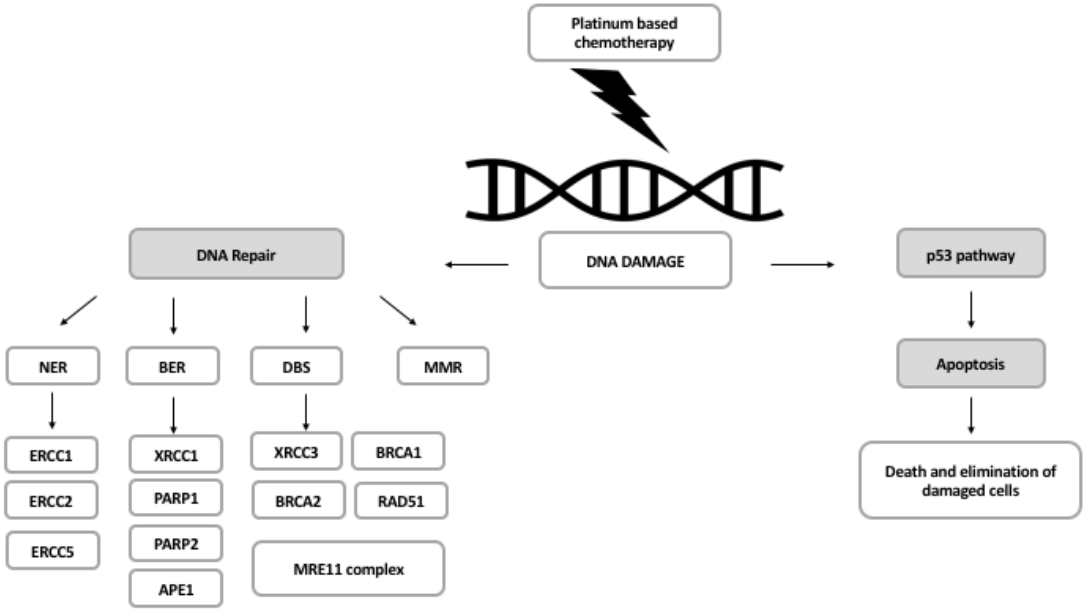

Figure 2. DNA repair pathways involved in removal DNA lesions produced by platinum compounds.

1.10.1.1 NER pathway

The NER pathway, through *ERCC1*, *ERCC2* and *ERCC5* genes, is able to repair helix-distorting DNA lesions, which prevent base pairing, blocking transcription and normal replication [122-124].

1.10.1.1.1 ERCC1

ERCC1 is the key enzyme in the NER pathway [122]. It heterodimerizes with excision repair cross-complementation group 4 (ERCC4 also called XPF), and the resultant ERCC1/ERCC4 complex makes an incision at the 5' end of the lesion, allowing removal of the damaged DNA strand and further polymerization and re-ligation [122,124].

Two polymorphisms in *ERCC1*; rs11615 (C→T synonymous substitution at codon 118, exon 4, Asn→Asn, C118T) and rs3212986 (C→A substitution in the 3'-untranslated region, C8092A); have been extensively investigated in advanced NSCLC patients treated with platinum-based chemotherapy (Table 2). The mechanisms underlying the effects of these base changes in platinum therapy effectiveness and toxicity are still unclear. Several studies have described some effects on *ERCC1* mRNA expression, whereas others have found no association [144-146].

Regarding *ERCC1* C118T, its association with clinical outcome to platinum-based chemotherapy remains unclear, with some studies reporting better ORR in patients carrying the C allele

[110,125-128], and others in patients with the TT genotype (Table 2). Six meta-analyses have evaluated the influence of *ERCC1* C118T polymorphism [58-60,95,156,157] and the two largest (23 studies/3231 patients and 21 studies/1281 patients, respectively), have not found any association between the ERCC1 C118T polymorphism and ORR (odds ratio (OR)=0.94; 95% confidence interval ($CI_{95\%}$)=0.72-1.23; I^2=60%; $P_{heterogeneity}$ <0.01; CT/TT vs CC and OR=0.95; $CI_{95\%}$=0.85-1.06; I^2=66.90%; $P_{heterogeneity}$=0.386; CT/TT vs CC) [60,157]. The same contradictory results have been reported in the case of PFS and OS, with some studies finding an association of the CC genotype with a better outcome and others reaching opposite conclusions (Table 2). Finally, a meta-analysis including 13 studies, found a significant association of ERCC1 C118T polymorphism with a longer OS (hazard ratio (HR)=1.26; $CI_{95\%}$=1.02-1.55; I^2=67%; $P_{heterogeneity}$<0.01; CT/TT vs CC) but not with PFS (HR=1.23; $CI_{95\%}$=0.90-1.69; I^2=70.7%; $P_{heterogeneity}$ <0.01; CT/TT vs CC) [60].

The *ERCC1* C8092A polymorphism has also been associated with response in two studies in 115 and 163 Asian patients, which reported worse ORR to platinum-based chemotherapy in patients carrying the A allele (OR=0.23; $CI_{95\%}$=0.10-0.57 for AC/AA vs CC and OR=0.44; $CI_{95\%}$=0.27-0.74 for A vs C allele, respectively) [128,129]. Studies in Caucasian population have not found significant associations between *ERCC1* C8092A polymorphism and outcome in advanced NSCLC patients (Table 2) and four meta-analyses have reached the same conclusion [58,60,95,157]; the largest one, a meta-analysis carried out with 10 studies and 1.311 patients also failed to show influence of this polymorphism on response to platinum-based chemotherapy (OR=1.05; $CI_{95\%}$=0.83-1.32; I^2=39.3%; $P_{heterogeneity}$=0.096; AC/AA vs CC) [60]. Regarding OS and PFS, results for *ERCC1* C8092A are again contradictory, with some reports associating the CC genotype with better and worse OS and PFS (Table 2). A recent meta-analysis, including 6 studies and 999 cases, could not find any significant association between the polymorphism and OS (HR=1.26; $CI_{95\%}$=0.81-1.95; I^2=87%; $P_{heterogeneity}$ =<0.01; AC/AA vs CC) [60].

Finally, although the association between toxicity and *ERCC1* polymorphisms has also been extensively investigated, no significant findings have been reported (Table 2).

Table 2. Influence of ERCC1 polymorphisms on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs11615														
2004	Asian (Korea)	109	IIIB-IV	CDDP DTX/GMZ/PTX	0.61 (0.28-1.31)	CC	3.24 (1.82-5.76)	CC						[137]
2004	Caucasian (Spain)	62	IIIB-IV	CDDP DTX	0.47 (0.13-1.75)	CC	3.54 (1.25-10.04)	CC	2.41 (1.15-5-04)	CC	Grade 2-4 (p>0.05*)			[80]
2004	Caucasian (USA)	128	IIIA-IV	Platinum-based			0.84 (0.53-1.34)	CC						[141]
2005	Caucasian (USA)	214	IIA-IV	Platinum-based							Hematological Gastrointestinal	1.08 (0.59-1.91) 1.47 (0.66-3.28)	CC	[162]
2006	Asian (Korea)	245	IIIA-IV	CDDP-based			1.42 (1.08-1-86)	CC						[136]
2006	Caucasian (Spain)	135	III-IV	CDDP GMZ			0.69 (0.39-1.23)	CC						[147]
2007	Asian (China)	76	IIIA-IV	Platinum-based	0.26 (0.09-0.74)	CC								[125]
2008	Caucasian (Italy)	65	IIIB-IV	CDDP+GMZ	1.89 (0.35-10.19)	CC	0.51 (0.17-1.52)	CC	0.36 (0.13. 0.98)	CC	Grade 3-4: Neutropenia (p=0.31*) Trombocitopenia (p=0.72*) Anemia (p=0.35*) Non-hematological (p=0.35*)			[146]
2009	Caucasian (Greece)	119	IIIA-IV	Platinum-based	0.53 (0.19-1.46)	CC	1.37 (0.76-1.52)	CC			Severe Toxicity (p>0.05*)			[142]
2009	Caucasian-Asian (USA-Japan)	381	III-IV	Platinum-based			1.20 (0.74-1.96)	TT	1.11 (0.69 -1.82)	TT	Neutropenia grade 3-4	0.57 (0.20-1.61)	TT	[159]
2010	Asian (China)	90	IIIB-IV	CDDP-based	0.68 (0.26-1.75)	CC								[166]
2010	Asian (China)	115	IIIB-IV	CDDP/CBDCA DTX/GMZ/VNR	3.14 (1.33-7.41)	CC								[129]
2010	Caucasian (USA)	130	IIIB-IV	CDDP/CBDCA GMZ/PTX/VNR	0.85 (0.35-2.06)	T	0.59 (0.36-0.96)	CC						[138]
2010	Asian (China)	95	IIIB-IV	CDDP GMZ/PTX/VNR	0.80 (0.36-1.79)	CC					Hematological (p=0.269*) Gastrointestinal (p=0.343*) Nephrotoxicity/ Hepatotoxicity (p=0.489*)			[158]

Table 2. (Continued).

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
							rs11615							
2010	Asian (Japan)	122	I-III	Platinum-based			0.95 (0.33-2.74)	T	0.49 (0.20-1.18)	T				[140]
2010	Asian (Japan)	640	IB-IV	Platinum-based	1.13 (0.81-1.59)	CC								[64]
2011	Caucasian (Italy)	80	IIIB-IV	CDDP/CBDCA GMZ/PTX/VP16	p>0.05*		p>0.05**		p>0.05**		Hematological/ Non-hematological (p>0.05*)			[161]
2011	Caucasian (Italy)	192	IIIB-IV	CDDP GMZ/PTX/VNR	1.23 (0.62-2.43)	CC	p>0.05**		p>0.05**		Grade 3-4	Hematological: 1.02 (0.40–2.60) CT 2.68 (0.86-8.36) TT Non-hematological 3.20 (0.69-15.2) CT 2.20 (0.29-16.8) TT	CC	[160]
2011	Caucasian (France)	85	II-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR					p=0.24**					[148]
2011	Caucasian (Spain)	94	IIIB-IV	CDDP+VNR	1.11 (0.40-3.09)	CC	1.22 (0.94-1.66)	CC	0.93 (0.53-1.62) CC 0.92 (0.57-1.49) CT	TT	Severe Toxicity (p>0.05*)			[163]
2011	Caucasian (Spain)	493	IIIB-IV	CDDP+DTX							Neutropenia (p=0.53*)			[164]
2011	Asian (Japan)	90	I-IV	Platinum-based			2.00 (1.01-3.93)	CC						[135]
2012	Caucasian (Netherlands)	137	IIIB-IV	CDDP GMZ	0.40 (0.16-0.98)	CC	1.50 (1.02-3.59)	CC	1.62 (1.01-2.61)	CC	Severe Toxicity (p>0.05*)			[110]
2012	Asian (China)	142	IIIB-IV	CDDP PTX/VNR	0.40 (0.18-0.86)	CC	1.86 (1.17-2.97)	CC	1.87 (1.38-2.54)	CC				[126]
2012	Asian (China)	568	III-IV	Platinum-based			1.45 (1.03-2.03)	CC						[134]
2012	Asian (China)	340	IIIB-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR			0.74 (0.56-0.97)	CC	p=0.512**					[139]
2012	Asian (China)	89	III-IV	CDDP DTX/GMZ/VNR	2.00 (0.79-5.09)	CC								[152]
2012	Caucasian (Poland)	43	IIIB-IV	CDDP/CBDCA GMZ/PTX/VNR/VP16	2.19 (0.52-9.27)	CC	0.503 (0.13-1.14)	TT	0.44 (0.08-0.88)	TT				[151]
2012	Asian (China)	62	IIIB-IV	Platinum-based	0.74 (0.24-2.23)	CC	0.62 (0.17-2.18) CC 0.55 (0.14-2.16) CT	TT	p=0.357**					[153]
2012	Asian (China)	632	I-IV	CDDP GMZ			1.15 (0.81-1.28)	CC						[155]
2013	Caucasian (Italy)	98	IIIA-IV	CDDP-based	1.05 (0.39-2.78)	CC	0.62 (0.37-1.03)	T	0.85 (0.52-1.39)	T				[154]
2013	Asian (China)	135	IIIB-IV	CDDP/CBDCA GMZ	1.35 (0.60-3.06)	CC	p=0.73**		0.90 (0.61-1.34)	CC				[149]

Table 2. (Continued).

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival					
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	Type	OR (95% CI)	Ref. Cat.	
rs11615														
2014	Asian (China)	163	IIIA-IV	Platinum-based	0.53 (0.33-0.86)	C	1.97 (1.20-3.34)	C						[128]
2014	Asian (China)	91	IIIB-IV	CDDP DTX/GMZ/VNR	10.16 (1.78-11.16)	CC			p<0.01**					[105]
2014	Asian (China)	161	IIIA-IV	CDDP/CBDCA DTX/GMZ/PMX/PTX/VNR	0.66 (0.33-1.29)	CC			p>0.05**					[130]
2014	Caucasian (Spain)	74	IIIA-IV	CDDP/CBDCA DTX/GMZ/PMX/PTX/VNR	0.11 (0.01-0.66) III 0.61 (0.21-1.67) IV CT 1.08 (0.25-4.38) IV CC	T III TT IV					Severe Toxicity (p>0.05*)			[132]
2014	Asian (China)	192	IIIA-IV	Platinum-based	0.50 (0.32-0.78)	C	1.57 (1.02-2.47)	C-						[133]
2014	Asian (China)	187	IIIA-IV	CDDP/CBDCA PTX	0.66 (0.36-1.23)	CC	1.39 (0.72-2.69)	CC						[61]
2014	Caucasian (Poland)	115	II-IV	CDDP/CBDCA PMX	p=0.802*		0.66 (0.33-1.33)	CC	0.68 (0.36-1.26)	CC				[150]
2015	Caucasian (Greece)	107	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	p=0.812*		0.76 (0.44-1.32)	TT	0.76 (0.47-1.23)	TT	Hematological (p>0.05*)			[143]
2015	Asian (China)	240	III-IV	CDDP DTX/GMZ/PTX/VNR	1.31 (0.70-2.44) CT 2.73 (1.21-6.18) TT	CC	0.76 (0.41-1.41) CT 0.38 (0.14-0.96) TT							[131]
2015	Caucasian (Poland)	55	IIIB-IV	CDDP/CBDCA+VNR							Hematological (p>0.05*) Nephrotoxicity (p>0.05*) Hepatotoxicity (p>0.05*)			[81]
rs3212986														
2005	Caucasian (USA)	214	IIA-IV	Platinum-based							Hematological Gastrointestinal	1.06 (0.59-1.91) 1.47 (0.66-3.28)	CC	[162]
2009	Caucasian (Greece)	119	IIIA-IV	Platinum-based	1.35 (0.62-2.95)	CC	0.5 (0.33-0.84)	CC			Severe Toxicity (p>0.05*)			[142]
2010	Asian (China)	115	IIIB-IV	CDDP/CBDCA DTX/GMZ/VNR	0.23 (0.10-0.57)	CC								[129]
2010	Asian (Japan)	122	I-III	Platinum-based			5.01 (1.67-14.99)	CC	2.30 (1.03-5.15)	CC				[140]
2011	Caucasian (Spain)	493	IIIB-IV	CDDP DTX							Neutropenia (p=0.92*)			[164]
2011	Asian (China)	300	IV	CDDP/CBDCA GMZ/PTX/VNR	1.17 (0.72-1.93)	CC	p>0.911*	CC			Grade 1-3 (p>0.05*)			[165]
2013	Caucasian (Italy)	98	IIIB-IV	CDDP-based	1.81 (0.78-4.19)	CC	0.90 (0.59-1.36)	A	0.67 (0.44-1.01)	A				[154]
2014	Asian (China)	163	IIIA-IV	Platinum-based	0.44 (0.27-0.74)	C	1.97 (1.13-3.35)	C	2.30 (1.03-5.15)	CC				[128]

Table 2. (Continued).

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs3212986														
2014	Caucasian (Spain)	74	IIIA-IV	CDDP/CBDCA DTX/GMZ/PMX/PTX/VNR	2.13 (0.47-12.46) III AC 0.71 (0.07-9.18) III AA 1.68 (0.65-4.36) IV	CC					Severe Toxicity (p=0.806*)			[132]
2015	Caucasian (Greece)	107	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	p=0.104*		0.64 (0.41-1.0)	CC	0.50 (0.32-0.77)	CC	Hematological (p=0.537*)			[143]
2015	Caucasian (Poland)	55	IIIB-IV	CDDP/CBDCA VNR							Hematological (p>0.05*) Nephrotoxicity (p>0.05*) Hepatotoxicity (p>0.05*)			[81]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine; VP16: etopósido.
*p-value for chi-square test; ** p-value for long rank test.
The blanks mean that the paper did not provide any information on this parameter.

1.10.1.1.2 ERCC2

The ERCC2 protein is a component of the general transcription factor IIH (TFIIH) and its helicase activity plays a key role in gene transcription and nucleotide excision repair [123].

Numerous SNPs in *ERCC2* gene have been described, of which rs13181 (A→C substitution at codon 751, exon 23, Lys→Gln) and rs1799793 (G→A substitution at codon 312, exon 10, Asp→Asp) are the most investigated. Although both SNPs cause suboptimal DNA repair capacity [167,168], no significant association has been found between *ERCC2* Lys751Gln or Asp312Asn and ORR or PFS (Table 3). This lack of association has been confirmed in two recent meta-analyses, that included 22 studies/3240 patients [60] and 12 studies/1737 patients [157]. In contrast, several studies have found an association between OS and both SNPs (Table 3). The genotype CC for *ERCC2* Lys751Gln has been correlated with a longer OS compared to the AC/AA genotypes in Asian population (HR=1.54; $CI_{95\%}$=1.03-2.29; AC/AA vs CC) [173]. In the case of Asp312Asn, the AG/AA genotype has been associated with poor survival both in Caucasian and Asian populations (Table 3). A meta-analysis has also confirmed these results in Asian (HR=2.07; $CI_{95\%}$=1.11-3.88; I^2=62.7%; $P_{heterogeneity}$=0.07; AG/AA vs GG) but not in Caucasian population (HR=0.84; $CI_{95\%}$=0.62-1.14; I^2=52.2%; $P_{heterogeneity}$=0.08; AG/AA vs GG) [60].

A relationship between *ERCC2* Asp312Asn and Lys751Gln and hematological toxicity has been reported in two small studies including 55 and 62 patients [80,81], in which the A-allele of *ERCC2* Lys751Gln was associated with increased grade 2-3 neutropenia (p=0.04) [80] and the G-allele Asp312Asn with reduced frequency of severe hematological toxicity (OR=0.08; $CI_{95\%}$=0.01-0.40; p=0.0005; AG/GG vs AA) [81].The AA genotype of *ERCC2* Lys751Gln has also been associated with higher risk of severe nephrotoxicity (OR=0.07; $CI_{95\%}$=0.02-0.31; AC/CC vs AA) [81]. Other studies in larger patient populations (65 to 493) failed to find such associations between *ERCC2* polymorphisms and platinum-based chemotherapy toxicity (Table 3).

Finally, three polymorphisms in *ERCC2* (rs50872, rs238405, rs238416) were found to be correlated with clinical outcome in a cohort of 129 Asian advanced NSCLC patients [181], with rs50872 associated with longer OS (p=0.009) and PFS (p=0.032) in all patients, rs238405 in patients treated with a combination of platinum and taxanes and rs238416 in those receiving platinum and gemcitabine doublets [181].

Table 3. Influence of ERCC2 polymorphisms on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs13181														
2003	Caucasian (Spain)	38	IIIB-IV	CDDP GMZ	2.04 (0.49-8.45)	CC								[170]
2004	Caucasian (Spain)	62	IIIB-IV	CDDP DTX	0.90 (0.32-2.53)	CC	0.87 (0.65.1.51)	CC	0.90 (0.54-1.51)	CC	Neutropenia grade 2-4 (p=0.04*)			[80]
2004	Asian (Korea)	109	IIIB-IV	CDDP DTX/GMZ/PTX	0.74 (0.22-2.51)	CC	p=0.47**							[137]
2006	Caucasian (United Kingdom)	108	III-IV	Platinum-based	0.74 (0.31-1.80)	CC	0.95 (0.50-1.80)	CC						[169]
2006	Caucasian (Spain)	135	III-IV	CDDP GMZ			0.99 (0.65-1.51)	CC	0.82 (0.57-1.51)	CC				[147]
2007	Caucasian (Italy)	188	IIIA-IV	CDDP/CBDCA GMZ	0.94 (0.56-1.58) AC 0.81 (0.39-1.66) CC	AA	1.11 (0.82-1.51) AC 1.10 (0.70-1.75) CC	AA						[176]
2008	Caucasian (Italy)	65	IIIB-IV	CDDP GMZ	0.61 (0.21-1.79)	CC	1.09 (0.50-2.36)	CC	1.49 (0.78-2.86)	CC	Grade 3-4: Neutropenia (p=0.71*) Trombocitopenia (p=0.90*) Anemia (p=0.32*) Non-hematological (p=0.65*)			[146]
2009	Caucasian (Greece)	119	IIIA-IV	Platinum-based	1.47 (0.61-3.56)	CC	0.70 (0.44-1.12)	CC			Severe Toxicity (p>0.05*)			[142]
2009	Caucasian-Asian (USA-Japan)	381	III-IV	Platinum-based			1.20 (0.74-1.96)	TT	1.11 (0.69 -1.82)	TT	Neutropenia grade 3-4	1.33 (0.54-3.33)		[159]
2009	Asian (China)	108	IIIB-IV	Platinum-based	0.36 (0.08-1.69)	CC	0.55 (0.29-1.07)	CC						[177]
2010	Asian (Japan)	640	IB-IV	Platinum-based	0.68 (0.27-1.74)	CC								[64]
2010	Asian (China)	115	IIIB-IV	CDDP/CBDCA DTX/GMZ/VNR	0.56 (0.21-1.45)	CC								[129]
2011	Caucasian (Italy)	192	IIIB-IV	CDDP GMZ/PTX/VNR	1.22 (0.62-2.38)	CC	0.98 (0.66-1.47) AC 0.80 (0.49-1.29) CC	AA	0.93 (0.66-1.31) AC 0.68 (0.44-1.03) CC	AA	Grade 3-4	Hematological: 1.22 (0.49-3.01) AC 0.98 (0.31-3.03) CC Non-hematological 0.52 (0.15-1.81) AC 0.98 (0.26-3.75) CC	AA	[160]
2011	Caucasian (Spain)	94	IIIB-IV	CDDP VNR	1.17 (0.50-2.74)	CC	0.99 (0.60-1.67) AC 0.73 (0.36-1.48) CC	AA	0.84 (0.52-1.35) AC 0.90 (0.47-1.72) CC	AA	Severe Toxicity (p>0.05*)			[163]
2011	Asian (China)	199	III-IV	Platinum based			1.33 (0.86-2.04)	CC	1.51 (0.92-2.48)	CC				[179]
2011	Caucasian (France)	85	II-IV	CDDP/CBDCA GMZ/DTX/PTX/VNR					0.80 (0.57-1.11)	CC				[148]

Table 3. (Continued).

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs13181														
2011	Asian (China)	115	IIIB-IV	Platinum based							Grade 1-4 (p>0.05*)			[180]
2011	Caucasian (Spain)	493	IIIB-IV	CDDP DTX							Neutropenia grade 1-4 (p=0.72*)			[164]
2012	Caucasian (Netherlands)	137	IIIB-IV	CDDP GMZ	0.95 (0.45-2.01)	CC	0.8 (0.54-1.20) AC 0.66 (0.35-1.24) CC	AA	0.84 (0.58-1.21) AC 0.85 [0.47-1.52) CC	AA	Severe Toxicity (p>0.05*)			[110]
2012	Asian (China)	340	IIIB-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR			1.20 (0.87-1.65)	CC	p=0.41**					[139]
2012	Asian (China)	353	III-IV	CDDP/CBDCA DTX/GMZ/PTX	1.48 (0.65-3.40)	CC	1.54 (1.03-2.29)	CC	p=0.062**					[173]
2012	Caucasian (Italy)	208	IIIB-IV	CBDCA PMX	p=0.99*		p=0.06**		p=0.12**					[70]
2012	Asian (China)	355	IIIB-IV	CDDP/CBDCA GMZ/PTX/VNR			p=0.019** elderly patients p=0.042 **non-adenocarcinoma							[182]
2012	Caucasian (Spain)	180	IIIB-IV	CDDP VNR	0.75 (0.36-1.55)	CC			1.209 (0.8-1.7)	CC	Grade ≥3: Hematological (p>0.05*) Asthenia (p>0.05*) Gastrointestinal (p>0.05*)			[172]
2012	Asian (China)	62	IIIB-IV	Platinum based	0.18 (0.01-3.42)	CC	0.91 (0.21-3.91)	CC	p=0.428**					[153]
2013	Caucasian (Italy)	98	IIIB-IV	CDDP-based	0.57 (0.25-1.31)	CC	0.99 (0.64-1.51)	C	0.78 (0.51-1.20)	C				[154]
2013	Asian (China)	496	III-IV	Platinum-based	0.70 (0.42-1.17)	CC	1.34 (0.79-1.75)	CC	1.42 (0.73-1.82)	CC				[171]
2013	Asian (China)	115	IIIB-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR			1.20 (0.78-1.83)	CC	1.28 (0.83-1.96)	CC				[178]
2014	Asian (China)	161	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/PMX/VNR	1.06 (0.47-2.39)	AA	p>0.05**							[130]
2014	Caucasian (Spain)	74	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/PMX/VNR	2.06 (0.53-8.88) III AC 5.03 (0.44-142.6) III CC 1.16 (0.42-3.24) IV AC 0.41 (0.05-2.37) IV CC	AA					Severe Toxicity (p>0.05*)			[132]
2014	Asian (China)	375	IIIA-IV	CDDP/CBDCA DTX/PTX/VNR	0.61 (0.34-1.18)	CC	1.41 (0.85-1.85)	CC	1.34 (0.75-1.82)	CC				[174]
2014	Asian (China)	93	IIIB-IV	CDDP GMZ	p=0.517*		p=0.03*		p=0.39*					[175]
2015	Caucasian (Greece)	107	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	p=0.666*		1.28 (0.79-2.08)	AA	1.14 (0.74-1.76)	AA	Hematological (p=0.826*)			[143]

Table 3. (Continued).

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs13181														
2015	Caucasian (Poland)	55	IIIB-IV	CDDP/CBDCA VNR							Hematological (p>0.05*) Nephrotoxicity Hepatotoxicity (p>0.05*)	0.07 (0.02-0.31)	AA	[81]
rs1799793														
2003	Caucasian (Spain)	38	IIIB-IV	CDDP GMZ	3.33 (0.66-16.74)	GG								[170]
2004	Caucasian (Spain)	62	IIIB-IV	CDDP DTX	0.72 (0.26-2.02)	GG	1.12 (0.55-2.28)	GG	1.55 (1.04-2.32)	GG	Grade 2-4 (p>0.05*)			[80]
2004	Asian (Korea)	109	IIIB-IV	CDDP DTX/GMZ/PTX	1.08 (0.26-4.57)	GG	p=0.56**							[137]
2006	Caucasian (UK)	108	III-IV	Platinum-based	0.94 (0.38-2.33)	GG								[169]
2008	Caucasian (Italy)	65	IIIB-IV	CDDP GMZ	0.69 (0.24-2.00)	GG	p=0.72**		p=0.78**		Grade 3-4: Neutropenia (p=0.17*) Trombocitopenia (p=0.35*) Anemia (p=0.66*) Non-hematological (p=0.60*)			[146]
2009	Caucasian (Greece)	119	IIIA-IV	Platinum-based	0.78 (0.34-1.80)	GG	0.80 (0.53-1.37)	GG			Severe Toxicity (p>0.05*)			[142]
2011	Caucasian (Spain)	94	IIIB-IV	CDDP VNR	1.10 (0.47-2.56)	GG	0.82 (0.52-1.31) AG 0.87 (0.44-1.70) AA	GG	0.79 (0.47-1.33) AG 0.80 (0.40-1.62) AA	GG	Severe Toxicity (p>0.05*)			[163]
2011	Caucasian (Spain)	493	IIIB-IV	CDDP DTX							Neutropenia grade 1-4 (p=0.89*)			[164]
2012	Caucasian (Netherlands)	137	IIIB-IV	CDDP GMZ	1.90 (0.87-4.15)	GG	0.58 (0.39-0.85)	GG	0.92 (0.63-1.34)	GG	Severe Toxicity (p>0.05*)			[110]
2012	Asian (China)	353	III-IV	CDDP/CBDCA DTX/GMX/PTX	1.64 (0.71-3.79)	GG	1.55 (1.04-2.32)	GG	p=0.790**					[173]
2012	Caucasian (Spain)	180	IIIB-IV	CDDP VNR	1.22 (0.62-2.38)	GG			p=0.64**	CC	Grado ≥3: Hematological (p>0.05*) Asthenia (p>0.05*) Gastrointestinal (p>0.05*)			[172]
2013	Asian (China)	496	III-IV	Platinum-based	0.72 (0.48-1.08)	GG	1.85 (1.12-3.15)	GG	1.27 (0.68-1-84)	GG				[171]
2013	Asian (China)	62	IIIB-IV	Platinum-based	2.33 (0.14-39.39)	GG	14.04 (2.25-87.51)	GG	p=0.672**					[153]

Table 3. (Continued).

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs1799793														
2014	Caucasian (Spain)	74	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/PMX/VNR	1.26 (0.31-5.43) AG III Inf. (0.00-NR) AA III 0.77 (0.27-2.13) AG IV 0.98 (0.21 -4.25) AA IV	GG					Severe Toxicity (p>0.05*)			[132]
2014	Asian (China)	375	IIIA-IV	CDDP/CBDCA DTX/PTX/VNR	0.67 (0.36-0.97)	GG	1.73 (0.97-2.92)	GG	1.29 (0.67-1.93)	GG				[174]
2014	Asian (China)	93	IIIB-IV	CDDP GMZ	p=0.517*		p=0.03*		p=0.39*					[175]
2015	Caucasian (Greece)	107	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	p=0.288*		0.96 (0.60-1.53)	GG	0.69 (0.45-1.0)	GG	Hematological (p=0.999*)			[143]
2015	Caucasian (Poland)	55	IIIB-IV	CDDP/CBDCA VNR							Hematological Nephrotoxicity (p>0.05*) Hepatotoxicity (p>0.05*)	0.08 (0.01-0.40)	AA	[81]
rs50872														
2012	Asian (Korea)	129	IIIA-IV	CDDP/CBDCA DXT/GMZ/PTX			p=0.009**		p=0.032**					[181]
rs238405														
2012	Asian (Korea)	129	IIIA-IV	CDDP/CBDCA DXT/GMZ/PTX			p=0.012**		p=0.008**					[181]
rs238416														
2012	Asian (Korea)	129	IIIA-IV	CDDP/CBDCA DXT/GMZ/PTX			p=0.004**		p=0.033**					[181]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine; NR: not reached; Inf: Infinite.
*p-value for chi-square test; ** p-value for long rank test.
The blanks mean that the paper did not provide any information on this parameter.

1.10.1.1.3 ERCC5

ERCC5 encodes for a single-strand specific DNA endonuclease, which cleaves the damaged DNA strand 3' to the lesion during nucleotide excision repair [183].

The TT genotype in the *ERCC5* rs1047768 (T→C substitution at codon 46, exon 2, His→His) has been associated with increased ORR (OR=1.90; $CI_{95\%}$=1.10-3.28; TT vs CC), OS (HR=0.52; $CI_{95\%}$=0.31, 0.96; TT vs CC) and PFS (HR=0.47; $CI_{95\%}$=0.22-0.82; TT vs CC) [184]. A meta-analysis which included 5 studies with 846 cases, has recently confirmed these results for ORR (relative risk (RR)=1.23; $CI_{95\%}$=1.11-1.36 I^2=0%; $P_{heterogeneity}$=0.480; CT/TT vs CC) [157] while no association of rs1047768 with toxicity was found in 74 Spanish NSCLC patients [132]. In contrast, *ERCC5* rs17655 (G→C substitution at codon 1104, exon 15, Asp→His) has been related with a higher infection rate (p=0.017) in 388 Chinese NSCLC patients [74] but not with clinical outcome (Table 4).

Three additional polymorphisms in *ERCC5* (rs2094258, rs2296147, rs873601) have also been investigated. The G-allele for *ERCC5* rs2094258 has been associated with increased OS (HR=0.51; $CI_{95\%}$=0.39-0.82; AG/GG vs AA) and PFS (HR=0.44; $CI_{95\%}$=0.34-0.78; AG/GG vs AA) in 433 advanced NSCLC patients treated with platinum-based chemotherapy [187]. In the case of *ERCC5* rs2296147, two studies in 433 and 277 Chinese patients found a significant association with OS (HR=0.66; $CI_{95\%}$=0.48-0.99 for CT/TT vs CC and HR=0.49; $CI_{95\%}$=0.36, 0.68 for T vs C allele) and PFS (HR=0.73; $CI_{95\%}$=0.51-0.97 for CT/TT vs CC and HR=0.52; $CI_{95\%}$=0.38-0.70 for T vs C allele) [185,187]. Finally, the allele G of rs873601 correlated with a better OS (HR=0.72; $CI_{95\%}$=0.55-0.97; G vs A allele) in 277 Chinese stage III/IV NSCLC patients [185].

1.10.1.2 BER pathway

The BER pathway removes small, non-helix-distorting base lesions from the genome, including single strand breaks produced by oxidation, methylation, deamination and hydroxylation [188]. These lesions may not block transcription and normal replication, but they usually cause miscoding. In this process, the enzymes poly (ADP ribose) polymerase 1 and 2 (PARP1 and PARP2) play a key role, along with apurinic/apyimidinic endonuclease 1 (APE1) and XRCC1 [189,190].

Table 4. Influence of ERCC5 polymorphisms on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival					
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	Type	OR (95% CI)	Ref. Cat.	
rs1047768														
2009	Asian (China)	82	IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	1.64 (0.56-5.83) CT 1.02 (0.37-3.86) TT	CC								[186]
2014	Caucasian (Spain)	74	IIIA-IV	CDDP/CBDCA DTX/GMZ/VNR/PMX/PTX	3.12 (0.61-17.66) III CT 0.90 (0.14-5.78) III TT 1.42 (0.52-3.97) IV CT 0.61 (0.13-2.48) IV TT	CC III TT IV					Severe Toxicity (p>0.05*)			[132]
2014	Asian (China)	378	I-IV	CDDP DTX/GMZ/PTX/VNR	1.29 (0.76-2.21) CT 1.90 (1.10-3.28) TT	CC	0.88 (0.58-1.31) CT 0.52 (0.31-0.96) TT	CC	0.85 (0.58-1.24) CT 0.47 (0.22-0.82) TT	CC				[184]
2014	Asian (China)	277	III-IV	Platinum basaed			0.94 (0.71-1.25)	T	0.99 (0.75-1.30)	T				[185]
rs17655														
2009	Asian (China)	82	IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	0.95 (0.29-3.03) CG 1.333 (0.34-5.18) GG	CC								[186]
2011	Caucasian (France)	85	II-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR					p=0.84**					[148]
2012	Asian (China)	388	IIIB-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR							Infection	1.72 (1.00-13.22)	CC	[74]
2012	Asian (China)	340	IIIB-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR			p=0.425**		p=0.191**					[139]
2013	Asian (China)	433	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR			0.95 (0.72-1.32)	GG	0.97 (0.71-1.34)	GG				[187]
2014	Caucasian (Spain)	74	IIIA-IV	CDDP/CBDCA DTX/GMZ/VNR/PMX/PTX	0.44 (0.09-2.21) III CG 0.23 (0.02-3.12) III CC 1.44 (0.56-3.79) IV C	GG III/IV					Severe Toxicity (p>0.05*)			[132]
2014	Asian (China)	277	III-IV	Platinum-based			0.82 (0.62-1.08)	C	0.83 (0.63-1.09)	C				[185]
2014	Asian (China)	378	I-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	1.13 (0.69-1.86) CG 1.40 (0.75-2.59) GG	CC	0.94 (0.64-1.38) CG 0.90 (0.55-1.47) GG	CC	0.92 (0.64-1.32) CG 0.83 (0.52-1.32) GG	CC				[184]
rs2094258														
2013	Asian (China)	433	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR			0.51 (0.39-0.82)	AA	0.44 (0.34-0.78)	AA				[187]
rs2296147														
2013	Asian (China)	433	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR			0.66 (0.48-0.99)	CC	0.73 (0.51-0.97)	CC				[187]
2014	Asian (China)	277	III-IV	Platinum-based			0.49 (0.36-0.68)	C	0.52 (0.38-0.70)	C				[185]
rs873601														
2014	Asian (China)	277	III-IV	Platinum-based			0.72 (0.55-0.97)	A	0.84 (0.64-1.09)	A				[185]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine.
*p-value for chi-square test; ** p-value for long rank test.
The blanks mean that the paper did not provide any information on this parameter.

1.10.1.2.1 XRCC1

The XRCC1 protein interacts with DNA polymerase-beta, DNA ligase III and PARP (poly ADP-ribose polymerase), repairing the damaged DNA strand [191].

XRCC1 rs1799782 (A→G substitution at codon 194, exon 6, Arg→Trp) and rs25487 (A→G substitution at codon 399, exon 10, Gln→Arg) are the most studied polymorphisms in *XRCC1*. The T-allele of *XRCC1* Arg194Trp has been associated with better ORR in Asian population, but not in Caucasian patients (Table 5). A recent meta-analysis, which involved 11 studies and compiled 1.329 cases, has reported similar results in Asian population (OR=0.38; $CI_{95\%}$=0.30-0.48; I^2=0%; $P_{heterogeneity}$=0.830; CT/TT vs CC) [62]. For *XRCC1* Gln399Arg, several studies in Asian population have found a relationship between genotype GG and higher ORR (Table 5). This result has been confirmed by a recent meta-analysis, which evaluated 13 studies and 1.334 cases from Asian population (OR=2.05; $CI_{95\%}$=1.62-2.60; I^2=26%; $P_{heterogeneity}$=0.18; GG vs AG/AA) [62]. However, more recent studies in 325 and 147 Chinese patients have reported poor ORR for GG genotype [104,197]. No significant association has been reported in Caucasian population (Table 5).

The *XRCC1* Gln399Arg polymorphism has also been associated with OS, PFS and toxicity (Table 5). A longer OS for the AA genotype has been reported in two studies in Asian (OR=1.69; $CI_{95\%}$=1.19-2.39 for GG vs AG/AA and OR=0.17; $CI_{95\%}$=0.06-0.41 for AA vs GG) and one in Caucasian populations (OR=0.47; $CI_{95\%}$=0.23-0.95 for A vs G allele) [76,104,154]. The GG genotype showed significant results for PFS in 111 stage-IV NSCLC patients (OR=1.77; $CI_{95\%}$=1.14-2.73 for AG/AA vs GG) [108] and has also been linked with increased hematological (OR=0.32; $CI_{95\%}$=0.12-0.86 for AG/AA vs GG and OR=0.22; $CI_{95\%}$=0.06-0.82 for AG/AA vs GG) and gastrointestinal toxicity (OR=0.29; $CI_{95\%}$=0.11-0.83 for AG/AA vs GG) [75,76,81]. In contrast, other study reported higher incidence of hematological toxicity for AA/AG genotype (OR=2.14; $CI_{95\%}$=1.21-3.78 for AG/AA vs GG) [76]. Finally, no associations between OS, PFS, toxicity and *XRCC1* Arg194Trp SNPs have been found (Table 5).

1.10.1.2.2 PARP1, PARP2 and APE1

The role of *PARP1*, *PARP2* and *APE1* polymorphisms on clinical outcomes to cisplatin in advanced NSCLC has been less investigated. The T-allele for *APE1* rs1760944 polymorphism was found to be associated with gastrointestinal toxicity in 235 patients [76], and the GG genotype for *APE1*

Table 5. Influence of XRCC1 polymorphisms on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs1799782														
2004	Asian (China)	105	IIIB-IV	Platinum-based	0.34 (0.14-0.80)	C								[193]
2006	Asian (China)	200	IIIB-IV	Platinum-based	0.43 (0.23-0.78)	C								[195]
2009	Asian (China)	82	IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	0.28 (0.11-0.73)	C								[186]
2009	Asian (China)	164	III-IV	CDDP-based	0.49 (0.25-0.96)	C								[192]
2011	Asian (China)	130	IIIB-IV	Platinum-based	0.39 (0.18-0.86)	C								[194]
2012	Asian (China)	460	III-IV	CDDP DTX/GMZ/PTX/VNR			1.23 (0.81-1.89) CT 0.45 (0.23-0.87) TT	CC						[198]
2013	Asian (China)	147	IIIB-IV	CDDP/CBDCA DTX/GMZ/PMX/VNR	0.92 (0.43-1.96)	CC	0.88 (0.48-1.61)	CC	1.08 (0.68-1.72)	CC				[197]
2014	Caucasian (Spain)	74	IIIA-IV	CDDP/CBDCA DTX/GMZ/PMX/PTX/VNR	5.86 (0.64-151.17) III 0.46 (0.06-2.15) IV	CC					Severe Toxicity (p>0.05*)			[132]
2014	Asian (China)	375	IIIA-IV	CDDP/CBDCA DTX/PTX/VNR	1.10 (0.72-1.71)	CC	0.81 (0.56-1.66)	CC	0.85 (0.57-1.59)	CC				[174]
2014	Asian (China)	378	I-IV	CDDP DTX/GMZ/PTX/VNR	1.28 (0.78-2.09) CT 1.63 (0.84-3.12) TT	CC	0.91 (0.61-1.33) CT 0.82 (0.47-1.39) TT	CC	0.95 (0.66-1.37) CT 0.79 (0.47-1.31) TT	CC				[184]
2015	Asian (China)	325	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	1.18 (0.68-2.05) CT 1.41 (0.61-3.47) TT	CC	0.75 (0.45-1.26) CT 0.74 (0.33-1.60) TT	CC						[104]
2015	Caucasian (Poland)	55	IIIB-IV	CDDP/CBDCA VNR							Hematological (p>0.05*) Nephrotoxicity (p>0.05*) Hepatotoxicity (p>0.05*)			[81]
2015	Asian (China)	322	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	1.29 (0.74-2.26) CT 1.98 (0.84-5.07) TT	CC	0.77 (0.44-1.34) CT 0.66 (0.30-1.49) TT	CC						[199]
rs25487														
2004	Asian (China)	105	IIIB-IV	Platinum-based	2.65 (1.12-6.26)	A								[193]
2007	Caucasian (Italy)	188	IIIA-IV	CDDP/CBDCA GMZ	0.67 (0.31-1.45) AA 0.79 (0.47-1.31) AG	GG	0.61 (0.36-1.03) AG 1.09 (0.80-1.47) AA	GG						[176]
2009	Caucasian (Greece)	119	IIIA-IV	Platinum-based	1.15 (0.49-2.72)	G	1.43 (0.85-2.40) AG 4.58 (1.92-10.92) AA	GG			Severe Toxicity (p>0.05*)			[142]
2009	Asian (China)	82	IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	1.00 (0.38-2.65)	A								[186]
2009	Asian (China)	164	III-IV	CDDP-based	1.20 (0.63-2.30)	A								[192]
2011	Asian (China)	111	IV	CDDP/CBDCA DTX/GMZ/PMX/VNR	3.70 (1.31-10.47)	GG			1.77 (1.14-2.73)	GG				[108]
2011	Asian (China)	130	IIIB-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	2.98 (1.38-6.40)	A								[194]
2012	Caucasian (Netherlands)	137	IIIB-IV	CDDP+GMZ	p>0.05*		0.62 (0.34-1.11) AG 0.56 (0.30-1.01) GG	AA	0.91 (0.53-1.58) AG 1.03 (0.59-1.82) GG	AA	Severe Toxicity (p>0.05*)			[110]

Table 5. (Continued).

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs25487														
2012	Asian (China)	89	III-IV	CDDP DXT/GMZ/VNR	4.81 (1.78-13.01)	A								[152]
2012	Asian (China)	460	III-IV	CDDP DTX/GMZ/PTX/VNR			0.76 (0.53-1.07) AG 0.42 (0.21-0.82) AA	GG						[198]
2013	Caucasian (Italy)	98	IIIB-IV	CDDP-based	1.22 (0.32-4.64)	G	0.47 (0.23-0.95)	G	0.83 (0.41-1.70)	G				[154]
2013	Asian (Korea)	382	III-IV	CDDP PTX	3.92 (1.37-11.21)	G	1.41 (0.88-2.26)	G						[196]
2013	Asian (China)	147	IIIB-IV	CDDP/CBDCA DTX/GMZ/PMX/VNR	2.35 (1.11-5.00)	GG	0.89 (0.49-1.62)	GG	0.79 (0.50-1.25)	GG				[197]
2014	Asian (China)	375	IIIA-IV	CDDP/CBDCA DTX/PTX/VNR	1.69 (1.12-2.64)	GG	0.55 (0.23-0.94)	GG	0.61 (0.31-1.22)	GG				[174]
2014	Asian (China)	487	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	p=0.930*		1.69 (1.19-2.39)	A	p=0.205**		Hematological Gastrointestinal	2.14 (1.21-3.78) 0.91 (0.45-1.84)	GG	[76]
2014	Caucasian (Spain)	74	IIIA-IV	CDDP/CBDCA DTX/GMZ/PMX/PTX/VNR	0.96 (0.24-3.88) III AG 3.89 (0.43-95.90) III AA 0.67 (0.23-1.88) IV AG 0.35 (0.08-1.35) IV AA	GG					Severe Toxicity (p>0.05*)			[132]
2014	Asian (China)	378	I-IV	CDDP DTX/GMZ/PTX/VNR	1.53 (0.94-2.57) AG 2.27 (1.64-6.97) AA	GG	0.87 (0.60-1.24) AG 0.51 (0.26-0.98) AA	GG	0.81 (0.58-1.15) AG 0.48 (0.25-0.88) AA	GG				[184]
2015	Asian (China)	97	IIIB-IV	CDDP DXT/GMZ/PXT/VNR	1.407 (0.539-3.675)	GG			0.89 (0.58-1.38)	GG	Hematological Gastrointestinal	0.32 (0.12-0.86) 0.29 (0.11-0.83)	GG	[75]
2015	Asian (China)	325	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	1.97 (1.13-3.43) AG 3.76 (1.52-10.53) AA	GG	0.49 (0.29-0.83) AG 0.17 (0.06-0.41) AA	GG						[104]
2015	Caucasian (Greece)	107	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	p=0.529*		1.01 (0.64-1.5)	GG	0.83 (0.55-1.26)	GG	Hematological (p>0.05*)			[143]
2015	Caucasian (Poland)	55	IIIB-IV	CDDP/CBDCA VNR							Hematological	0.22 (0.06-0.82)	GG	[81]
2015	Asian (China)	322	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	1.53 (0.92-2.54) AG 3.37 (1.44-8.53) AA	GG	0.53 (0.31-0.91) AG 0.39 (0.18-0.83) AA	GG						[199]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine.
*p-value for chi-square test; ** p-value for long rank test.
The blanks mean that the paper did not provide any information on this parameter.

rs3136820 showed better OS compared to the TT genotype in 147 patients (HR=0.33; $CI_{95\%}$=0.12-0.92) [197]. A polymorphism in *PARP1* (rs1136410) has also been associated with survival; in particular, patients with CC genotype showed lower PFS than CT/TT patients (HR=1.90; $CI_{95\%}$=1.02-3.52) [197]. Finally, no studies have evaluated polymorphisms in *PARP2* and their associations with clinical outcomes or toxicity.

1.10.1.3 DSB pathway

The DSB pathway is involved in repairing the most severe lesions, affecting both strands, which may result in cell death or a diversity of genetic alterations, such as deletions and chromosomal aberrations [200]. Homologous recombination and non-homologous end-joining are the two main mechanisms of the DSB pathway [200]. Key proteins involved are MRE11 (MRE11 homolog A, double strand break repair nuclease), NBN (nibrin) and RAD50 (RAD50 double strand break repair protein), which form the MRE11 complex, BRCA1 (breast cancer 1), BRCA2 (breast cancer 2), RAD51 (RAD51 recombinase) and XRCC3 [200,201]. Despite of the essential function of these proteins in DSB pathway, only a polymorphism in XRCC3 gene has been extensively studied (Table 6).

The C→T substitution at codon 241, exon 7 of *XRCC3* (Thr→Met, rs861539) has been associated with ORR and OS, but not with PFS or toxicity (Table 6).The genotype CT/TT has shown higher ORR (OR=2.72; $CI_{95\%}$=1.17-6.31 for CT/TT vs CC) in 137 Caucasian stage III-IV NSCLC patients [110], a result confirmed by a meta-analysis that included 7 studies and 1.186 patients (OR=1.51; $CI_{95\%}$=1.10-2.07; I^2=0%; $P_{heterogeneity}$=0.618; CT/TT vs CC) [63]. The TT genotype has also been associated with prolonged OS in 135 Caucasian stage III-IV NSCLC patients (OR=0.43; $CI_{95\%}$=0.22-0.82 for TT vs CC) [147].

1.10.2 p53 pathway

The *TP53* tumor suppressor gene regulates the DNA repair through its ability to act as a transcription factor and to interact with damaged DNA, promoting the activation of DNA repair mechanisms [205,206]. In addition, *TP53* may induce apoptosis, arresting the cell cycle progression, when DNA damage is extensive (Figure 2) [207,208].

Table 6. Influence of XRCC3 polymorphism on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs861539														
2006	Caucasian (Spain)	135	III-IV	CDDP GMZ			0.60 (0.37-0.95) CC 0.43 (0.22-0.82) TT	CC						[147]
2011	Caucasian (Italy)	192	IIIB-IV	CDDP GMZ/PXT/VNR	1.60 (0.81-3.19)	CC	p>0.05**		p>0.05**		Grade 3-4	Hematological: 0.95 (0.37-2.43) CT 0.90 (0.32-2.52) CC Non-hematological 0.53 (0.17-1.68) CT 0.23 (0.05-1.19) CC		[160]
2011	Caucasian (Italy)	80	IIIB-IV	CDDP/CBDCA GMZ/PTX/VP16	p>0.05*		p>0.05**		p>0.05**		Hematological/ Non-hematological (p>0.05*)			[161]
2011	Caucasian (Spain)	493	IIIB-IV	CDDP DTX							Neutropenia grade 1-4 (p=0.37*)			[164]
2011	Caucasian (USA)	228	IA-IV	Platinum-based			1.09 (0.78-1.54)	CC						[203]
2012	Caucasian (Netherlands)	137	IIIB-IV	CDDP GMZ	2.72 (1.17-6.31)	CC	1.12 (0.74-1.71) CT 1.54 (0.87-2.72) TT	CC	0.82 (0.56-1.21) CT 1.21 (0.73-2.02) TT	CC	Severe Toxicity (p>0.05*)			[110]
2012	Asian (China)	62	IIIB-IV	Platinum-based	0.18 (0.01-3.42)	CC	0.91 (0.21-3.91)	CC	p=0.428**					[153]
2012	Asian (China)	340	IIIB-IV	CDDP/CBDCA DTX/GMZ/PXT/VNR			p=0.205**		p=0.532**					[139]
2012	Caucasian (Spain)	180	IIIB-IV	CDDP VNR	1.42 (0.71-2.82)	CC			p=0.44**	CC	Grado ≥3: Hematological (p>0.05*) Asthenia (p>0.05*) Gastrointestinal (p>0.05*)			[172]
2012	Caucasian (Poland)	171	I-IV	Platinum-based			0.89 (0.57-1.40)	CC	1.06 (0.71-1.60)	CC				[202]
2012	Asian (China)	355	IIIB-IV	CDDP/CBDCA DTX/GMZ/PXT/VNR	0.89 (0.40-1.97)	CC	p=0.003** Gemcitabine group		p>0.05**					[182]
2013	Caucasian (Italy)	98	IIIB-IV	CDDP-based	0.91 (0.30-2.75)	C	1.0 (0.59-1.71)	C	1.11 (0.65-1.89)	C				[154]
2014	Asian (China)	378	I-IV	CDDP DTX/GMZ/PTX/VNR	1.24 (0.78-1.98) CT 1.78 (0.81-3.85) TT	CC	0.93 (0.65-1.34) CT 0.94 (0.50-1.75) TT	CC	0.92 (0.65-1.30) CT 0.81 (0.43-1.49) TT	CC				[204]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine; VP16: etopósido.
*p-value for chi-square test; ** p-value for long rank test.
The blanks mean that the paper did not provide any information on this parameter.

1.10.2.1 TP53

The *TP53* tumor suppressor gene has a prominent role in carcinogenesis [208]. It has an anti-cancer function, inhibiting angiogenesis, inducing apoptosis of tumor cells and maintaining. genomic stability [208]. This gene is frequently somatically mutated in NSCLC (40-70%), which may modify p53 activity and induce resistance to platinum based chemotherapy [209].

In tumor cell lines with a wild type *TP53* gene, the GG genotype of a common *TP53* polymorphism located at codon 72, exon 4 (C→G, rs1042522, Pro72Arg), enhances the risk of death by promoting the mitochondrial localization of p53 [210]. When the p53 protein is mutated, the polymorphism has the opposite effect [211,212]. In NSCLC patients treated with platinum compounds, the p53 mutant protein with GG genotype has been described to abolish the function of p73, a related p53 protein, and therefore inhibit apoptosis [64]. The CC genotype has shown better response (OR=3.02; $CI_{95\%}$=1.77-5.18; CC vs GG) in 640 NSCLC patients treated with platinum compounds [64] while the GG variant has been associated with higher gastrointestinal toxicity (OR=0.24; $CI_{95\%}$=0.08-0.81 for GC/CC vs GG) (Table 7) [88].

1.10.2.2 MDM2

The MDM2 protein is a negative regulator of p53 that binds to the p53 protein inducing its ubiquitination and degradation [87]. Two polymorphisms in *MDM2* (C→T, rs1470383 and A→G, rs1690924) have been associated with gastrointestinal and hematological toxicity, but not with OS and PFS in 663 Chinese NSCLC patients (Table 7) [65]. Patients with the AA genotype for MDM2-rs1470383 showed lower hematological chemotherapy-related toxicity than those with the GG genotype (OR=4.10; $CI_{95\%}$=1.73-9.71 for GG vs AA) [65]. In contrast, the GG genotype for *MDM2* rs1690924 has been related to lower gastrointestinal toxicity (OR=2.32; $CI_{95\%}$=1.30-4.14 for AG vs AA) (Table 7) [65]. The relationship between these SNPs with response has not been evaluated.

An association has also been found between a SNP in the promoter region of *MDM2* (T→G, rs2279744) and clinical outcomes in NSCLC patients treated with cisplatin/carboplatin [88,89]. The GG genotype correlated with longer OS (HR=1.33; $CI_{95\%}$=1.03-1.72 for GT/TT vs GG) in 568 NSCLC patients [89] and lower hematological toxicity in 444 Chinese advanced NSCLC patients (OR=2.18; $CI_{95\%}$=1.12-4.25 for GT/TT vs GG) [88].

Table 7. Influence of TP53 and MDM2 polymorphisms on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival					
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	Type	OR (95% CI)	Ref. Cat.	
							TP53 rs1042522							
2010 2014	Asian (Japan) Asian (China)	640 444	III-IV IIIA-IV	Platinum-based CDDP/CBDCA DTX/GMZ/PTX/VNR	3.02 (1.77-5.18) p=0.937*	GG					Gastrointestinal	0.24 (0.08-0.81)	GG	[64] [88]
							MDM2 rs1470383							
2015	Asian (China)	663	IIIA-IV	Platinum-based			1.18 (0.96-1.44) AG 1.20 (0.72-2.00) GG	AA	1.08 (0.88-1.34) AG 1.13 (0.66-1.93) GG	AA		Gastrointestinal: 0.80 (0.42-1.51) AG 0.43 (0.06-3.31) GG Hematological: 1.02 (0.68-1.54) AG 4.10 (1.73-9.71) GG	AA	[65]
							MDM2 rs1690924							
2015	Asian (China)	663	IIIA-IV	Platinum-based			1.08 (0.89-1.31) AG 0.74 (0.51-1.06) GG	AA	0.92 (0.76-1.12) AG 0.76 (0.53-1.09) GG	AA	Gastrointestinal Hematological	Gastrointestinal: 2.32 (1.30-4.14) AG 0.58 (0.13-2.57) GG Hematological: 1.04 (0.72-1.52) AG 0.76 (0.37-1.55) GG	AA	[65]
							MDM2 rs229744							
2011 2014	Asian (China) Asian (China)	561 444	I-IV IIIA-IV	Platinum-based CDDP/CBDCA DTX/GMZ/PTX/VNR	 p=0.225*		1.33 (1.03-1.72)	GG			 Gastrointestinal	 2.18 (1.12-4.25)	 GG	[89] [88]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine.
*p-value for chi-square test.
The blanks mean that the paper did not provide any information on this parameter.

1.10.3 PI3K/PTEN/AKT pathway

The PTEN/PI3K/AKT signaling is involved in a great variety of cell processes, such as cell proliferation and survival, which may be altered by polymorphisms in the genes that integrate the pathway [213].

1.10.3.1 PI3K

The PI3K enzyme catalyzes the conversion of PIP2 into PIP3, activating the downstream AKT signaling [214]. Despite of this key role, none of the polymorphisms studied to date in the *PI3K* gene (A→G, rs7651265; C→G, rs7640662; T→C, rs7621329, A→C rs6443624; G→A, rs2699887) have shown a significant association with clinical outcomes in NSCLC patients treated with platinum based chemotherapy [90]. Only the AA genotype for rs2699887 has been correlated with increased grade 3-4 toxicity in 168 Caucasian stage IIIB/IV NSCLC patients (OR=3.86; $CI_{95\%}$=1.08-13.82 for GG vs AA) [90].

1.10.3.2 PTEN

PTEN, a well-known tumor suppressor gene, codifies a protein phosphatase that hydrolyzes PIP3 and inhibits the PTEN/PI3K/AKT pathway [215,216].Three polymorphisms in *PTEN* (T→A/C, rs2299939; T→G rs12569398, G→C rs12557281) have been evaluated, and only the AA genotype in rs2299939 has been associated with severe toxicity in 168 Caucasian stage IIIB/IV NSCLC patients (OR=0.44; $CI_{95\%}$=0.20-0.95 for AC/CC vs AA) [90]. The influence of these SNPs on ORR, PFS and OS has not been determined.

1.10.3.3 AKT

The AKT protein is the main downstream target of PI3K pathway. It triggers the phosphorylation of a series of intermediates effectors, promoting cell cycle progression, cell proliferation, transcription and cell migration [217-219].

The T allele in the *AKT* polymorphisms rs3803304 and rs2498804 have been associated with longer PFS in advanced NSCLC patients treated with cisplatin-based chemotherapy (HR=0.66; $CI_{95\%}$=0.45-0.97 for CT/TT vs CC and HR=0.52; $CI_{95\%}$=0.35-0.77 for GT/TT vs GG, respectively) [90]. In contrast, the GG genotype of AKT rs1130214 have been correlated with shorter PFS in 168 advanced NSCLC patients (HR=0.62; $CI_{95\%}$=0.42-0.91 for GT/TT vs GG) [90], while it was associated with better OS (HR=2.78; $CI_{95\%}$=1.11-6.99 for TT vs GG) and PFS (HR=1.48; $CI_{95\%}$=1.02-

2.15 for TT vs GG) in 310 early NSCLC patients receiving cisplatin-based adjuvant therapy [220]. No association with toxicity has been described in any polymorphism, and ORR has not been evaluated [90].

1.10.4 TGF-β pathway

The TGF-β pathway regulates tumorigenesis and tumor progression through its effects on cellular proliferation, survival, angiogenesis and invasion, via cross talk with SMAD transcriptional regulators [94].

The CT/TT genotypes for the SMAD3 polymorphisms rs6494633 and rs11632964 have been associated with better OS (HR=1.20; $CI_{95\%}$=1.01-1.43 for CC vs CT/TT and HR=1.52; $CI_{95\%}$=1.05-2.17 for CC vs CT/TT, respectively) in 598 Caucasian stage IIIA/IV NSCLC patients [91]. In contrast, polymorphisms in TGF-β receptor do not seem to be associated with response and survival to platinum based chemotherapy [91].

1.10.5 Cellular efflux transporters

Proteins involved in drug efflux are responsible of extruding drugs out of the cell [98,102,103] and polymorphisms in the corresponding genes have been associated with efficacy of platinum based chemotherapy in advanced NSCLC.

1.10.5.1 ABCB1

ABCB1 is the most extensively studied transmembrane cellular efflux transporter. It belongs to the ATP-binding cassette family, and pumps out the cell an enormous variety of drugs, including platinum compounds [98,102,103]. Polymorphisms in this gene lead to lower expression and activity of ABCB1 protein, increasing levels of drugs outside the cells [221]. Contradictory results have been obtained when analyzing the association of the silent polymorphism rs1045642, (C→T substitution at codon 1142, exon 26, position 3435, C3435T) [221], with ORR to platinum drugs in advanced NSCLC (Table 8). Although a meta-analysis, including 5 studies with a total of 379 Asian and Caucasian patients, reported higher ORR for the CC variant (OR=1.82; $CI_{95\%}$=1.17-2.85; I^2=0%; $P_{heterogeneity}$=0.77; CC vs CT/TT) [95]. The C-allele has also been associated with better OS (HR=0.77; $CI_{95\%}$=0.59-0.99 for C vs T allele) and PFS (HR=1.91; $CI_{95\%}$=1.13-3.22 for CT vs CC) in two studies including 160 and 94 advanced NSCLC patients, respectively [96,163]. A correlation with grade 3-4 gastrointestinal toxicity was also initially reported in 62 stage IIIB-IV NSCLC patients (p=0.03) [80], but in further studies failed to confirm it [97,158,163].

Table 8. Influence of ABCB1 polymorphisms on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival					
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	Type	OR (95% CI)	Ref. Cat.	
rs1045642														
2004	Caucasian (Spain)	62	IIIB-IV	CDDP DTX	1.26 (0.42-3.83)	T	p=0.62**		p=0.58**		Grade 3-4: Gastrointestinal (p=0.03*)			[80]
2008	Asian (China)	69	III-IV	CDDP VNR	2.90 (1.05-8.00)	T								[223]
2009	Asian (China)	54	IIIB-IV	CDDP DTX	p=0.123*									[222]
2010	Asian (China)	95	IIIB-IV	CDDP GMZ/PXT/VNR	1.60 (0.64-4.02)	T					Hematological (p=0.941*) Hepatotoxicity/ Nephrotoxicity (p=0.335*) Gastrointestinal (p=0.912*)			[158]
2010	Caucasian (Spain)	94	IIIB-IV	CDDP VNR	1.92 (0.76-4.85)	T	1.14 (0.64-2.04)	T	1.91 (1.13-3.22) CT 1.64 (0.85-3.18) TT	CC	Grade 3-4 (p>0.05*)			[163]
2011	Asian (China)	103	IIIB-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	1.62 (1.11-2.37)	CC	1.13 (0.80-1.59)	CC						[224]
2014	Asian (China)	161	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/PMX/VNR	0.48 (0.24-0.93)	CC	p>0.05**							[130]
2014	Caucasian (USA)	160	III-IV	CDDP/CBDCA DTX/PTX			0.77 (0.59-0.99)	C	0.62 (0.38-1.00)	C				[96]
2014	Caucasian (USA)	86	IIIB-IV	CDDP/CBDCA GMZ/PTX/VP16			p=0.04**		p>0.05**		Grade 3-4: Hematological/ Non-hematological (p>0.05*)			[97]
rs2032582														
2010	Asian (China)	95	IIIB-IV	CDDP GMX/PTX/VNR	1.88 (1.01-3.50) T 0.47 (0.18-1.25) A 1.38 (0.78-2.45) A/T	G					Hematological (p=0.880*) Hepatotoxicity/ Nephrotoxicity (p=0.995*) Gastrointestinal (p=0.030*)			[158]
rs1128503														
2014	Caucasian (USA)	160	III-IV	CDDP/CBDCA DTX/PTX			1.53 (1.11-2.09)	T	2.04 (1.11-3.77)	T				[96]

Table 8. (Continued).

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
							rs1128503							
2014	Caucasian (USA)	86	IIIB-IV	CDDP/CBDCA GMZ/PTX/VP16			p>0.05**		0.54 (-1.11--0.12)	CC	Grade 3-4: Hematological/ Non-hematological (p>0.05*)			[97]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine; VP16: etopósido.
*p-value for chi-square test; ** p-value for long rank test.
The blanks mean that the paper did not provide any information on this parameter.

A meta-analysis in Asian population, with a total of 3 studies and 96 patients, has reported better ORR for the GG variant (OR=2.61; $CI_{95\%}$=1.44-4.74; I^2=0%; $P_{heterogeneity}$=0.51; GG vs GT/GA/TT/AA) of a polymorphism in linkage disequilibrium with C3435T SNP (Ala893Ser, rs2032582), which modifies the expression of the ABCB1 protein [95,158]. In contrast, no significant association with toxicity was reported and PFS and OS were not evaluated (Table 8).

Finally, contradictory reports have also been published regarding the silent SNP rs1128503 (Gly412Gly) The C-allele was associated with longer OS (HR=1.53; $CI_{95\%}$=1.11-2.09 for C vs T allele) and PFS (HR=2.04; $CI_{95\%}$=1.11-3.77 for C vs T allele) in 160 stage III-IV NSCLC patients [96] while an analysis of 86 stage IIIB-IV NSCLC patients reported shorter PFS for CC genotype (HR=0.541; $CI_{95\%}$=-1.11- -0.12 for CT/TT vs CC) in [97]. No association with toxicity was found and ORR was not studied (Table 8).

1.10.6 Gluthathione metabolic pathway

The glutathione metabolic pathway mediates platinum detoxification through glutathione conjugation [225]. Glutathione S-transferase (GSTPs) enzymes catalyze this process. The major subclasses of GSTs are GSTM1, GSTP1, GSTT1, and GSTA1 [226]; being GSTP1 the most abundant isoform in the lung and the enzyme mainly involved in platinum detoxification in NSCLC patients [227,228]. A single nucleotide substitution at exon 5 (G→A, rs1695, Ile105Val) has been demonstrated to alter GSTP1 activity [229], with the Val variant being more active against cisplatin and carboplatin compounds [108,110,230]. Several studies have reported that the AG/GG genotypes in rs1695 are associated with better ORR, PFS and OS and that the GG genotype correlates with less severe hematological toxicity (p=0.02), but higher neurotoxicity (p=0.01) (Table 9).

1.10.7 Folate metabolism

Folate metabolisms is involved in various intracellular processes such as DNA methylation, cell proliferation and synthesis of nucleic and amino acids [231]. Genetic alterations in these genes may disrupt folate metabolism function, inducing DNA hypomethylation and consequently activating proto-oncogenes [232-236]. Thus, polymorphisms in folate metabolism genes may promote tumor development and modify sensitivity of tumor cells to platinum compounds.

Table 9. Influence of GSTP1 polymorphisms on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival		Type	OR (95% CI)	Ref. Cat.	
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.				
rs1695														
2006	Caucasian (United Kingdom)	108	III-IV	Platinum-based	0.96 (0.39-2.36)	AA	0.83 (0.44-1.58) AG 1.14 (0.52-2.50) GG	AA			Hematological (p=0.02*)			[109]
2010	Asian (China)	113	IIIA-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	3.030 (1.28-7.19)	AA								[106]
2011	Asian (China)	111	IV	CDDP/CBDCA DTX/GMZ/PMX/VNR	3.961 (1.53-10.25)	G			1.852 (1.19-2.89)	G				[108]
2012	Caucasian (Netherlands)	137	IIIB-IV	CDDP GMZ	0.47 (0.22-1.02)	AA	1.34 (0.89-2.02) AG 1.32 (0.72-2.42) GG	AA	1.38 (0.92-1.97) AG 0.85 (0.48-1.49) GG	AA	Neurotoxicity (p=0.01*)			[110]
2014	Asian (China)	91	IIIB-IV	CDDP DTX/GMZ/VNR	6.931 (0.12-0.74)	AA			p<0.01**					[105]
2015	Asian (China)	97	IIIB-IV	CDDP DTX/GMZ/VNR/PTX	4.302 (1.19-15.52)	AA			1.639 (1.01-2.65)	AA	Gastrointestinal (p>0.05*) Hematological (p>0.05*) Skin (p>0.05*)			[75]
2015	Asian (China)	325	IIIB-IV	CDDP DTX/GMZ/PTX/VNR	2.31 (1.35-3.95) AG 5.68 (1.61-30.46) GG	AA	0.75 (0.46-1.22) AG 0.36 (0.11-0.98) GG	AA						[104]
2015	Asian (China)	282	IIIB-IV	Platinum-based	1.59 (0.87-2.89) AG 2.18 (1.16-4.12) GG	AA	0.58 (0.31-1.07) AG 0.48 (0.25-0.93) GG	AA						[107]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine.
*p-value for chi-square test; ** p-value for long rank test.
The blanks mean that the paper did not provide any information on this parameter.

1.10.7.1 MTHFR

MTHFR is a crucial enzyme in the folate metabolism. Several polymorphisms in this gene lead to a production of an enzyme with decreased activity and their effects have been linked with DNA hypomethylation, therefore influencing platinum therapy outcomes [232-236].

A C→T transition at codon 222, exon 4 (Val→Ala, rs1801133) has been found to be associated with response, survival and toxicity of platinum compounds drugs. A meta-analysis compiling data from 3 studies and 147 patients, both in Asian and Caucasian populations, has shown better response in individuals with TT genotype (OR=1.72; $CI_{95\%}$=1.01-2.93; I^2=16%; $P_{heterogeneity}$=0.31; TT vs CT/CC) [66]. The TT variant has also been associated with higher OS (p=0.026) and PFS (p=0.012) in 208 Italian stage IIIB/IV NSCLC patients (Table 10) [70]. Likewise, the genotype CC has been correlated with higher hematological toxicity in 1004 Chinese stage III/IV NSCLC patients (OR=0.40; $CI_{95\%}$=0.19-0.85 for CT vs CC) (Table 10) [79]

Another polymorphism in MTHFR, which results in an A→C substitution at codon 429, exon 7 (Glu→Ala, rs1801131), has also been associated to platinum based chemotherapy outcomes [79]. Carriers of AA genotype presented lower ORR (OR=1.52; $CI_{95\%}$=1.04-2.23 for AC vs AA), PFS (p=0.03) and higher gastrointestinal toxicity in a study conducted in 1.004 Chinese stage III/IV NSCLC patients (OR=0.40; $CI_{95\%}$=0.22-0.75 for AC vs AA) (Table 10) [79].

1.10.7.2 MTR

MTR is an important vitamin B12-dependent enzyme, which catalyzes the final step in folate metabolism [111].

A transition from A to G at position 2756, exon 26 (rs1805087) causes an amino acid change of Asp to Gly, which has been reported to modify enzyme activity and alter DNA methylation processes [237,238]. However, no significant was found with ORR (OR=0.66; $CI_{95\%}$=0.23-1.89 for AG/GG vs AA) and OS (HR=0.99; $CI_{95\%}$=0.23-1.89 for AG/GG vs AA) in two studies including 465 I-IV and 101 IIIB/IV NSCLC patients treated with platinum based chemotherapy (Table 10) [67,68].

1.10.7.3 SLC19A1

The reduced folate carrier SLC19A1 is responsible for the transport of folate drugs into the cell, such as pemetrexed, a drug that is usually given in combination with carboplatin/cisplatin [239]. Polymorphisms in this gene may modify the passage of this drug into the tumor cell.

A single nucleotide transition (G→A, rs1051266) at exon 2, which induces an Arg→His replacement in codon 27, has demonstrated to alter pemetrexed-platinum combination efficacy [71]. The GG genotype has been associated with better OS (HR=1.76; $CI_{95\%}$=1.11-2.78 for AG/AA vs GG) in 136 lung cancer patients (Table 10) [71]. However, no significant associations have been reported for ORR, PFS and toxicity (Table 10).

1.10.8 Cytokine signaling

Innate immune cells induce inflammation as a physiological process aimed to combat infection. However, chronic inflammation may cause persistent tissue damage and cellular proliferation leading to metaplasia and dysplasia [240,241]. Consequently, there are prominent connections between chronic inflammation, infection and early stage of neoplastic development. Clinical and epidemiological studies have reported that 20% of tumors are associated to chronic infection, 30% associated to tobacco smoking and pollutant inhalation and 35% are related to nutrition [242].

Growth, differentiation, and activation of immune cells are mediated by a family of cytokines referred as interleukins (ILs) [243]. During tumor development, ILs act as autocrine and paracrine growth factors, inhibiting apoptosis at the site of inflammation [116]. *IL* polymorphisms have recently been associated with OS in stage IIIB-IV NSCLC patients receiving platinum based chemotherapy. In the case of the *IL1B* rs1143634 polymorphism, the T-allele has been correlated with better OS (HR=0.78; 95%CI=0.63-0.98 for CT/TT vs CC) and PFS (HR=0.73; $CI_{95\%}$=0.57-0.93 for CT/TT vs CC) in 651 Caucasian patients [78]. Likewise, the CC genotype for *IL6* rs1800795 was found to be associated with better OS (HR=1.68; $CI_{95\%}$=1.08-2.63 for CG/GG vs CC) in 414 Portuguese stage I-IV NSCLC patients [77]. Finally, the CT/TT genotypes for *IL12A* (rs662959) (HR=1.41; $CI_{95\%}$=1.08-1.83 for CT/TT vs CC), AC/CC for *IL13* (rs1881457) (HR=1.29; $CI_{95\%}$=1.00-1.66 for AC/CC vs AA) and GG for *IL16* (rs7170924) (HR=0.65; $CI_{95\%}$=0.50-0.83 for GT/TT vs GG) have been associated with lower PFS in 651 Caucasian stage I-IV NSCLC patients [78].

Table 10. Influence of folate metabolisms polymorphisms on clinical outcomes and toxicity in NSCLC patients.

Year	Population	N	Stage	Chemotherapy agents	Outcomes						Side Effects			Ref.
					Overall Response Rate		Overall survival		Progression-free survival					
					OR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	HR (95% CI)	Ref. Cat.	Type	OR (95% CI)	Ref. Cat.	
							MTHFR rs1801133							
2012	Caucasian (Italy)	208	IIIB-IV	CBDCA+PMX	p=0.081*		p=0.026**		p=0.012**					[70]
2014	Asian (China)	1004	III-IV	CDDP/CBDCA+DTX/GMZ/ PTX/VNR	1.06 (0.74-1.53) CT 1.05 (0.65-1.69) TT	CC	0.98 (0.83-1.16) CT 1.02 (0.82-1.28) CC	CC	0.90 (0.74-1.08) CT 0.84 (0.65-1.09) CC	CC	Hematological	0.40 (0.19-0.85) CT 0.40 (0.13-1.21) TT	CC	[79]
							MTHFR rs1801131							
2014	Asian (China)	1004	III-IV	CDDP/CBDCA DTX/GMZ/PTX/VNR	1.52 (1.04-2.23) AC 0.70 (0.28-1.73) CC	AA	1.02 (0.87-1.20) AC 1.13 (0.69-1.84) AA	AA	0.97 (0.80-1.17) AC 0.99 (0.55-1.78) CC	AA	Gastrointestinal	0.40 (0.22-0.75) AC 2.01 (0.64-6.35) CC	AA	[79]
							MTR rs1805087							
2007	Caucasian (United Kingdom)	465	I-IV	Platinum-based			0.99 (0.23-1.89)	AA						[68]
2010	Asian (China)	101	IIIB-IV	CDDP/CBDCA DTX/GMZ/VNR	0.66 (0.23-1.89)	AA								[67]
							SLC19A1 rs1051266							
2009	Caucasian (Australia)	127	IIIB-IV	CBDCA PMX					p=0.8**					[72]
2010	Caucasian (USA)	54	IIIB-IV	GMZ PMX			p=0.03**				Dyspnea (p>0.05*) Fatigue (p>0.05*) Hematological (p>0.05*) Hepatotoxicity (p>0.05*)			[73]
2012	Caucasian (Italy)	208	IIIB-IV	CBDCA PMX	p=0.148*		p=0.634**		p=0.746**					[70]
2013	Asian (China)	45	IIIB-IV	CDDP PMX	p=0.701*		p=0.022**		p=0.137**					[69]
2014	Caucasian (United Kingdom)	94	IIIB-IV	CDDP/CBDCA PMX			1.76 (1.11-2.78)	GG	p>0.05**		Hematological (p>0.05*) Gastrointestinal (p>0.05*)			[71]

N: number of patients; OR: odds ratio; HR: hazard ratio; 95% CI: 95% confidence interval; Ref. Cat.: reference category; Ref.: reference; CDDP: cisplatin; CBDCA: carboplatin; GMZ: gemcitabine; DTX: docetaxel; PMX: pemetrexed; PTX: paclitaxel; VNR: vinorelbine.
*p-value for chi-square test; ** p-value for long rank test.
The blanks mean that the paper did not provide any information on this parameter.

HYPOTHESIS AND OBJECTIVES

2 HYPOTHESIS

"Genetic markers predict response and toxicity of treatment with platinum-based chemotherapy in patients diagnosed with NSCLC".

3 OBJECTIVES

3.1 Main objective

To evaluate the influence of genetic markers as prognostic and predictive markers in patients diagnosed with NSCLC and treated with platinum-based chemotherapy.

3.2 Specific objectives

- To determine the presence of gene polymorphisms involved in response and toxicity in patients with NSCLC treated with platinum-based chemotherapy.
- To evaluate the response to platinum-based chemotherapy treatment in patients diagnosed with NSCLC.
- To evaluate the overall survival and progression-free survival in patients diagnosed with NSCLC treated with platinum-based chemotherapy.
- To evaluate the toxicity profile in patients diagnosed with NSCLC treated with platinum-based chemotherapy.
- To evaluate the influence of genetic markers in effectiveness of the treatment.
- To evaluate the influence of genetic markers in toxicity of the treatment.

MATERIAL AND METHODS

4 MATERIAL AND METHODS

4.1 Study design

A retrospective-prospective cohorts study was conducted.

4.2 Ethics statement

This study was performed under the approval of the Complejo Hospitalario Universitario de Granada (CHUG) Ethics and Research Committee and in accordance with the declaration of Helsinki (Annex 1). A written informed consent form was signed by the patients for blood sample collection and genotyping analysis (Annex 2 and 3). The identification of samples was based on non-patient codes.

4.3 Target population

Patients with NSCLC treated with platinum-based chemotherapy.

4.4 Geographic and temporal scope

The study included patients diagnosed histologically or cytologically as NSCLC (stages I-IV) between December 2003 and November 2015 and treated with cisplatin or carboplatin in combination with a third-generation drug (gemcitabine, paclitaxel, pemetrexed and vinorelbine) at the CHUG.

4.5 Inclusion criteria:

- Age ≥18 years old.
- Patients histologically or cytologically diagnosed as NSCLC (stages I-IV).
- Patients treated intravenously with cisplatin or carboplatin in combination with a third-generation drug (gemcitabine, paclitaxel, pemetrexed and vinorelbine).
- Voluntary acceptance to participate in the study and a signed written consent form according to the to law 14/2007 on Biomedical Research Acceptance.
- Normal results of hematological function (hemoglobin>9g/dl, neutrophil count>1500/mm3, and platelet count>100000/mm3), liver function (bilirubin< 1.5 times

the normal upper limit, aspartate aminotransferase and alanine aminotransferase<2.5 times the normal upper limit) and renal function (creatine clearance rate>50 ml/s).

- Measurable disease by chest computed tomography (CT) scan.
- Follow-up period of at least 6 months after NSCLC diagnosis.

4.6 Exclusion criteria

- Hypersensitivity to the study drugs, to any of the excipients, or premedication required for chemotherapy regimen.
- Pregnant women, lactating or those who wanted to become pregnant during the study period.
- Patients treated in other hospital.

4.7 Variables

4.7.1 Dependent variables

4.7.1.1 Survival

Survival was evaluated through OS and PFS, which were measured as follows:

OS as time from cancer diagnosis until final follow-up or death

PFS as the time from initiation of treatment to relapse, death or last known follow-up.

Mortality related data were collected from clinical records and the population-based Cancer Registry of Granada.

4.7.1.2 Response

Platinum-based chemotherapy response was evaluated based on the Response Evaluation Criteria in Solid Tumors (RECIST) guideline (Version 1.1) [244], and was defined as follows:

Complete Response (CR): Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.

Partial Response (PR): At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters.

Progressive Disease (PD): At least a 20% increase in the sum of diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progression).

Stable Disease (SD): Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study.

Patients classified as CR+PR were catalogued as responders to treatment and SD+PD as non-responders.

4.7.1.3 Toxicity

Toxicity was evaluated according to the Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0 [245]. Based on the severity of adverse events, it was classified into grade 0-2 and 3-4. General toxicity was defined as 3-4 when at least one adverse event was present in grade 3-4, and 0-2 otherwise. The occurrence of more than one and more than two adverse events was also analyzed.

The types of adverse events collected were: asthenia, gastrointestinal toxicity, hematological toxicity, infection, neurotoxicity and nephrotoxicity.

4.7.2 Independent variables

4.7.2.1 Sociodemographic variables

Sociodemographic data including gender, family history of cancer, previous non-lung cancer, previous lung disease, smoking status and age at diagnosis was collected from clinical records.

4.7.2.2 Clinical variables

Clinical data were also collected from clinical records and comprised tumor histology and stage, chemotherapy agents, surgery, radiotherapy and EGFR status. The tumor staging was performed based on AJCC cancer staging manual [18].

All patients received intravenously cisplatin or carboplatin in combination with a third-generation drug (gemcitabine, paclitaxel, pemetrexed and vinorelbine) according to the National Comprehensive Cancer Network version 4.2016 guidelines [19]. Hematology and biochemistry analyses were done at the end of each cycle.

The status of EGFR was analyzed by cobas® EGFR Mutation Test [246].

4.7.2.3 Genetic variables

4.7.2.3.1 DNA isolation

Blood samples (3 ml) were collected in BD Vacutainer® K3E Plus Blood Collection Tubes, and saliva samples were collected in 50 ml BD Falcon™ conical tubes (BD, Plymouth, UK). DNA was extracted from white blood or epithelial cells using the QiaAmp DNA Mini Kit (Qiagen GmBH, Hilden, Germany) and stored at -40ºC. DNA quality was checked by 1% agarose gel electrophoresis.

4.7.2.3.2 Detection of gene polymorphisms

4.7.2.3.3 Conventional PCR

SLC19A1 Arg27His (rs1051266) and ABCB1 Ala893Ser/Thr (rs2032582) gene amplification were performed by polymerase chain reaction (PCR) on an Applied Biosystems 2720 Thermal Cycler (Life Technologies Corporation, Carlsbad, California, USA) in separated tubes for each polymorphism. The final volume was 25 µL for PCR plus restriction fragment length polymorphism (PCR-RFLP) and 12.5 µL for direct sequencing, containing 50 ng of DNA. PCR products were checked on 2% agarose gels with ethidium bromide and visualized under UV light.

PCR reaction mix for ABCB1 Ala893Ser/Thr (rs2032582) gene polymorphisms contained 400 nM of each primer, 1.5 mM of MgCl2, 0.2 mM of dNTPs (Qiagen GmBH, Hilden, Germany), 0.75 U AmpliTaq Gold® DNA Polymerase (Applied Biosystems, Foster City, CA, USA) and 8% DMSO in 1x PCR Gold Buffer. For SLC19A1 Arg27His (rs1051266) PCR reaction mix was composed by 400 nM of each primer, 4 mM of MgCl2, 1 mM of dNTPs (Qiagen GmBH, Hilden, Germany), 0.75 U AmpliTaq Gold® DNA Polymerase (Applied Biosystems, Foster City, CA, USA) and 8% DMSO in 1x PCR Gold Buffer.

The amplification program was conducted using a touchdown PCR program with an initial denaturation step of 15 min at 94°C, followed by 35 cycles of 30 seconds at 94°C, 60 seconds at

65-55ºC (annealing temperature decreased by 0.5°C per cycle), 30 seconds at 72°C and a final elongation step of 7 min at 72°C.

4.7.2.3.4 Direct sequencing

ABCB1 Ala893Ser/Thr (rs2032582) gene polymorphism was determined by PCR and direct sequencing. PCR products were purified using the illustra Exostar Kit for Enzymatic PCR and Sequencing Clean-up (GE Healthcare Life Sciences, Chicago, Illinois, USA). Sequencing reaction was performed using 1 µl of Big Dye Terminator v1.1 cycle sequencing reactive (Applied Biosystems, Foster City, CA, USA), 2 µl of the purified PCR product and 119 nM of the reverse PCR primer. Sequencing products were then purified by ethanol precipitation and analyzed on an ABI PRISM 3130xl Genetic Analyzer (Applied Biosystems, Foster City, CA, USA).

4.7.2.3.5 PCR-RFLP

SLC19A1 Arg27His (rs1051266) gene polymorphism was determined by PCR plus restriction fragment length polymorphism (PCR-RFLP) with HhaI restriction enzyme. Amplified DNA was digested overnight with the correspondent restriction enzyme at 37ºC, according to the manufacturer's specifications (New England Biolabs® Inc, Massachusetts, USA). The digestion fragments were analysed on 3.5% agarose gel with ethidium bromide and visualized under ultraviolet light. The results of PCR-RFLP were confirmed by direct sequencing in 10% of samples.

4.7.2.4 Real Time PCR

ERCC1 C118T (rs11615), ERCC1 C8092A (rs3212986), ERCC2 Lys751Gln (rs13181), ERCC2 Asp312Asn (rs1799793), ERCC2 (rs50872), ERCC2 (rs238416), ERCC5 His46His (rs1047768), ERCC5 Asp1104His (rs17655), XRCC1 Arg194Trp (rs1799782), XRCC1 Gln399Arg (rs25487), MDM2 (rs1470383), MDM2 (rs1690924), ABCB1 C3435T (rs1045642), ABCB1 C1236T (rs1128503), MTHFR A1298C (rs1801131), MTHFR C677T (rs1801133), MTR (rs1805087), IL1B (rs12621220), IL1B (rs1143623), IL1B (rs16944), IL1B (rs1143627), IL6 (rs1800795), IL16 (rs7170924) gene polymorphisms were analyzed by Real-Time PCR via allelic discrimination plots using TaqMan® real time PCR primers and probes on the Applied Biosystems StepOne Real Time PCR System (Applied Biosystems, Foster City, CA, USA). The PCR cycling conditions were as follows: initial denaturation at 95°C for 10 minutes, followed by 40 cycles of denaturation at 95°C for 15 seconds and annealing/extension at 60°C for 60 seconds. Fluorescence signal intensity due to degradation of the TaqMan® probe was quantified during the annealing phase of each PCR cycle. Genotype analysis of the DNA samples was performed in duplicate. The

presence of wild-type and variant alleles was defined by comparing the relative end-point fluorescence created by the degradation of each fluorescently labelled TaqMan® probe (VIC/FAM). Table 11 summarizes the characteristics of each SNP analyzed and the methodology used.

Table 11. Characteristics of each SNP analyzed and the methodology used.

Gene Polymorphism	Primers	Technique	Enzyme	Fragments (base pairs)
ERCC1 C118T (rs11615)		TaqMan		
ERCC1 C8092A (rs3212986)		TaqMan		
ERCC2 Lys751Gln (rs13181)		TaqMan		
ERCC2 Asp312Asn (rs1799793)		TaqMan		
ERCC2 (rs50872)		TaqMan		
ERCC2 (rs238416)		TaqMan		
ERCC5 His46His (rs1047768)		TaqMan		
ERCC5 Asp1104His (rs17655)		TaqMan		
XRCC1 Arg194Trp (rs1799782)		TaqMan		
XRCC1 Gln399Arg (rs25487)		TaqMan		
MDM2 (rs1470383)		TaqMan		
MDM2 (rs1690924)		TaqMan		
MDM2 (rs1690924)		TaqMan		
ABCB1 C3435T (rs1045642)		TaqMan		
ABCB1 C1236T (rs1128503)		TaqMan		
ABCB1 Ala893Ser/Thr (rs2032582)	Forward primer: GAATATAGCAAATCTTGGGACAGGAATAA Reverse primer: AATGGCCTGAAAACTGAAAAAGTCTGT	Sequencing		
MTHFR A1298C (rs1801131)		TaqMan		
MTHFR C677T (rs1801133)		TaqMan		
MTR (rs1805087)		TaqMan		
SLC19A1 Arg27His (rs1051266)	Forward primer: CTGCAGACCATCTTCCAAGGTG Reverse primer: GTAGGGGGTGATGAAGCTCTC	PCR-RFLP	HhaI	GG: 174+74 AG:248+174+74 AA: 248
IL1B (rs12621220)		TaqMan		
IL1B (rs1143623)		TaqMan		
IL1B (rs16944)		TaqMan		
IL1B (rs1143627)		TaqMan		
IL6 (rs1800795)		TaqMan		
IL16 (rs7170924)		TaqMan		

4.7.3 Statistical Analysis

Deviation from Hardy Weinberg equilibrium and pairwise linkage disequilibrium for each polymorphism were calculated using the free, open-source whole genome association analysis toolset PLINK [247].

Quantitative data were estimated as the mean (± standard deviation) for normally-distributed variables or medians and percentiles (25 and 75) for non-normal distributed variables. The Shapiro-Wilks test was used to assess normality.

The bivariate association between response with demographic, clinical and genetic variables was tested using the Pearson's chi-square or Fisher's exact test, and evaluated by relative risk and their corresponding 95% confidence intervals. The influence of SNPs on response and toxicity were analyzed using a logistic regression model (backward stepwise method).

The Kaplan-Meier method and the log-rank test were employed to assess associations between survival with demographic, clinical and genetic variables. Multivariate Cox proportional hazard regression model (backward stepwise method) was used to estimate the adjusted hazards ratio and 95% confidence interval for potential prognostic factors for survival.

All tests were two-sided and a probability of 0.05 or smaller was considered statistically significant. Data analysis was performed using R 3.0.1 [248].

RESULTS

5 RESULTS

5.1 Patients characteristics

A total of 141 NSCLC patients were recruited in the study. The baseline characteristics are listed in Table 12. The median age was 61 [52, 67] years, and 104 were males (104/141; 73.76%); 70.5% presented advanced stage (IIIB-IV) (98/139). All patients were treated with platinum-based chemotherapy in addition to one third-generation chemotherapy drug, such as gemcitabine (21/141; 14.89%), paclitaxel (33/141; 23.40%), pemetrexed (37/141; 26.24%) or vinorelbine (50/141; 35.46%).

Response to chemotherapy (CR + PR) was shown in 98 patients (98/141; 70.5%). Regarding treatment options, response to chemotherapy was higher in those patients who received adjuvant chemotherapy (93.8%; 15/16), whereas response to chemotherapy given as palliative treatment was 50.8% (31/61). During follow-up, 75 death events were documented. Median OS and PFS were 32.2 [27.0, 52.2] and 14.3 [10.2, 18.4] months for all patients, respectively. However, for patients with advanced stage median OS was 25.8 [21.1-32.2] months and median PFS 10.2 [8.37-5.0] months. During follow-up, grade 3-4 toxicity profile presented was: 29.79% (42/141) general toxicity, 2.13% (3/141) asthenia, 2.13% (3/141) gastrointestinal toxicity, 24.11% (34/141) hematological toxicity, 3.55% (5/141) infection, 2.13% (3/141) nephrotoxicity and 0.71% (1/141) neurotoxicity.

5.2 Influence of clinico-pathologic characteristics on clinical outcomes of platinum-based chemotherapy

5.2.1 Response

Response was better in squamous cell carcinoma (p=0.0342; RR=1.30; $CI_{95\%}$=1.02-1.66; Table 13), I, II and IIIA stage (p=0.0053; RR=1.41; $CI_{95\%}$=1.11-1.79; Table 13), and tumor resection (p=0.0004; RR=1.56; $CI_{95\%}$=1.22-1.99; Table 13).

Table 12. Clinico-pathologic characteristics of the 141 NSCLC patients treated with platinum based chemotherapy.

	n	%
Gender		
Female	37	25.24
Male	104	73.76
Family history of cancer		
Yes	85	60.28
No	56	39.72
Previous non-lung cancer		
Yes	23	16.31
No	118	83.69
Previous lung disease		
Yes	34	24.11
No	107	75.89
Smoking status		
Current-Smokers	72	51.06
Former-smokers	52	36.88
Non-smokers	17	12.06
Age at NSCLC diagnosis	61 [52, 67]	
≤60	68	48.23
>60	73	51.77
Histology		
Adenocarcinoma	87	61.70
Squamous cell carcinoma	51	36.17
Unknown	3	2.13
Tumor stage		
I, II or IIIA	41	29.5
Neoadjuvant Chemotherapy	20	48.78
Adjuvant Chemotherapy	16	39.02
Chemoradiotherapy	5	12.20
IIIB or IV	98	70.5
Chemoradiotherapy	37	37.76
Palliative	61	62.24
Chemotherapy agents		
Gemcitabine	21	14.89
Paclitaxel	33	23.40
Pemetrexed	37	26.24
Vinorelbine	50	35.46
Surgery		
Yes	33	23.4
No	108	76.6
Radiotherapy		
Yes	45	31.91
No	96	68.09
EGFR status		
Wild-type	72	51.06
Mutated	16	11.35
Unknown	53	37.59
Response		
CR	38	27.14
PR	60	42.86
SD	9	6.43
PD	33	23.57
Response/No response (divided by stage)		
I, II or IIIA		
Neoadjuvant Chemotherapy	18/2	90.0/10.0
Adjuvant Chemotherapy	15/1	93.8/6.2
Chemoradiotherapy	3/2	60.0/40.0
IIIB or IV		
Chemoradiotherapy	30/7	81.1/18.9
Palliative	31/30	50.8/49.2
Survival		
PFS	14.3 [10.2-18.4]	
OS	32.2 [27.0-52.2]	
Toxicity		
Grade 0-2	99	70.21
Grade 3-4	42	29.79

Qualitative variables: number (percentage)
Quantitative variables:
Normal distribution: mean ± standard deviation.
Non-normal distribution: P_{50} [P_{25}, P_{75}].
CR: complete response; PR: partial response; SD: stable disease; PD: progressive disease

Table 13. Association of clinical characteristics with response.

Characteristic	N	Response		χ^2	p-value	Reference Category	RR	95% CI
		PR+CR N (%)	SD+PD N(%)					
Gender								
Female	37	28 (75.7)	9 (24.3)	0.4478	0.5034			
Male	103	70 (68.0)	33 (32.0)					
Family history								
Yes	85	59 (69.4)	26 (30.6)	0.00	1.00			
No	55	39 (70.9)	16 (29.1)					
Personal history of cancer								
Yes	23	18 (78.3)	5 (21.7)	0.4856	0.4859			
No	117	80 (68.4)	37 (31.6)					
Previous lung disease								
Yes	34	27 (79.4)	7 (20.6)	1.3485	0.2455			
No	106	71 (67.0)	35 (33.0)					
Smoking status								
Current-Smokers	72	55 (76.4)	17 (23.6)	2.9046	0.2340			
Former-smokers	51	32 (62.7)	19 (37.3)					
Non-smokers	17	11 (64.7)	6 (35.3)					
Age at NSCLC diagnosis								
≤60	68	44 (64.7)	24 (35.3)	1.3086	0.2527			
>60	72	54 (75.0)	18 (25.0)					
Histology								
Adenocarcinoma	87	55 (63.2)	32 (36.8)	4.4829	0.0342	Squamous cell carcinoma	1.30	1.02-1.66
Squamous cell carcinoma	50	41 (82.0)	9 (18.0)					
Tumor stage								
I, II and IIIA	41	36 (87.8)	5 (12.2)	7.7851	0.0053	I, II and IIIA	1.41	1.11-1.79
IIIB and IV	98	61 (62.2)	37 (37.8)					
Chemotherapy agents								
Gemcitabine	20	11 (55.0)	9 (45.0)	6.8378	0.0773			
Paclitaxel	33	26 (78.8)	7 (21.2)					
Pemetrexed	37	22 (59.5)	15 (40.5)					
Vinorelbine	50	39 (78.0)	11 (22.0)					
Surgery								
Yes	32	31 (96.9)	1 (3.1)	12.656	0.0004	Yes	1.56	1.22-1.99
No	108	67 (62.0)	41 (38.0)					
Radiotherapy								
Yes	45	35 (77.8)	10 (22.2)	1.4035	0.2361			
No	95	63 (66.3)	32 (33.7)					
EGFR status								
Wild type	71	46 (64.8)	25 (35.2)	1.6244	0.2025			
Mutated	16	7 (43.8)	9 (56.2)					

N: number of patients; PR+CR: partial response + complete response; SD+PD: stable disease + progressive disease; RR: relative risk; 95% CI: 95% confidence interval.

5.2.2 Survival

The clinical factors associated with OS were: female ($p_{log\text{-}rank}$=0.0082; 85.4 vs 27.0 months; Table 14; Figure 3), squamous cell carcinoma ($p_{log\text{-}rank}$=0.0021; 59.4 vs 26.1 months; Table 14; Figure 4), I, II and IIIA stage ($p_{log\text{-}rank}$=<0.001; 85.4 vs 25.8 months; Table 14; Figure 5), paclitaxel chemotherapy agent ($p_{log\text{-}rank}$=<0.001; Table 14; Figure 6) and tumor resection ($p_{log\text{-}rank}$ =<0.001; 114.0 vs 26.1 months; Table 14; Figure 7).

Similarly, median PFS was better in female ($p_{log\text{-}rank}$ =0.0418; 19.6 vs 11.0 months; Table 15; Figure 8), squamous cell carcinoma ($p_{log\text{-}rank}$=0.0034; 27.3 vs 11.4 months; Table 15; Figure 9), I, II and IIIA stage ($p_{log\text{-}rank}$=<0.001; 44.0 vs 10.2 months; Table 15; Figure 10), paclitaxel as chemotherapy agent ($p_{log\text{-}rank}$=<0.001; Table 15; Figure 11), tumor resection ($p_{log\text{-}rank}$=<0.001; 82.2 vs 10.0 months; Table 15; Figure 12). A trend towards better PFS was showed in those patients who received concomitant or concurrent radiotherapy but it was not statistically significant ($p_{log\text{-}rank}$=0.0566; 23.2 vs 11.2 months; Table 15; Figure 13).

Table 14. Association of clinical characteristics with overall survival.

Characteristic	OS								
	N	Events	MST (mo)	95% CI	Log-Rank p-value	Reference Category	Univariate Cox Model		
							HR	95% CI	p-value
Gender									
Female	37	16	85.4	47.4-NR	0.0082	Female	2.10	1.20-3.67	0.0096
Male	104	60	27.0	24.2-39.1					
Family history									
Yes	85	48	31.1	25.8-60.7	0.8580				
No	56	28	39.1	26.1-73.9					
Personal history of cancer									
Yes	23	12	48.4	31.1-NR	0.2710				
No	118	64	30.7	25.8-52.2					
Previous lung disease									
Yes	34	14	39.1	24.5-NR	0.3180				
No	107	62	30.7	25.8-52.2					
Smoking status									
Current-Smokers	72	38	34.1	27.7-60.7					
Former-smokers	52	26	25.4	21.1-NR	0.8510				
Non-smokers	17	12	48.4	26.9-NR					
Age at NSCLC diagnosis									
≤60	68	39	27.9	23.1-60.7	0.6100				
>60	73	37	39.1	31.1-64.7					
Histology									
Adenocarcinoma	87	56	26.1	23.1-32.4	0.0021	Squamous cell carcinoma	2.23	1.32-3.76	0.0027
Squamous cell carcinoma	51	19	59.4	41.8-NR					
Tumor stage									
I, II and IIIA	41	16	85.4	64.7-NR	<0.001	I, II and IIIA	3.21	1.82-5.66	<0.001
IIIB and IV	98	59	25.8	21.1-32.2					
Chemotherapy agents									
Gemcitabine	21	13	26.9	21.1-NR	<0.001	Paclitaxel	2.23	1.07-4.68	0.0334
Paclitaxel	37	16	85.4	54.3-NR			1.00		
Pemetrexed	33	28	15.5	12.3-31.1			5.07	2.60-9.87	<0.001
Vinorelbine	50	19	43.1	32.0-NR			1.33	0.67-2.62	0.4164
Surgery									
Yes	33	11	114.0	73.9-NR	<0.001	Yes	5.55	2.76-11.12	<0.001
No	108	65	26.1	21.4-32.2					
Radiotherapy									
Yes	45	17	41.8	28.1-NR	0.2090				
No	96	59	30.0	24.5-54.3					
EGFR status									
Wild type	16	13	26.9	20.9-NR	0.4160				
Mutated	72	48	27.7	23.1-39.1					

N: number of patients; MST: median survival time (months); NR: not reached; HR: hazard ratio; 95% CI: 95% confidence interval.

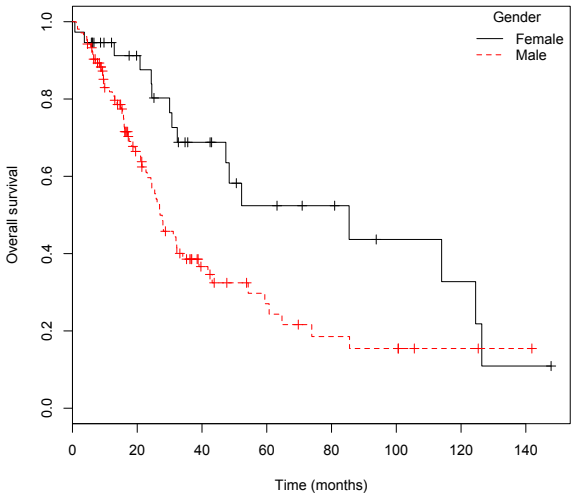

Figure 3. Kaplan-Meier curve for overall survival according to gender.

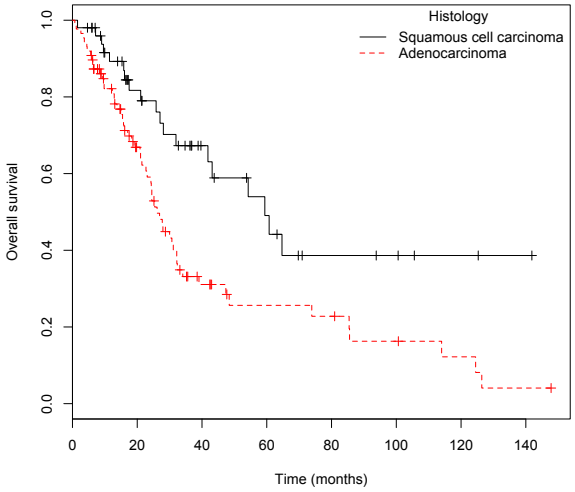

Figure 4. Kaplan-Meier curve for overall survival according to tumor histology.

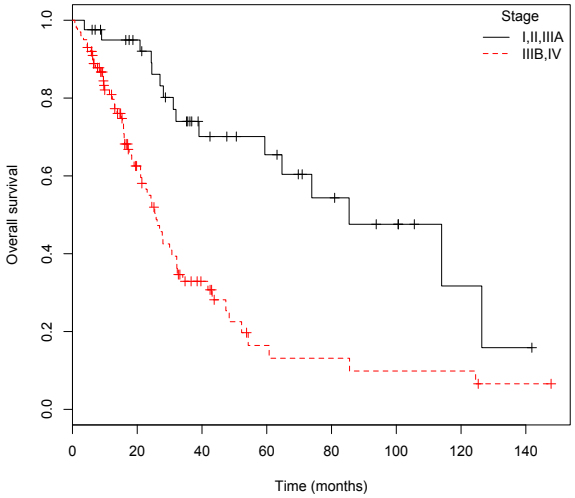

Figure 5. Kaplan-Meier curve for overall survival according to tumor stage.

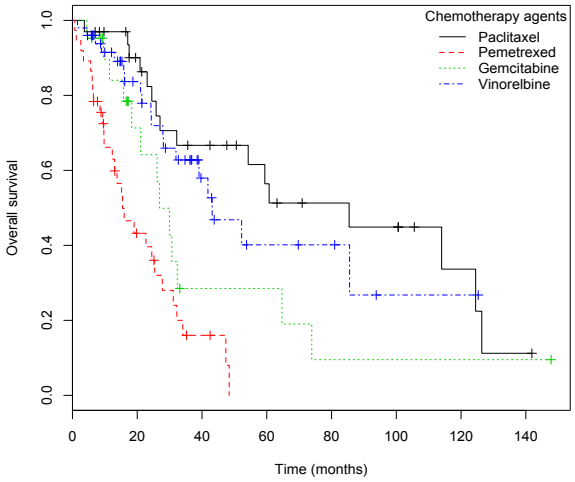

Figure 6. Kaplan-Meier curve for overall survival according to chemotherapy agents.

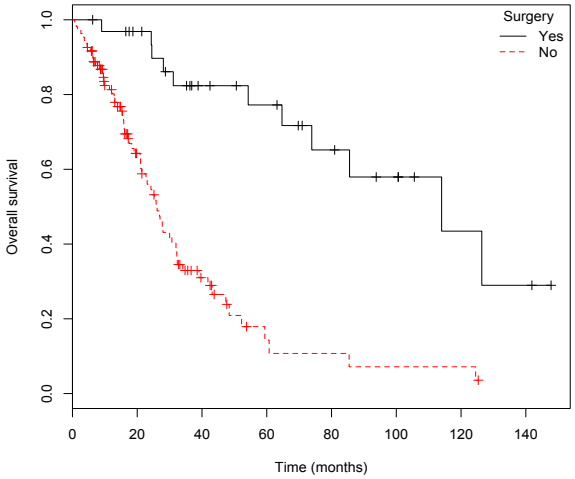

Figure 7. Kaplan-Meier curve for overall survival according to surgery.

Table 15. Association of clinical characteristics with progression-free survival.

Characteristic	PFS								
	N	Events	MST (mo)	95% CI	Log-Rank p-value	Reference Category	Univariate Cox Model		
							HR	95% CI	p-value
Gender									
Female	37	22	19.6	15.5-NR	0.0418	Female	1.63	1.01-2.63	0.0439
Male	104	78	11.0	8.5-17.1					
Family history									
Yes	85	63	14.6	11.0-20.4	0.6410				
No	56	37	10.9	6.9-23.2					
Personal history of cancer									
Yes	23	15	16.9	10.0-NR	0.4100				
No	118	85	13.2	10.1-18.7					
Previous lung disease									
Yes	34	21	19.2	10.20-38.4	0.2630				
No	107	79	12.9	9.17-17.6					
Smoking status									
Current-Smokers	72	49	16.1	10.3-34.4	0.3500				
Former-smokers	52	37	11.9	8.4-18.4					
Non-smokers	17	14	15.5	4.7-NR					
Age at NSCLC diagnosis									
≤60	68	51	10.9	7.6-19.6	0.7640				
>60	73	49	16.8	11.2-23.3					
Histology									
Adenocarcinoma	87	69	11.4	7.5-16.9	0.0034	Squamous cell carcinoma	1.89	1.23-2.91	0.0039
Squamous cell carcinoma	51	30	27.3	10.9-44.0					
Tumor stage									
I, II and IIIA	41	21	44.0	23.33-NA	<0.001	I, II and IIIA	2.81	1.71-4.63	<0.001
IIIB and IV	98	78	10.2	8.37-15.0					
Chemotherapy agents									
Gemcitabine	21	19	10.2	6.5-18.4	<0.001	Paclitaxel	3.20	1.65-6.24	<0.001
Paclitaxel	33	21	23.2	11.9-NR			1.00		
Pemetrexed	37	33	7.0	5.2-13.2			3.79	2.08-6.92	<0.001
Vinorelbine	50	27	34.4	9.2-NR			1.06	0.59-1.91	0.8363
Surgery									
Yes	33	14	82.2	44.0-NR	<0.001	Yes	4.93	2.73-8.91	<0.001
No	108	86	10.0	7.6-13.2					
Radiotherapy									
Yes	45	25	23.2	12.8-39.1	0.0566	Yes	1.55	0.98-2.44	0.0588
No	96	75	11.2	7.5-16.9					
EGFR status									
Wild type	72	62	10.3	7.7-17.1	0.3170				
Mutated	16	14	6.0	4.7-NR					

N: number of patients; MST: median survival time (months); NR: not reached; HR: hazard ratio; 95% CI: 95% confidence interval.

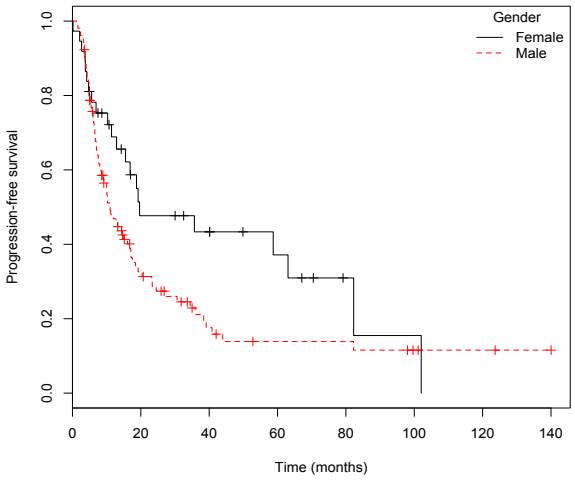

Figure 8. Kaplan-Meier curve for progression-free survival according to gender.

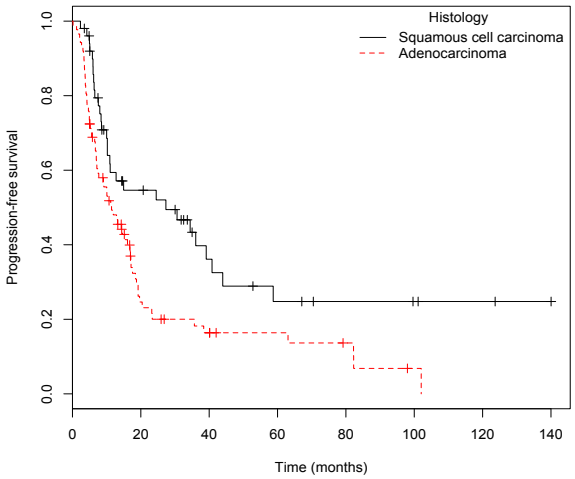

Figure 9. Kaplan-Meier curve for progression-free survival according to tumor histology.

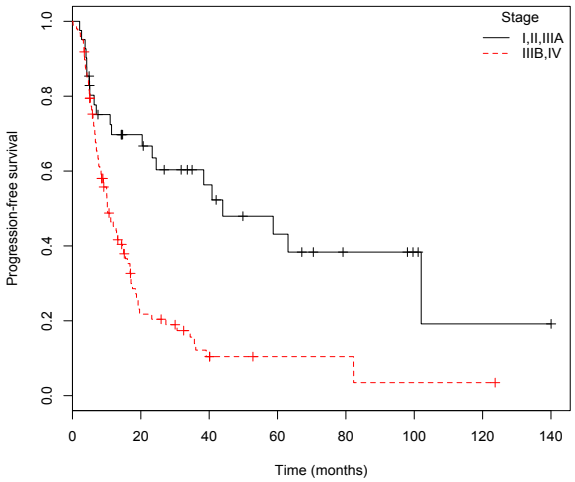

Figure 10. Kaplan-Meier curve for progression-free survival according to tumor stage.

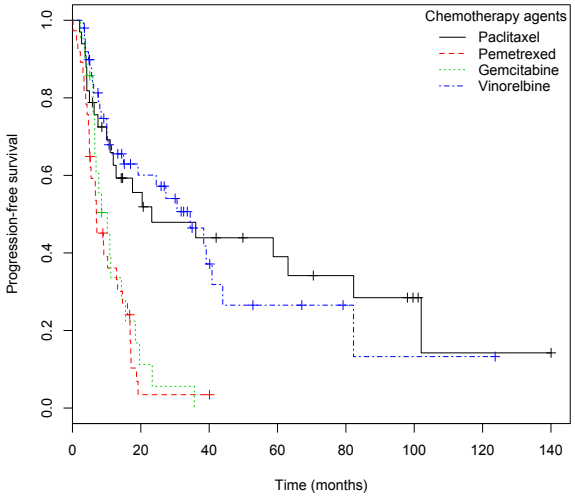

Figure 11. Kaplan-Meier curve for progression-free survival according to chemotherapy agents.

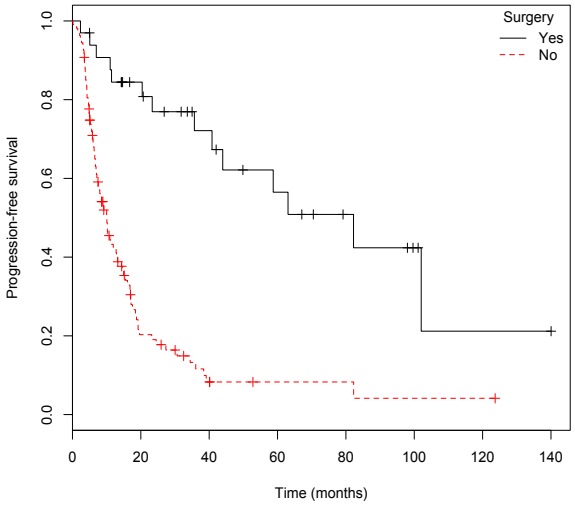

Figure 12. Kaplan-Meier curve for progression-free survival according to surgery.

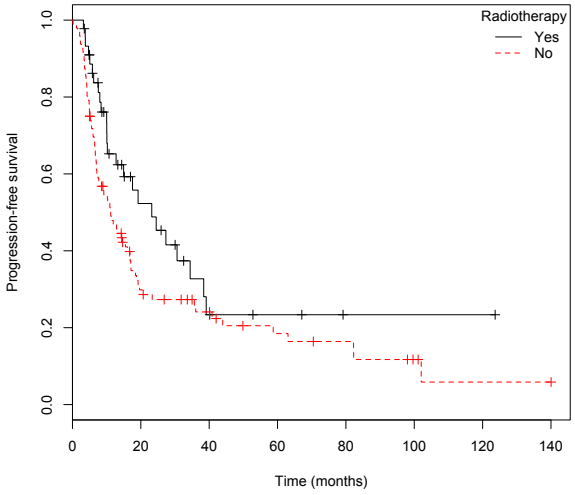

Figure 13. Kaplan-Meier curve for progression-free survival to concomitant or concurrent radiotherapy.

5.2.3 Toxicity

General toxicity was associated with personal history of cancer (p=0.0077; RR=7.99; $CI_{95\%}$=1.73-36.8; Table 16) and chemotherapy agents (p=0.0003; Table 16). Hematological toxicity was also associated with personal history of cancer (p=0.0311; RR=6.43; $CI_{95\%}$=1.18-34.91; Table 17) and chemotherapy agents (p=0.0022; Table 17). Nephrotoxicity was associated with histology (p=0.0486; Table 18).

Asthenia, gastrointestinal toxicity, infection, neurotoxicity and the occurrence of multiple adverse events were not associated with clinical or demographic characteristics (Tables 19, 20, 21, 22, 23, 24).

Table 16. Association of clinical characteristics with toxicity.

Characteristic	N	Toxicity		χ^2	p-value	Reference Category	RR	95% CI
		Grade 3-4 N (%)	Grade 2-3 N(%)					
Gender								
Female	37	11 (29.7)	26 (70.3)	0	1			
Male	104	31 (29.8)	73 (70.2)					
Family history								
Yes	56	21 (24.7)	64 (75.3)	2.0659	0.1506			
No	85	21 (37.5)	35 (62.5)					
Personal history of cancer								
Yes	23	1 (4.3)	22 (95.7)	7.1128	0.0077	Yes	7.99	1.73-36.80
No	118	41 (34.7)	77 (65.3)					
Previous lung disease								
Yes	34	11 (32.4)	23 (67.6)	0.0257	0.8727			
No	107	31 (29.0)	76 (71.0)					
Smoking status								
Current-Smokers	72	19 (26.4)	53 (73.6)	0.8539	0.6525			
Former-smokers	52	17 (32.7)	35 (67.3)					
Non-smokers	17	6 (35.3)	11 (64.7)					
Age at NSCLC diagnosis								
≤60	68	21 (30.9)	47 (69.1)	0.0081	0.9282			
>60	73	21 (28.8)	52 (71.2)					
Histology								
Adenocarcinoma	87	25 (28.7)	62 (71.3)	0.0180	0.8932			
Squamous cell carcinoma	51	16 (31.4)	35 (68.6)					
Tumor stage								
I, II and IIIA	41	12 (29.3)	29 (70.7)	0	1			
IIIB and IV	98	29 (29.6)	69 (70.4)					
Chemotherapy agents								
Gemcitabine	21	14 (66.7)	7 (33.3)	19.1570	0.0003	Paclitaxel	3.67	1.70-7.93
Paclitaxel	33	6 (18.2)	27 (81.8)				1.00	
Pemetrexed	37	6 (16.2)	31 (83.8)				0.89	0-Inf
Vinorelbine	50	16 (32.0)	34 (68.0)				1.76	0.67-4.64
Surgery								
Yes	33	11 (33.3)	22 (66.7)	0.0850	0.7707			
No	108	31 (28.7)	77 (71.3)					
Radiotherapy								
Yes	45	9 (20.0)	36 (80.0)	2.3788	0.1230			
No	96	33 (34.4)	63 (65.6)					
EGFR status								
Wild type	72	23 (31.9)	49 (68.1)		0.7669*			
Mutated	16	4 (25.0)	12 (75.0)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval; Inf: infinite.
*p-value for Fisher's Exact Test.

Table 17. Association of clinical characteristics with hematological toxicity.

Characteristic	N	Hematological Toxicity		χ^2	p-value	Reference Category	RR	95% CI
		Grade 3-4 N (%)	Grade 2-3 N(%)					
Gender								
Female	37	10 (27.0)	27 (73.0)	0.0669	0.7959			
Male	104	24 (23.1)	80 (76.9)					
Family history								
Yes	85	18 (21.2)	67 (78.8)	0.6452	0.4218			
No	56	16 (28.6)	40 (71.4)					
Personal history of cancer								
Yes	23	1 (4.3)	22 (95.7)	4.6479	0.0311	Yes	6.43	1.18-34.91
No	118	33 (28.0)	85 (72.0)					
Previous lung disease								
Yes	34	9 (26.5)	25 (73.5)	0.0192	0.8897			
No	107	25 (23.4)	82 (76.6)					
Smoking status								
Current-Smokers	72	15 (20.8)	57 (79.2)	0.9085	0.6349			
Former-smokers	52	14 (26.9)	38 (73.1)					
Non-smokers	17	5 (29.4)	12 (70.6)					
Age at NSCLC diagnosis								
≤60	68	21 (30.9)	47 (69.1)	2.613	0.1060			
>60	73	13 (17.8)	60 (82.2)					
Histology								
Adenocarcinoma	87	21 (24.1)	66 (75.9)	0	1			
Squamous cell carcinoma	51	12 (23.5)	39 (76.5)					
Tumor stage								
I, II and IIIA	41	10 (24.4)	31 (75.6)	<0.001	1			
IIIB and IV	98	23 (23.5)	75 (76.5)					
Chemotherapy agents								
Gemcitabine	21	11 (52.4)	10 (47.6)	14.6093	0.0022	Paclitaxel	3.46	1.36-8.77
Paclitaxel	33	5 (15.2)	28 (84.8)				1.00	
Pemetrexed	37	4 (10.8)	33 (89.2)				0.71	0.02-27.30
Vinorelbine	50	14 (28.0)	36 (72.0)				1.85	0.62-5.55
Surgery								
Yes	33	9 (27.3)	24 (72.7)	0.0636	0.8008			
No	108	25 (23.1)	83 (76.9)					
Radiotherapy								
Yes	45	7 (15.6)	38 (84.4)	2.0030	0.1570			
No	96	27 (28.1)	69 (71.9)					
EGFR status								
Wild type	16	4 (25.0)	12 (75.0)		1*			
Mutated	72	18 (25.0)	54 (75.0)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 18. Association of clinical characteristics with nephrotoxicity toxicity.

Characteristic	N	Nephrotoxicity		χ^2	p-value	Reference Category	RR	95% CI
		Grade 3-4 N (%)	Grade 2-3 N(%)					
Gender								
Female	37	0 (0.0)	37 (100.0)		0.5667*			
Male	104	3 (2.9)	101 (97.1)					
Family history								
Yes	85	0 (0.0)	85 (100.0)		0.0606*			
No	56	3 (5.4)	53 (94.6)					
Personal history of cancer								
Yes	23	0 (0.0)	23 (100.0)		1*			
No	118	3 (2.5)	115 (97.5)					
Previous lung disease								
Yes	34	2 (5.9)	32 (94.1)		0.1443*			
No	107	1 (0.9)	106 (99.1)					
Smoking status								
Current-Smokers	72	2 (2.8)	70 (97.2)		1*			
Former-smokers	52	1 (1.9)	51 (98.1)					
Non-smokers	17	0 (0.0)	17 (100.0)					
Age at NSCLC diagnosis								
≤60	68	1 (1.5)	67 (98.5)		1*			
>60	73	2 (2.7)	71 (97.3)					
Histology								
Adenocarcinoma	87	0 (0.0)	87 (100.0)		0.0486*			
Squamous cell carcinoma	51	3 (5.9)	48 (94.1)					
Tumor stage								
I, II and IIIA	41	0 (0.0)	41 (100.0)		0.5551*			
IIIB and IV	98	3 (3.1)	95 (96.9)					
Chemotherapy agents								
Gemcitabine	21	2 (9.5)	19 (90.5)					
Paclitaxel	33	0 (0.0)	33 (100.0)		0.0869*			
Pemetrexed	37	0 (0.0)	37 (100.0)					
Vinorelbine	50	1 (2.0)	49 (98.0)					
Surgery								
Yes	33	0 (0.0)	33 (100.0)		1*			
No	108	3 (2.8)	105 (97.2)					
Radiotherapy								
Yes	45	1 (2.2)	44 (97.8)		1*			
No	96	2 (2.1)	94 (97.9)					
EGFR status								
Wild type	16	0 (0.0)	16 (100.0)		1*			
Mutated	72	1 (1.4)	71 (98.6)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 19. Association of clinical characteristics with asthenia.

Characteristic	N	Asthenia		χ^2	p-value	Reference Category	RR	95% CI
		Grade 3-4 N (%)	Grade 2-3 N(%)					
Gender								
Female	37	0 (0.0)	37 (100.0)		0.5667*			
Male	104	3 (2.9)	101 (97.1)					
Family history								
Yes	85	1 (1.2)	84 (98.8)		0.5628*			
No	56	2 (3.6)	54 (96.4)					
Personal history of cancer								
Yes	23	0 (0.0)	23 (100.0)		1*			
No	118	3 (2.5)	115 (97.5)					
Previous lung disease								
Yes	34	1 (2.9)	33 (97.1)		0.5660*			
No	107	2 (1.9)	105 (98.1)					
Smoking status								
Current-Smokers	72	1 (1.4)	71 (98.6)		0.7094*			
Former-smokers	52	2 (3.8)	50 (96.2)					
Non-smokers	17	0 (0.0)	17 (100.0)					
Age at NSCLC diagnosis								
≤60	68	0 (0.0)	68 (100.0)		0.2456*			
>60	73	3 84.1)	70 (95.9)					
Histology								
Adenocarcinoma	87	2 (2.3)	85 (97.7)		1*			
Squamous cell carcinoma	51	1 (2.0)	50 (98.0)					
Tumor stage								
I, II and IIIA	41	1 (2.4)	40 (97.6)		1*			
IIIB and IV	98	2 (2.0)	96 (98.0)					
Chemotherapy agents								
Gemcitabine	21	0 (0.0)	21 (100.0)					
Paclitaxel	33	1 (3.0)	36 (97.0)		1*			
Pemetrexed	37	1 (2.7)	32 (97.3)					
Vinorelbine	50	1 (2.0)	49 (98.0)					
Surgery								
Yes	33	1 (3.0)	32 (97.0)		0.5536*			
No	108	2 (1.9)	106 (98.1)					
Radiotherapy								
Yes	45	1 (2.2)	44 (97.8)		1*			
No	96	2 (2.1)	94 (97.9)					
EGFR status								
Wild type	16	0 (0.0)	16 (100.0)		1*			
Mutated	72	1 (1.4)	71 (98.6)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 20. Association of clinical characteristics with gastrointestinal toxicity.

Characteristic	N	Gastrointestinal Toxicity		χ^2	p-value	Reference Category	RR	95% CI
		Grade 3-4 N (%)	Grade 2-3 N(%)					
Gender								
Female	37	1 (2.7)	36 (97.3)		1*			
Male	104	2 (1.9)	102 (98.1)					
Family history								
Yes	85	2 (2.4)	83 (97.6)		1*			
No	56	1 (1.8)	55 (98.2)					
Personal history of cancer								
Yes	23	1 (4.3)	22 (95.7)		0.4163*			
No	118	2 (1.7)	116 (98.3)					
Previous lung disease								
Yes	34	1 (2.9)	33 (97.1)		0.5660*			
No	107	2 (1.9)	105 (98.1)					
Smoking status								
Current-Smokers	72	2 (2.8)	70 (97.2)		0.2310*			
Former-smokers	52	0 (0.0)	52 (100.0)					
Non-smokers	17	1 (5.9)	16 (94.1)					
Age at NSCLC diagnosis								
≤60	68	0 (0.0)	68 (100.0)		0.2456*			
>60	73	3 (4.1)	70 (95.9)					
Histology								
Adenocarcinoma	87	3 (3.4)	84 (96.6)		0.2959*			
Squamous cell carcinoma	51	0 (0.0)	51 (100.0)					
Tumor stage								
I, II and IIIA	41	1 (2.4)	40 (97.6)		1*			
IIIB and IV	98	2 (2.0)	96 (98.0)					
Chemotherapy agents								
Gemcitabine	21	2 (9.5)	19 (90.5)					
Paclitaxel	33	0 (0.0)	33 (100.0)		0.0869*			
Pemetrexed	37	0 (0.0)	37 (100.0)					
Vinorelbine	50	1 (2.0)	49 (98.0)					
Surgery								
Yes	33	1 (3.0)	32 (97.0)		0.5536*			
No	108	2 (1.9)	106 (98.1)					
Radiotherapy								
Yes	45	1 (2.2)	44 (97.8)		1*			
No	96	2 (2.1)	94 (97.9)					
EGFR status								
Wild type	16	0 (0.0)	16 (100.0)		1*			
Mutated	72	3 (4.2)	69 (95.8)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 21. Association of clinical characteristics with infection.

Characteristic	N	Infection		χ^2	p-value	Reference Category	RR	95% CI
		Grade 3-4 N (%)	Grade 2-3 N(%)					
Gender								
Female	37	1 (2.7)	36 (97.3)		1*			
Male	104	4 (3.8)	100 (96.2)					
Family history								
Yes	85	2 (2.4)	83 (97.6)		0.3859*			
No	56	3 (5.4)	53 (94.6)					
Personal history of cancer								
Yes	23	0 (0.0)	23 (100.0)		0.5917*			
No	118	5 (4.2)	113 (95.8)					
Previous lung disease								
Yes	34	0 (0.0)	34 (100.0)		0.3365*			
No	107	5 (4.7)	102 (95.3)					
Smoking status								
Current-Smokers	72	3 (4.2)	69 (95.8)		1*			
Former-smokers	52	2 (3.8)	50 (96.2)					
Non-smokers	17	0 (0.0)	17 (100.0)					
Age at NSCLC diagnosis								
≤60	68	1 (1.5)	67 (98.5)		0.3676*			
>60	73	4 (5.5)	69 (94.5)					
Histology								
Adenocarcinoma	87	4 (4.6)	83 (95.4)		0.6513*			
Squamous cell carcinoma	51	1 (2.0)	50 (98.0)					
Tumor stage								
I, II and IIIA	41	0 (0.0)	41 (100.0)		0.3215*			
IIIB and IV	98	5 (5.1)	93 (94.9)					
Chemotherapy agents								
Gemcitabine	21	0 (0.0)	21 (100.0)					
Paclitaxel	33	0 (0.0)	33 (100.0)		0.3383*			
Pemetrexed	37	3 (8.1)	34 (91.9)					
Vinorelbine	50	2 (4.0)	48 (96.0)					
Surgery								
Yes	33	0 (0.0)	30 (100.0)		0.5909*			
No	108	5 (4.6)	103 (95.4)					
Radiotherapy								
Yes	45	2 (4.4)	43 (95.6)		0.6542*			
No	96	3 (3.1)	93 (96.9)					
EGFR status								
Wild type	16	0 (0.0)	16 (100.0)		1*			
Mutated	72	4 (5.6)	68 (94.4)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 22. Association of clinical characteristics with neurotoxicity.

Characteristic	N	Neurotoxicity		χ^2	p-value	Reference Category	RR	95% CI
		Grade 3-4 N (%)	Grade 2-3 N(%)					
Gender								
Female	37	0 (0.0)	37 (100.0)		1*			
Male	104	1 (1.0)	103 (99.0)					
Family history								
Yes	85	0 (0.0)	85 (100.0)		0.3972*			
No	56	1 (1.8)	55 (98.2)					
Personal history of cancer								
Yes	23	0 (0.0)	23 (100.0)		1*			
No	118	1 (0.8)	117 (99.2)					
Previous lung disease								
Yes	34	0 (0.0)	34 (100.0)		1*			
No	107	1 (0.9)	106 (99.1)					
Smoking status								
Current-Smokers	72	1 (1.4)	71 (98.6)		1*			
Former-smokers	52	0 (0.0)	52 (100.0)					
Non-smokers	17	0 (0.0)	17 (100.0)					
Age at NSCLC diagnosis								
≤60	68	0 (0.0)	68 (100.0)		1*			
>60	73	1 (1.4)	72 (98.6)					
Histology								
Adenocarcinoma	87	0 (0.0)	87 (100.0)		0.3696*			
Squamous cell carcinoma	51	1 (2.0)	50 (98.0)					
Tumor stage								
I, II and IIIA	41	1 (2.4)	40 (97.6)		0.2950*			
IIIB and IV	98	0 (0.0)	98 (100.0)					
Chemotherapy agents								
Gemcitabine	21	0 (0.0)	21 (100.0)					
Paclitaxel	33	1 (3.0)	32 (97.0)		0.3830*			
Pemetrexed	37	0 (0.0)	37 (100.0)					
Vinorelbine	50	0 (0.0)	50 (100.0)					
Surgery								
Yes	33	1 (3.0)	32 (97.0)		0.2340*			
No	108	0 (0.0)	108 (100.0)					
Radiotherapy								
Yes	45	0 (0.0)	45 (100.0)		1*			
No	96	1 (1.0)	95 (99.0)					
EGFR status								
Wild type	16	0 (0.0)	16 (100.0)		1*			
Mutated	72	0 (0.0)	72 (100.0)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 23. Association of clinical characteristics with more than one adverse events.

Characteristic	N	Toxicity		χ^2	p-value	Reference Category	RR	95% CI
		>1 N (%)	0-1 N(%)					
Gender								
Female	37	29 (78.4)	8 (21.6)	2.1257	0.1448			
Male	104	66 (63.5)	38 (36.5)					
Family history								
Yes	85	61 (71.8)	24 (28.2)	1.4064	0.2357			
No	56	34 (60.7)	22 (39.3)					
Personal history of cancer								
Yes	23	15 (65.2)	8 (34.8)	0	1			
No	118	80 (67.8)	38 (32.2)					
Previous lung disease								
Yes	34	24 (70.6)	10 (29.4)	0.0618	0.8036			
No	107	71 (66.4)	36 (33.6)					
Smoking status								
Current-Smokers	72	51 (70.8)	21 (29.2)	0.8092	0.6673			
Former-smokers	52	33 (63.5)	19 (36.5)					
Non-smokers	17	11 (64.7)	6 (35.3)					
Age at NSCLC diagnosis								
≤60	68	49 (72.1)	19 (27.9)	0.9312	0.3346			
>60	73	46 (63.0)	27 (37.0)					
Histology								
Adenocarcinoma	87	60 (69.0)	27 (31.0)	0.1070	0.7436			
Squamous cell carcinoma	51	33 (64.7)	18 (35.3)					
Tumor stage								
I, II and IIIA	41	31 (75.6)	10 (24.4)	0.9822	0.3217			
IIIB and IV	98	64 (65.3)	34 (34.7)					
Chemotherapy agents								
Gemcitabine	21	12 (57.1)	9 (42.9)					
Paclitaxel	33	21 (63.6)	12 (36.4)	1.8378	0.6067			
Pemetrexed	37	26 (70.3)	11 (29.7)					
Vinorelbine	50	36 (72.0)	14 (28.0)					
Surgery								
Yes	33	22 (66.7)	11 (33.3)	<0.001	1			
No	108	73 (67.6)	35 (32.4)					
Radiotherapy								
Yes	45	32 (71.1)	13 (28.9)	0.2071	0.6491			
No	96	63 (65.6)	33 (34.4)					
EGFR status								
Wild type	72	49 (68.1)	23 (31.9)	0.0179	0.8937			
Mutated	16	10 (62.5)	6 (37.5)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 24. Association of clinical characteristics with more than two adverse events.

Characteristic	N	Toxicity		χ^2	p-value	Reference Category	RR	95% CI
		>2 N (%)	0-2 N(%)					
Gender								
Female	37	11 (29.7)	26 (70.3)	0.2138	0.6438			
Male	104	25 (24.0)	79 (76.0)					
Family history								
Yes	85	19 (22.4)	66 (77.6)	0.7555	0.3847			
No	56	17 (30.4)	39 (69.6)					
Personal history of cancer								
Yes	23	4 (17.4)	19 (82.6)	0.5146	0.4731			
No	118	32 (27.1)	86 (72.9)					
Previous lung disease								
Yes	34	10 (29.4)	24 (70.6)	0.1368	0.7115			
No	107	26 (24.3)	81 (75.7)					
Smoking status								
Current-Smokers	72	16 (22.2)	56 (77.8)	2.6109	0.2710			
Former-smokers	52	13 (25.0)	39 (75.0)					
Non-smokers	17	7 (41.2)	10 (58.8)					
Age at NSCLC diagnosis								
≤60	73	18 (26.5)	55 (73.5)	0.0029	0.9574			
>60	68	18 (24.7)	50 (75.3)					
Histology								
Adenocarcinoma	87	25 (28.7)	62 (71.3)	0.9740	0.3237			
Squamous cell carcinoma	51	10 (19.6)	41 (80.4)					
Tumor stage								
I, II and IIIA	41	9 (22.0)	32 (78.0)	0.2256	0.6348			
IIIB and IV	98	27 (27.6)	71 (72.4)					
Chemotherapy agents								
Gemcitabine	21	7 (33.3)	14 (66.7)					
Paclitaxel	37	7 (21.2)	26 (78.8)	1.6671	0.6805			
Pemetrexed	33	11 (29.7)	26 (70.3)					
Vinorelbine	50	11 (22.0)	39 (78.0)					
Surgery								
Yes	33	8 (24.2)	25 (75.8)	<0.001	1			
No	108	28 (25.9)	80 (74.1)					
Radiotherapy								
Yes	45	8 (17.8)	37 (82.2)	1.5340	0.2155			
No	96	28 (29.2)	68 (70.8)					
EGFR status								
Wild type	72	18 (18.8)	54 (82.2)		0.7521*			
Mutated	16	3 (25.0)	13 (75.0)					

N: number of patients; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

5.3 Genotypes Distribution

Genotype frequencies were in agreement with the values expected under the Hardy-Weinberg equilibrium model. Linkage disequilibrium values D' and r^2 are shown in Table 25. In particular, IL1B rs1143627/IL1B rs16944 and IL1B rs1143623/IL1B rs12621220 pairs were in strong linkage disequilibrium.

Table 25. Linkage disequilibrium

CHR	BP	SNP	Gene	CHR	BP	SNP	Gene	R2	D
1	11854476	rs1801131	MTHFR	1	11856378	rs1801133	MTHFR	0.305884	1.000
2	113594387	rs1143627	IL-1B	2	113594867	rs16944	IL-1B	0.983219	1.000
2	113594387	rs1143627	IL-1B	2	113595829	rs1143623	IL-1B	0.631679	1.000
2	113594387	rs1143627	IL-1B	2	113598255	rs12621220	IL-1B	0.631679	1.000
2	113594867	rs16944	IL-1B	2	113595829	rs1143623	IL-1B	0.605375	0.977
2	113594867	rs16944	IL-1B	2	113598255	rs12621220	IL-1B	0.605375	0.977
2	113595829	rs1143623	IL-1B	2	113598255	rs12621220	IL-1B	1	1.000
7	87138645	rs1045642	ABCB1	7	87160618	rs2032582	ABCB1	0.637051	0.865
7	87138645	rs1045642	ABCB1	7	87179601	rs1128503	ABCB1	0.550816	0.764
7	87160618	rs2032582	ABCB1	7	87179601	rs1128503	ABCB1	0.678695	0.841
19	45854919	rs13181	ERCC2	19	45857049	rs238416	ERCC2	0.324509	0.966
19	45854919	rs13181	ERCC2	19	45867259	rs1799793	ERCC2	0.604276	0.772
19	45854919	rs13181	ERCC2	19	45912736	rs3212986	ERCC1	0.26341	0.554
19	45854919	rs13181	ERCC2	19	45923653	rs11615	ERCC1	0.247201	0.512
19	45867259	rs1799793	ERCC2	19	45912736	rs3212986	ERCC1	0.318222	0.653
19	45867259	rs1799793	ERCC2	19	45923653	rs11615	ERCC1	0.274173	0.559
19	45912736	rs3212986	ERCC1	19	45923653	rs11615	ERCC1	0.493236	0.975

CHR: chromosome; BP: physical position (base-pair)

5.4 Influence of gene polymorphisms on clinical outcomes outcomes of platinum-based chemotherapy

5.4.1 Response

XRCC1 Gln399Arg was associated with response. Patients carrying the GG genotype showed significantly better ORR compared to those with AG/AA genotypes (p=0.0343; RR=1.29; $CI_{95\%}$=1.02-1.63; Table 26). A trend towards better ORR was showed for those patients with ERCC1 C8092A-GG genotype, but it was not statistically significant (p=0.0825; RR=1.24; $CI_{95\%}$=0.97-1.58; Table 26). Logistic regression model adjusted by resection revealed that XRCC1 Gln399Arg-GG genotype and ERCC1 C8092A-GG were independently associated with response ($p_{likelihood\ ratio\ test}=8.663\cdot10^{-7}$; Table 27).

Table 26. Association of gene polymorphisms with response.

Gene	SNPs	Genotype	N	Response		χ^2	p-value	Ref. Cat.	RR	95% CI
				CR+PR N (%)	SD+PD N (%)					
ABCB1	rs1045642	CC	48	35 (72.9)	13 (27.1)					
		CT	62	43 (69.4)	19 (30.6)	0.3655	0.8330			
		TT	30	20 (66.7)	10 (33.3)					
		C	110	78 (70.9)	32 (29.1)	0.0505	0.8222			
		T	92	63 (68.5)	29 (31.5)	0.1223	0.7266			
	rs1128503	CC	52	38 (73.1)	14 (26.9)					
		CT	63	45 (71.4)	18 (28.6)	1.4861	0.4757			
		TT	25	15 (60.0)	10 (40.0)					
		C	115	83 (72.2)	32 (27.8)	0.9275	0.3355			
		T	88	60 (68.2)	28 (31.8)	0.1763	0.6746			
	rs2032582	GG	69	34 (73.9)	12 (26.1)					
		G (GT/AG)	46	49 (71.0)	20 (29.0)	1.5597	0.4585			
		T (TT/AT)	25	15 (60.0)	10 (40.0)					
		G vs TT/AT	115	83 (72.2)	32 (27.8)	0.9275	0.3355			
		G/T vs GG	94	64 (68.1)	30 (31.9)	0.2606	0.6097			
ERCC1	rs11615	CC	19	14 (73.7)	5 (26.3)					
		CT	72	48 (66.7)	24 (33.3)	0.7846	0.6755			
		TT	49	36 (73.5)	13 (26.5)					
		C	91	62 (68.1)	29 (31.9)	0.2153	0.6426			
		T	121	84 (69.4)	37 (30.6)	0.0116	0.9142			
	rs3212986	GG	74	57 (77.0)	17 (23.0)					
		GT	56	34 (60.7)	22 (39.3)	4.0393	0.1327			
		TT	10	7 (70.0)	3 (30.0)					
		G	130	91 (70.0)	39 (30.0)		1.00*			
		T	66	41 (62.1)	25 (37.9)	3.0153	0.0825	T	1.24	0.97-1.58
ERCC2	rs13181	GG	24	14 (58.3)	10 (41.7)					
		GT	55	40 (72.7)	15 (27.3)	1.8823	0.3902			
		TT	61	44 (72.1)	17 (27.9)					
		G	79	54 (68.4)	25 (31.6)	0.0885	0.7660			
		T	116	84 (72.4)	32 (27.6)	1.2668	0.2604			
	rs1799793	AA	23	15 (65.2)	8 (34.8)					
		AG	55	38 (69.1)	17 (30.9)	0.4688	0.7911			
		GG	62	45 (72.6)	17 (27.4)					
		A	78	53 (67.9)	25 (32.1)	0.1668	0.6830			
		G	117	83 (70.9)	34 (29.1)	0.0892	0.7652			
	rs50872	CC	81	59 (56.7)	22 (24.3)					
		CT	54	36 (37.8)	18 (16.2)		0.5884*			
		TT	5	3 (3.5)	2 (1.5)					
		C	135	95 (70.4)	40 (29.6)		0.6361*			
		T	59	39 (66.1)	20 (33.9)	0.4520	0.5014			
	rs238416	AA	19	11 (57.9)	8 (42.1)					
		AG	67	50 (74.6)	17 (25.4)	2.0653	0.3561			
		GG	54	37 (68.5)	17 (31.5)					
		A	86	61 (70.9)	25 (29.1)	0.0129	0.9095			
		G	121	87 (71.9)	34 (28.1)	0.9395	0.3324			
ERCC5	rs17655	CC	79	58 (73.4)	21 (26.6)					
		CG	51	33 (64.7)	18 (35.3)	1.1201	0.5712			
		GG	10	7 (70.0)	3 (30.0)					
		C	130	91 (70.0)	39 (30.0)		1.00*			
		G	61	40 (65.6)	21 (34.4)	0.6696	0.4132			
	rs1047768	CC	43	30 (69.8)	13 (30.2)					
		CT	70	51 (72.9)	19 (27.1)	0.9099	0.6345			
		TT	27	17 (63.0)	10 (37.0)					
		C	113	81 (71.7)	32 (28.3)	0.4283	0.5128			
		T	97	68 (70.1)	29 (29.9)	<0.001	1.00			
IL1B	rs12621220	CC	73	51 (69.9)	22 (30.1)					
		CT	60	41 (68.3)	19 (31.7)		0.7496*			
		TT	7	6 (85.7)	1 (14.3)					
		C	133	92 (69.2)	41 (30.8)		0.6744*			
		T	67	47 (70.1)	20 (29.9)	<0.001	1.00			
	rs1143623	CC	7	6 (85.7)	1 (14.3)					
		CG	60	41 (68.3)	19 (31.7)		0.7496*			
		GG	73	51 (69.9)	22 (30.1)					
		C	67	47 (70.1)	20 (29.9)	<0.001	1.00			
		G	133	92 (69.2)	41 (30.8)		0.6744*			

Table 26. (Continued).

Gene	SNPs	Genotype	N	Response		χ^2	p-value	Ref. Cat.	RR	95% CI
				CR+PR N (%)	SD+PD N (%)					
IL1B	rs16944	AA	14	8 (57.1)	6 (42.9)					
		AG	70	51 (72.9)	19 (27.1)	1.3776	0.5022			
		GG	56	39 (69.6)	17 (30.4)					
		A	84	59 (70.2)	25 (29.8)	<0.001	1.0			
		G	126	90 (71.4)	36 (28.6)		0.3559*			
	rs1143627	CC	15	9 (60.0)	6 (40.0)					
		CT	69	50 (72.5)	19 (27.5)	0.91713	0.6322			
		TT	56	39 (69.6)	17 (30.4)					
		C	84	59 (70.2)	25 (29.8)	<0.001	1.00			
		T	125	89 (71.2)	36 (28.8)		0.3822*			
IL6	rs1800795	CC	17	9 (52.9)	8 (47.1)					
		CG	69	50 (72.5)	19 (27.5)	2.6822	0.2616			
		GG	54	39 (72.2)	15 (27.8)					
		C	86	59 (68.6)	27 (31.4)	0.0703	0.7908			
		G	123	89 (72.4)	34 (27.6)	1.8364	0.1754			
IL16	rs7170924	GG	79	51 (64.6)	28 (35.4)					
		GT	52	40 (76.9)	12 (23.1)	2.5606	0.2780			
		TT	9	7 (77.8)	2 (22.2)					
		G	131	91 (69.5)	40 (30.5)		0.7241*			
		T	61	47 (77.0)	14 (23.0)	1.9976	0.1575			
MDM2	rs1470383	CC	7	6 (85.7)	1 (14.3)					
		CT	34	26 (76.5)	8 (23.5)		0.4208*			
		TT	99	66 (66.7)	33 (33.3)					
		C	41	32 (78.0)	9 (22.0)	1.2877	0.2565			
		T	133	92 (69.2)	41 (30.8)		0.6744*			
	rs1690924	AA	56	41 (73.2)	15 (26.8)					
		AG	64	44 (68.8)	20 (31.2)	0.7406	0.6905			
		GG	19	12 (63.2)	7 (36.8)					
		A	120	85 (70.8)	35 (29.2)	0.1666	0.6832			
		G	83	56 (67.5)	27 (32.5)	0.2863	0.5926			
MTHFR	rs1801131	AA	74	53 (71.6)	21 (28.4)					
		AC	53	36 (67.9)	17 (32.1)	0.2050	0.9026			
		CC	13	9 (69.2)	4 (30.8)					
		A	127	89 (70.1)	38 (29.9)		1.00*			
		C	66	45 (68.2)	21 (31.8)	0.0669	0.7959			
	rs1801133	CC	44	29 (65.9)	15 (34.1)					
		CT	73	52 (71.2)	21 (28.8)	0.5712	0.7516			
		TT	23	17 (73.9)	6 (26.1)					
		C	117	81 (69.2)	36 (30.8)	0.0396	0.8422			
		T	96	69 (71.9)	27 (28.1)	0.2667	0.6055			
MTR	rs1805087	AA	99	71 (71.7)	28 (28.3)					
		AG	37	24 (64.9)	13 (35.1)		0.8038*			
		GG	4	3 (75.0)	1 (25.0)					
		A	136	95 (69.9)	41 (30.1)		1.00*			
		G	41	27 (65.9)	14 (34.1)	0.2365	0.6267			
SLC19A1	rs1051266	AA	35	25 (71.4)	10 (28.6)					
		AG	63	43 (68.3)	20 (31.7)	0.1663	0.9202			
		GG	42	30 (71.4)	12 (28.6)					
		A	98	68 (69.4)	30 (30.6)	0.0016	0.9679			
		G	105	73 (69.5)	32 (30.5)	<0.001	1.00			
XRCC1	rs1799782	CC	119	81 (68.1)	38 (31.9)					
		CT	20	16 (80.0)	4 (20.0)		0.601*			
		TT	1	1 (100.0)	0 (0.0)					
		C	139	97 (69.8)	42 (30.2)	<0.001	1.00			
		T	21	17 (81.0)	4 (19.0)	0.8644	0.3525			
	rs25487	AA	20	14 (70.0)	6 (30.0)					
		AG	70	43 (61.4)	27 (38.6)	5.8776	0.0529			
		GG	50	41 (82.0)	9 (18.0)					
		A	90	57 (63.3)	33 (36.7)	4.4815	0.0343	1.29	A	1.02-1.63
		G	120	84 (70.0)	36 (30.0)	<0.001	1.00			

N: number of patients; CR: partial response + complete response; SD+PD: stable disease + progressive disease; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval

*p-value for Fisher's Exact Test.

Table 27. Influence of clinical characteristic and gene polymorphisms on response.

	Response	
	OR ($CI_{95\%}$)	**p-value**
Surgery (Yes)	25.38 (4.94-467.06)	0.0021
ERCC1 C8092A-GG	2.50 (1.12-5.69)	0.0268
XRCC1 Gln399Arg-GG	2.99 (1.26-7.62)	0.0161

OR: odds ratio; $CI_{95\%}$: 95% confidence interval

5.4.2 Survival

5.4.2.1 Overall survival

The bivariate analysis revealed that IL16 rs7170924 polymorphism was statistically significant associated with OS. Particularly, patients with GG genotype were in higher risk of death compared to those carrying the T-allele (p=0.0240; HR=1.76; $CI_{95\%}$=1.08-2.87; Table 28). The Figure 14 shows the Kaplan-Meier curve in accordance to IL16 rs7170924-T allele ($p_{log\text{-}rank}$=0.023). Median OS was 28.1 months ($CI_{95\%}$=24.3-41.8) for GG genotype, whereas for GT and TT genotypes, the median OS was 64.7 months ($CI_{95\%}$=27.7-not reached [NR]) and 73.9 months ($CI_{95\%}$=16.0-NR), respectively. Kaplan-Meier curve for MDM2 rs1690924-A allele showed a trend to higher risk of death for GG genotype, but this was not statistically significant ($p_{log\text{-}rank}$=0.086; Table 28) (Figure 15). Patients with GG genotype showed a median OS of 17.5 months ($CI_{95\%}$= 15.2-NR), whereas for AG and AA genotypes was 43.1 ($CI_{95\%}$=30.7-85.4) and 32.2 ($CI_{95\%}$=24.5-73.9) months, respectively. Patients with CC genotype for XRCC1 Arg194Trp polymorphism showed a trend to higher risk of death compared to those carrying the T-allele, but this was not statistically significant either (p=0.0777; HR=1.88; $CI_{95\%}$=0.93-3.79; Table 28). The Figure 16 shows the Kaplan-Meier curve in accordance to XRCC1 Arg194Trp-T allele ($p_{log\text{-}rank}$=0.073). Median OS was 30.0 months ($CI_{95\%}$=25.4-41.8) for CC genotype, whereas for CT genotype the median OS was 85.4 months ($CI_{95\%}$=48.4-NR). For the TT genotype, the survival median values exceeded the survival time of the further observation.

Multivariate Cox regression adjusted by gender, tumor histology, chemotherapy agents and surgery revealed that MDM2 rs1690924 gene polymorphism was associated to OS ($p_{likelihood\ ratio\ test}$=$3.391 \cdot 10^{-13}$; Table 29).

Table 28. Association of gene polymorphisms with overall survival.

Gene	SNPs	Genotype	N	OS							
				Events	MST (mo)	95% CI	Log Rank p-value	Ref. Cat.	Univariate Cox Model		
									HR	95% CI	p-value
ABCB1	rs1045642	CC	49	28	28.1	24.2-64.7					
		CT	62	36	39.1	25.8-73.9	0.697				
		TT	30	12	52.2	27.9-NR					
		C	111	64	32.0	25.8-48.4	0.428				
		T	92	48	43.1	27.9-60.7	0.572				
	rs1128503	CC	53	30	27.9	24.5-NR					
		CT	63	34	32.2	25.8-73.9	0.901				
		TT	25	12	52.2	30.0-NR					
		C	116	64	32.0	26.1-48.4	0.717				
		T	88	46	39.1	27.7-59.4	0.696				
	rs2032582	GG	47	27	30.7	24.5-NR					
		G(GT/AG)	69	38	39.1	25.8-73.9	0.880				
		T (TT/AT)	25	11	41.8	21.4-NR					
		G vs TT/AT	116	65	32.0	26.1-48.4	0.672				
		G/T vs GG	94	49	41.8	26.9-59.4	0.694				
ERCC1	rs11615	CC	20	9	41.8	15.8-NR					
		CT	72	40	32.4	24.5-60.7	0.876				
		TT	49	27	28.1	24.5-85.6					
		C	92	49	41.8	25.8-59.4	0.687				
		T	121	67	31.1	26.1-54.3	0.674				
	rs3212986	GG	74	42	32.4	27.7-60.7					
		GT	56	29	25.4	23.1-73.9	0.430				
		TT	11	5	64.7	34.1-NR					
		G	130	71	30.7	25.8-47.4	0.260				
		T	67	34	32.0	24.3-64.7	0.876				
ERCC2	rs13181	GG	25	12	26.1	15.8-NR					
		GT	55	29	47.4	31.1-114.0	0.385				
		TT	61	35	28.1	24.5-54.3					
		G	80	41	43.1	27.9-73.9	0.233				
		T	116	64	32.4	27.7-52.2	0.799				
	rs1799793	AA	24	11	52.2	26.1-NR					
		AG	55	33	32.4	25.4-59.4	0.754				
		GG	62	32	28.1	24.5-124.5					
		A	79	44	34.1	26.1-59.4	0.911				
		G	117	65	32.0	25.8-48.4	0.467				
	rs50872	CC	82	41	39.1	30.7-85.4					
		CT	54	34	27.0	21.1-59.4	0.191				
		TT	5	1	NR	NR-NR					
		C	136	75	32.2	27-52.2	0.570				
		T	59	35	27.0	21.1-59.4	0.115				
	rs238416	AA	19	12	25.8	20.9-NR					
		AG	67	37	34.1	27.7-73.9	0.601				
		GG	55	27	30.0	26.1-NR					
		A	86	49	32.2	25.4-52.2	0.502				
		G	122	64	32.4	27.7-54.3	0.357				
ERCC5	rs17655	CC	79	42	30.0	25.4-52.2					
		CG	52	29	34.1	24.5-85.6	0.514				
		GG	10	5	59.4	27.9-NR					
		C	131	71	32.0	26.1-48.4	0.286				
		G	62	34	43.1	27.0-85.6	0.437				
	rs1047768	CC	43	23	27.7	24.2-NR					
		CT	70	36	32.0	25.8-60.7	0.970				
		TT	28	17	39.1	24.5-NR					
		C	113	59	32.0	26.1-48.4	0.856				
		T	98	53	32.2	27.9-59.4	0.827				
IL1B	rs12621220	CC	73	42	31.1	23.1-54.3					
		CT	61	32	41.8	26.9-85.4	0.716				
		TT	7	2	27.0	17.1-NR					
		C	134	74	32.2	26.9-52.2	0.415				
		T	68	34	41.8	27.0-85.4	0.820				
	rs1143623	CC	7	2	27.0	17.1-NR					
		CG	61	32	41.8	26.9-85.4	0.716				
		GG	73	42	31.1	23.1-54.3					
		C	68	34	41.8	27.0-85.4	0.820				
		G	134	74	32.2	26.9-52.2	0.415				

Table 28. (Continued).

Gene	SNPs	Genotype	N	OS							
				Events	MST (mo)	95% CI	Log Rank p-value	Ref. Cat.	Univariate Cox Model		
									HR	95% CI	p-value
IL1B	rs16944	AA	14	7	21.1	11.4-NR					
		AG	71	39	34.1	27.7-60.7	0.881				
		GG	56	30	32.0	24.5-126.4					
		A	85	46	32.4	26.9-60.7	0.619				
		G	127	69	32.2	27.7-52.2	0.841				
	rs1143627	CC	15	7	27.0	11.4-NR					
		CT	70	39	34.1	26.9-60.7	0.882				
		TT	56	30	32.0	24.5-126.4					
		C	85	46	32.4	26.9-60.7	0.619				
		T	126	69	32.2	27.7-52.2	0.929				
IL6	rs1800795	CC	17	10	32.4	20.9-NR					
		CG	69	38	32.2	25.8-60.7	0.721				
		GG	55	28	30.0	24.5-NR					
		C	86	48	32.4	25.8-54.3	0.473				
		G	124	66	32.2	27.0-54.3	0.567				
IL16	rs7170924	GG	79	46	28.1	24.3-41.8					
		GT	53	24	64.7	27.7-NR	0.072				
		TT	9	6	73.9	16.0-NR					
		G	132	70	32.0	26.9-48.4	0.687				
		T	62	30	64.7	27.7-126.4	0.023	T	1.76	1.08-2.87	0.0240
MDM2	rs1470383	CC	7	4	126.4	27.9-NR					
		CT	35	21	27.7	24.2-NR	0.163				
		TT	99	51	32.4	26.1-73.9					
		C	42	25	28.1	24.5-64.7	0.685				
		T	134	72	32.0	26.1-48.4	0.160				
	rs1690924	AA	56	30	32.2	24.5-73.9					
		AG	65	33	43.1	30.7-85.4	0.222				
		GG	19	12	17.5	15.2-NR					
		A	121	63	39.1	27.9-59.4	0.086	A	1.71	0.92-3.19	0.0903
		G	84	45	32.2	27.7-64.7	0.827				
MTHFR	rs1801131	AA	75	40	28.1	24.5-60.7					
		AC	53	28	32.2	21.4-124.5	0.778				
		CC	13	8	43.1	30.0-NR					
		A	128	68	31.1	25.8-52.2	0.623				
		C	66	36	32.4	26.1-85.4	0.793				
	rs1801133	CC	44	26	32.4	27.7-59.4					
		CT	73	38	32.0	24.5-85.4	0.602				
		TT	24	12	25.8	21.1-NR					
		C	117	64	32.2	27.7-48.4	0.455				
		T	97	50	32.0	24.5-73.9	0.382				
MTR	rs1805087	AA	99	53	32.0	26.9-54.3					
		AG	37	21	28.1	22.7-NR	0.324				
		GG	5	2	124.5	64.7-NR					
		A	136	74	32.0	26.1-48.4	0.135				
		G	42	23	32.2	23.1-NR	0.660				
SLC19A1	rs1051266	AA	35	25	27.9	24.5-43.1					
		AG	64	32	41.8	25.4-85.6	0.392				
		GG	42	19	47.4	24.2-NR					
		A	99	57	30.7	26.1-48.4	0.330				
		G	106	51	47.4	26-64.7	0.204				
XRCC1	rs1799782	CC	120	67	30.0	25.4-41.8					
		CT	20	9	85.4	48.4-NR	0.186				
		TT	1	0	NR	NR-NR					
		C	140	76	32.2	27-52.2	0.495				
		T	21	9	85.4	48.4-NR	0.073	T	1.88	0.93-3.79	0.0777
	rs25487	AA	20	10	31.1	13.8-NR					
		AG	71	41	26.9	21.4-47.4	0.456				
		GG	50	25	48.4	30.7-NR					
		A	91	51	27.7	22.7-43.1	0.229				
		G	121	66	32.2	26.9-54.3	0.500				

N: number of patients; MST: median survival time (months); NR: not reached; Ref Cat: reference category; HR: hazard ratio; 95% CI: 95% confidence interval.

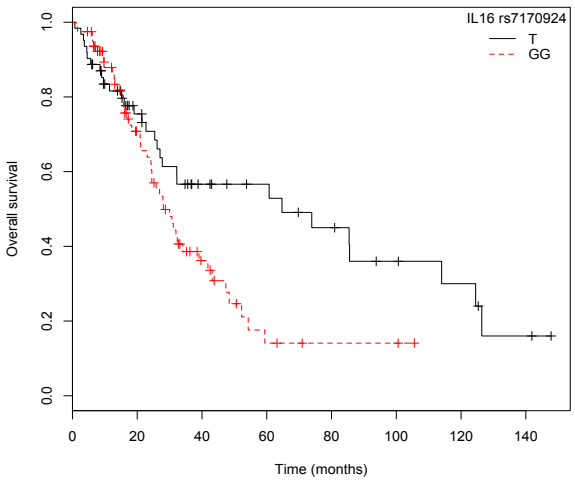

Figure 14. Kaplan-Meier curve for overall survival according to IL16 rs7170924-T allele.

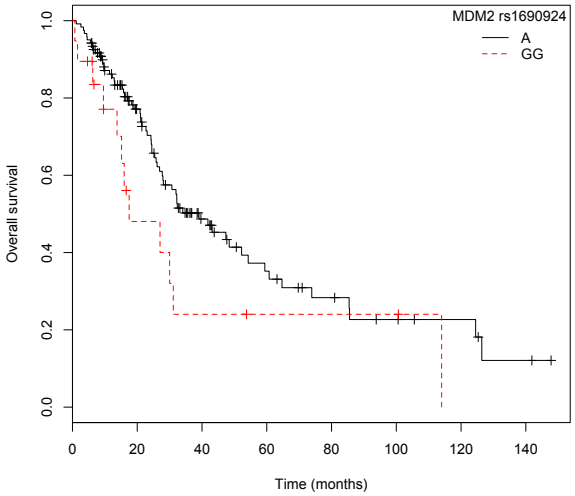

Figure 15. Kaplan-Meier curve for overall survival according to MDM2 rs1690924-A allele.

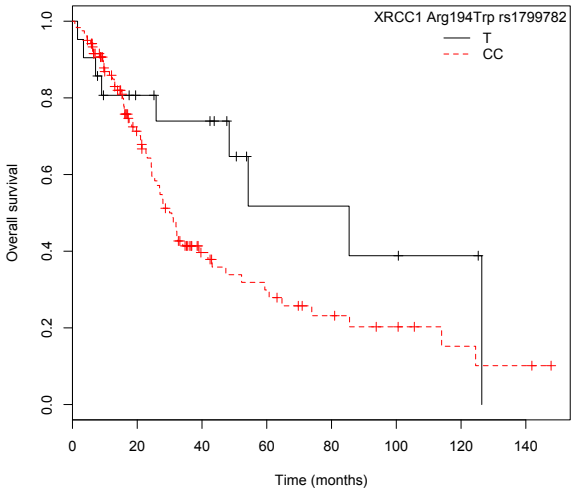

Figure 16. Kaplan-Meier curve for overall survival according to XRCC1 Arg194Trp-T allele.

Table 29. Influence of clinical characteristic and gene polymorphisms on overall survival.

	Overall survival	
	HR ($CI_{95\%}$)	**p-value**
Gender (Male)	3.21 (1.72-6.00)	0.0003
Histology (Adenocarcinoma)	2.21 (1.17-4.15)	0.0142
Chemotherapy agents (Gemcitabine and Pemetrexed)	2.35 (1.36-4.08)	0.0023
Surgery (No)	5.93 (2.79-12.59)	<0.001
MDM2 rs1690924-GG	1.99 (1.05-3.80)	0.0345

HR: hazard ratio; $CI_{95\%}$: 95% confidence interval.

5.4.2.2 Progression-free survival

No associations were demonstrated in the bivariate analysis between polymorphisms and PFS (Table 30). However, the A-allele for IL1B rs16944 ($p_{log\text{-}rank}$=0.085; Table 30) (Figure 17), the C-allele for IL1B rs1143627 ($p_{log\text{-}rank}$=0.085; Table 30) (Figure 18), the GG genotype for IL16 rs7170924 ($p_{log\text{-}rank}$=0.065; Table 30) (Figure 19), the A-allele for MTR rs1805087 ($p_{log\text{-}rank}$=0.106; Table 30) (Figure 20) and the AA genotype for SLC19A1 rs1051266 polymorphisms ($p_{log\text{-}rank}$=0.127; Table 30) (Figure 21) presented a trend to higher risk to progression. Median PFS for carriers of IL1B rs16944-AA genotype was 7.7 ($CI_{95\%}$=6.0-NR), whereas for AG and GG genotype was 10.9 ($CI_{95\%}$=8.4-17.1) and 19.2 ($CI_{95\%}$=15.0-35.7) months, respectively. For IL1B rs1143627, which was in linkage disequilibrium with IL1B rs16944, patients with CC genotype showed a median PFS of 8.0 months ($CI_{95\%}$=6.0-NR), whereas for CC and CT genotypes, the median PFS was 10.3 ($CI_{95\%}$=7.1-17.6) and 19.2 ($CI_{95\%}$=15.0-35.7) months, respectively. Carriers of IL16 rs7170924-GG genotype reported a median PFS of 11.4 months ($CI_{95\%}$=8.5-17.6), and those patients with GT and TT genotypes showed a median PFS of 17.1 ($CI_{95\%}$=10.9-82.3) and 13.2 ($CI_{95\%}$=7.6-NR) months, respectively. Patients with A-allele for MTR rs1805087 polymorphism showed a median PFS of 12.9 months ($CI_{95\%}$=10.2-17.6) versus GG genotype, which revealed a median PFS of 82.3 months ($CI_{95\%}$=82.3-NR). Median PFS for SLC19A1 Arg27His-AA genotype was 10.1 months ($CI_{95\%}$=7.0-20.4), whereas for G-allele the median PFS was 15.0 months ($CI_{95\%}$=10.9-23.2). A multivariate Cox regression model adjusted by surgery and concomitant or concurrent radiotherapy was performed to assess the impact of gene polymorphisms and PFS. IL1B rs16944, MTR rs1805087 and SLC19A1 Arg27His were significantly associated with PFS ($p_{likelihood\ ratio\ test}$= $1.11 \cdot 10^{-16}$; Table 31).

Table 30. Association of gene polymorphisms with progression-free survival.

Gene	SNPs	Genotype	N	PFS							
				Events	MST (mo)	95% CI	Log Rank p-value	Ref. Cat.	Univariate Cox Model		
									HR	95% CI	p-value
ABCB1	rs1045642	CC	49	37	11.9	7.1-20.4					
		CT	62	43	12.9	10.1-24.5	0.699				
		TT	30	20	17.6	8.5-44.0					
		C	111	80	12.8	10.1-18.7	0.617				
		T	92	63	14.6	10.2-19.2	0.412				
	rs1128503	CC	53	40	11.9	9.2-19.6					
		CT	63	41	15.0	10.1-24.5	0.677				
		TT	25	19	13.2	7.6-44.0					
		C	116	81	14.6	10.3-8.7	0.973				
		T	88	60	14.6	10.1-23.3	0.404				
	rs2032582	GG	47	36	14.3	8.4-20.4					
		G(GT/AG)	69	45	14.6	10.1-24.5	0.506				
		T (TT/AT)	25	19	13.2	6.9-39.1					
		G vs TT/AT	116	81	14.3	10.3-18.7	0.625				
		G/T vs GG	94	64	13.2	10.03-23.3	0.394				
ERCC1	rs11615	CC	20	11	16.1	6.5-NR					
		CT	72	51	15.5	11.0-27.3	0.335				
		TT	49	38	10.2	7.0-17.6					
		C	92	62	16.1	11.2-24.5	0.159				
		T	121	89	12.9	10.2-18.4	0.416				
	rs3212986	GG	74	57	12.8	9.2-18.4					
		GT	56	36	15.0	10.9-24.5	0.683				
		TT	11	7	18.7	5.4-NR					
		G	130	93	12.9	10.2-18.4	0.502				
		T	67	43	16.1	11.0-24.5	0.453				
ERCC2	rs13181	GG	25	16	15.0	6.5-NR					
		GT	55	39	12.9	9.2-30.6	0.735				
		TT	61	45	14.3	10.0-19.2					
		G	80	55	12.9	9.2-23.3	0.435				
		T	116	84	13.2	10.2-18.4	0.832				
	rs1799793	AA	24	15	18.7	7.7-NR					
		AG	55	42	12.8	9.1-23.2	0.575				
		GG	62	43	13.2	10.0-19.2					
		A	79	57	15.0	10.0-19.2	0.577				
		G	117	85	12.8	10.1-17.6	0.299				
	rs50872	CC	82	59	12.9	10.1-19.2					
		CT	54	39	14.6	7.5-24.5	0.791				
		TT	5	2	NR	4.3-NR					
		C	136	98	14.3	10.2-18.4	0.704				
		T	59	41	14.6	7.5-24.5	0.636				
	rs238416	AA	19	16	12.8	8.4-NR					
		AG	67	49	14.6	10.3-19.2	0.655				
		GG	55	35	11.2	7.7-36.1					
		A	86	65	14.6	10.3-19.2	0.598				
		G	122	84	14.3	10.2-18.7	0.380				
ERCC5	rs17655	CC	79	56	11.4	9.2-18.7					
		CG	52	38	13.2	8.0-30.6	0.344				
		GG	10	6	19.2	10.1-NR					
		C	131	94	12.9	10.2-17.1	0.166				
		G	62	44	16.1	10.0-30.6	0.362				
	rs1047768	CC	43	31	11.2	8.5-27.3					
		CT	70	49	16.1	10.3-20.4	0.832				
		TT	28	20	14.3	7.00-NR					
		C	113	80	13.2	10.2-18.7	0.852				
		T	98	69	15.0	10.3-19.2	0.545				
IL1B	rs12621220	CC	73	52	16.8	11.0-24.5					
		CT	61	45	10.3	7.0-18.4	0.452				
		TT	7	3	10.9	7.5-NR					
		C	134	97	14.3	10.2-18.4	0.463				
		T	68	48	10.9	7.5-18.4	0.416				
	rs1143623	CC	7	3	10.9	7.5-NR					
		CG	61	45	10.3	7.0-18.4	0.452				
		GG	73	52	16.8	11.0-24.5					
		C	68	48	10.9	7.5-18.4	0.416				
		G	134	97	14.3	10.2-8.4	0.463				

Table 30. (Continued).

Gene	SNPs	Genotype	N	PFS							
				Events	MST (mo)	95% CI	Log Rank p-value	Ref. Cat.	Univariate Cox Model		
									HR	95% CI	p-value
IL1B	rs16944	AA	14	9	7.7	6.0-NR					
		AG	71	54	10.9	8.4-17.1	0.225				
		GG	56	37	19.2	15.0-35.7					
		A	85	63	10.2	7.7-16.1	0.085	GG	1.43	0.95-2.15	0.0859
		G	127	91	15.0	11.0-19.2	0.718				
	rs1143627	CC	15	10	8.0	6.0-NR					
		CT	70	53	10.3	7.1-17.6	0.226				
		TT	56	37	19.2	15.0-35.7					
		C	85	63	10.2	7.7-16.1	0.085	TT	1.43	0.95-2.15	0.0859
		T	126	90	15.0	11.0-19.2	0.617				
IL6	rs1800795	CC	17	13	11.9	6.5-NR					
		CG	69	52	15.0	10.0-19.6	0.499				
		GG	55	35	14.3	10.0-44.0					
		C	86	65	12.9	10.0-19.2	0.248				
		G	124	87	14.6	10.2-19.2	0.562				
IL16	rs7170924	GG	79	60	11.4	8.5-17.6					
		GT	53	32	17.1	10.9-82.3	0.164				
		TT	9	8	13.2	7.6-NR					
		G	132	92	14.3	10.2-8.4	0.901				
		T	62	40	17.1	10.9-44.0	0.065	T	1.47	0.98-2.21	0.0657
MDM2	rs1470383	CC	7	5	39.1	17.1-NR					
		CT	35	26	11.0	9.2-19.2	0.275				
		TT	99	69	12.9	10.0-19.6					
		C	42	31	16.9	10.3-34.4	0.983				
		T	134	95	12.8	10.1-17.6	0.161				
	rs1690924	AA	56	44	16.9	10.2-23.3					
		AG	65	42	14.3	10.2-30.6	0.518				
		GG	19	13	7.0	6.0-NR					
		A	121	86	15.5	10.9-19.2	0.502				
		G	84	55	11.4	9.1-23.2	0.485				
MTHFR	rs1801131	AA	75	54	11.4	8.5-19.2					
		AC	53	37	14.3	10.0-19.6	0.419				
		CC	13	9	38.4	15.0-NR					
		A	128	91	11.9	10.0-17.6	0.189				
		C	66	46	16.1	10.9-30.6	0.523				
	rs1801133	CC	44	35	15.0	8.0-19.6					
		CT	73	49	16.9	10.1-24.5	0.530				
		TT	24	16	10.2	7.0-NR					
		C	117	84	15.0	10.9-19.2	0.920				
		T	97	65	12.9	10.1-23.2	0.270				
MTR	rs1805087	AA	99	72	13.2	10.1-19.2					
		AG	37	26	11.9	6.5-30.6	0.252				
		GG	5	2	82.3	82.3-NR					
		A	136	98	12.9	10.2-17.6	0.106	GG	3.04	0.74-12.48	0.124
		G	42	28	14.3	8.5-63.1	0.359				
SLC19A1	rs1051266	AA	35	30	10.1	7.0-20.4					
		AG	64	44	14.3	9.2-24.5	0.237				
		GG	42	26	17.1	10.2-39.1					
		A	99	74	11.4	9.2-18.7	0.206				
		G	106	70	15.0	10.9-23.2	0.127	G	1.39	0.91-2.14	0.128
XRCC1	rs1799782	CC	120	86	11.9	10.0-17.1					
		CT	20	13	17.6	11.0-NR	0.371				
		TT	1	1	18.7	NR-NR					
		C	140	99	13.2	10.2-18.4	0.942				
		T	21	14	18.7	11.0-NR	0.173				
	rs25487	AA	20	13	14.6	5.4-NR					
		AG	71	49	13.2	10.0-24.5	0.766				
		GG	50	38	16.8	9.2-23.2					
		A	91	62	13.2	10.1-19.2	0.886				
		G	121	87	13.2	10.2-19.2	0.514				

N: number of patients; MST: median survival time (months); NR: not reached; Ref Cat: reference category; HR: hazard ratio; 95% CI: 95% confidence interval.

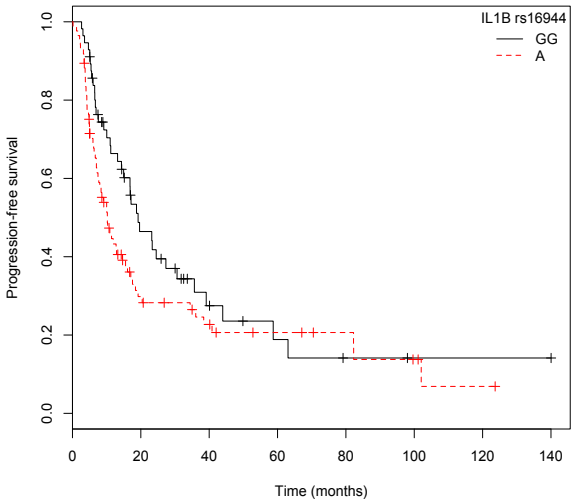

Figure 17. Kaplan-Meier curve for progression-free survival according to IL1B rs16944-A allele.

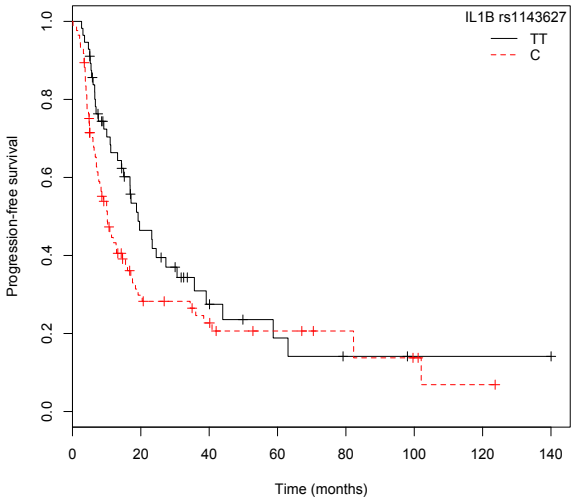

Figure 18. Kaplan-Meier curve for progression-free survival according to IL1B rs1143627-C allele.

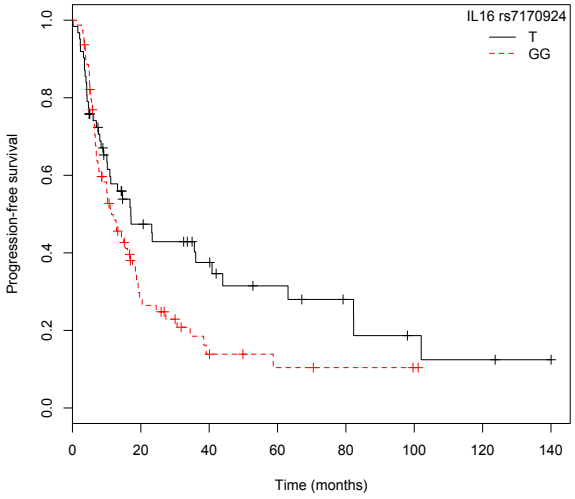

Figure 19. Kaplan-Meier curve for progression-free survival according to IL16 rs7170924-T allele.

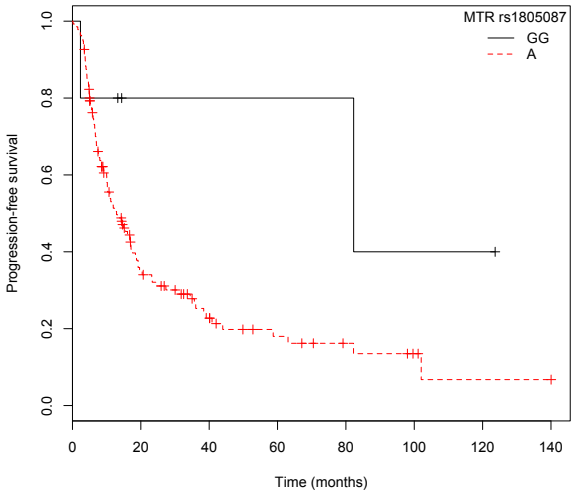

Figure 20. Kaplan-Meier curve for progression-free survival according to MTR rs1805087-A allele.

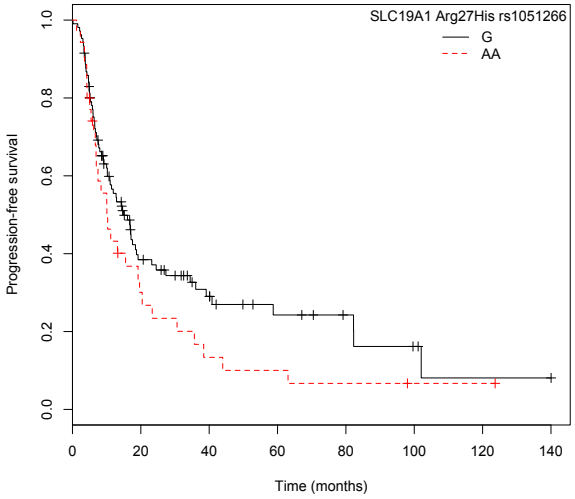

Figure 21. Kaplan-Meier curve for progression-free survival according to SLC19A1 rs1051266-G allele.

Table 31. Influence of clinical characteristic and gene polymorphisms on progression-free survival.

	Progression free-survival	
	HR ($CI_{95\%}$)	**p-value**
Surgery (No)	11.99 (5.83-24.66)	<0.001
Radiotherapy (No)	3.06 (1.88-4.97)	<0.001
IL1B rs16944- A allele	1.68 (1.11-2.54)	0.0141
MTR rs1805087-A allele	12.19 (2.57-57.71)	0.0016
SLC19A1 Arg27His-AA	1.75 (1.13-2.70)	0.0116

HR: hazard ratio; $CI_{95\%}$: 95% confidence interval.

5.4.3 Toxicity

5.4.3.1 General Toxicity

ERCC1 C118T, ERCC2 rs50872 and ABCB1 C1236T were associated with general toxicity. Patients with CT/TT genotypes for ERCC1 C118T (p=0.0186; RR=6.78; $CI_{95\%}$=1.38-33.4; Table 32), CC for ERCC2 rs50872 (p=0.0234; RR=2.03; $CI_{95\%}$=1.10-3.74; Table 32) or TT for ABCB1 C1236T (p=0.0148; RR=2.08; $CI_{95\%}$=1.15-3.75; Table 32) were in higher risk of grade 3-4 toxicity. Logistic regression analysis adjusted by chemotherapy agents and previous non-lung cancer revealed that ERCC1 C118T-T allele and ERCC2 rs50872-CC genotype were the only genetic independent factors associated with higher general toxicity ($p_{likelihood\ ratio\ test}=1.898\cdot10^{-7}$; Table 33).

ERCC2 Asp312Asn and IL1B polymorphisms were associated with multiple adverse events (>1). In particular, ERCC2 Asp312Asn (p=0.0378; Table 34), IL1B rs12621220-CT/TT (p=0.0163; RR=1.35; $CI_{95\%}$=1.06-1.72; Table 34), IL1B rs1143623-CG/CC (p=0.0163; RR=1.35; $CI_{95\%}$=1.06-1.72; Table 34), IL1B rs16944 (p=0.0298; Table 34) or IL1B rs1143627 (p=0.0450; Table 34) were in risk of experiencing more than one adverse event. A logistic regression analysis revealed that the AG genotype for ERCC2 Asp312Asn and CT/TT for IL1B-rs12621220 were independently associated with the occurrence of more than one adverse event ($p_{likelihood\ ratio\ test}=0.0009103$; Table 36). Similarly, carriers of ABCB1 C1236T-TT genotype (p=0.0097; RR=2.32; $CI_{95\%}$=1.23-4.39; Table 35), ERCC2 Asp312Asn (p=0.0464; Table 35) or XRCC1 Gln399Arg-AG/GG genotype (p=0.0459; RR=5.79; $CI_{95\%}$=1.03-32.47; Table 35), showed higher risk of experiencing more than two adverse events. Logistic regression analysis revealed that the G-allele of ERCC2 Asp312Asn and the TT genotype of ABCB1 C1236T were the only independent factors associated with higher risk of more than two adverse events ($p_{likelihood\ ratio\ test}=0.0007531$; Table 36).

5.4.3.2 Hematological Toxicity

ERCC2 rs50872 and IL16 rs7170924 presented influence on hematological toxicity (Table 36). In fact, patients carrying the CC genotype for ERCC2 rs50872 (p=0.0223; RR=2.34; $CI_{95\%}$=1.13-4.85; Table 37) or IL16 rs7170924 T-allele (p=0.0277; RR=2.06; $CI_{95\%}$=1.08-3.92; Table 37) presented higher grade of hematological toxicity. A multivariate logistic regression analysis adjusted by chemotherapy agents was used to further investigate the impact of ERCC2 rs50872 and IL16 rs7170924 on hematological toxicity. Both remaining significantly associated to grade 3-4 hematological toxicity ($p_{likelihood\ ratio\ test}=6.861\cdot10^{-5}$; Table 38).

Table 32. Association of gene polymorphisms with general toxicity.

Gene	SNPs	Genotype	N	Toxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
ABCB1	rs1045642	CC	49	17 (34.7)	32 (65.3)					
		CT	62	15 (24.2)	47 (75.8)	1.672	0.4334			
		TT	30	10 (33.3)	20 (66.7)					
		C	111	32 (28.8)	79 (71.2)	0.0644	0.7997			
		T	92	25 (27.2)	67 (72.8)	0.5423	0.4615			
	rs1128503	CC	53	16 (30.2)	37 (69.8)					
		CT	63	13 (20.6)	50 (79.4)	8.4252	0.0148			
		TT	25	13 (52.0)	12 (48.0)					
		C	116	29 (25)	87 (75)	5.9362	0.0148	C	2.08	1.15-3.75
		T	88	26 (29.5)	62 (70.5)	0	1			
	rs2032582	GG	47	15 (31.9)	32 (68.1)					
		G (GT/AG)	69	16 (23.2)	53 (76.8)	3.953	0.1386			
		T (TT/AT)	25	11 (44.0)	14 (56.0)					
		G vs TT/AT	116	31 (26.7)	85 (73.3)	2.1671	0.1410			
		G/T vs GG	94	27 (28.7)	67 (71.3)	0.0381	0.8451			
ERCC1	rs11615	CC	20	1 (5.0)	19 (95.0)					
		CT	72	26 (35.1)	46 (63.9)	7.2681	0.0264			
		TT	49	15 (30.6)	34 (69.4)					
		C	92	27 (29.3)	65 (70.7)	0	1			
		T	121	41 (33.9)	80 (66.1)	5.5352	0.0186	CC	6.78	1.38-33.4
	rs3212986	GG	74	20 (27.0)	54 (73.0)					
		GT	56	20 (35.7)	36 (64.3)	1.9186	0.3832			
		TT	11	2 (18.2)	9 (81.8)					
		G	130	40 (30.8)	90 (69.2)		0.5061*			
		T	67	22 (32.8)	45 (67.2)	0.3236	0.5695			
ERCC2	rs13181	GG	25	5 (20.0)	20 (80.0)					
		GT	55	16 (29.1)	39 (70.9)	1.7854	0.4095			
		TT	61	21 (34.4)	40 (65.6)					
		G	80	21 (26.2)	59 (73.8)	0.7499	0.3865			
		T	116	37 (31.9)	79 (68.1)	0.8811	0.3479			
	rs1799793	AA	24	6 (25.0)	18 (75.0)					
		AG	55	18 (32.7)	37 (67.3)	0.5072	0.776			
		GG	62	18 (29.0)	44 (71.0)					
		A	79	24 (30.4)	55 (69.6)	0	1			
		G	117	36 (30.8)	81 (69.2)	0.1011	0.7505			
	rs50872	CC	82	31 (37.8)	51 (62.2)					
		CT	54	11 (20.4)	43 (79.6)		0.0318*			
		TT	5	0 (0.0)	5 (100.0)					
		C	136	42 (30.9)	94 (69.1)		0.3219*			
		T	59	11 (18.6)	48 (81.4)	5.1419	0.0234	T	2.03	1.10-3.74
	rs238416	AA	19	7 (36.8)	12 (63.2)					
		AG	67	20 (29.9)	47 (70.1)	0.6186	0.734			
		GG	55	15 (27.3)	40 (72.7)					
		A	86	27 (31.4)	59 (68.6)	0.1111	0.7389			
		G	122	35 (28.7)	87 (71.3)	0.2054	0.6504			
ERCC5	rs17655	CC	79	25 (31.6)	54 (68.4)					
		CG	52	16 (30.8)	36 (69.2)	2.0265	0.363			
		GG	10	1 (10.0)	9 (90.0)					
		C	131	41 (31.3)	90 (68.7)		0.2814*			
		G	62	17 (27.4)	45 (72.6)	0.129	0.7195			
	rs1047768	CC	43	13 (30.2)	30 (69.8)					
		CT	70	23 (32.9)	47 (67.1)	1.2549	0.534			
		TT	28	6 (21.4)	22 (78.6)					
		C	113	36 (31.9)	77 (68.1)	0.7217	0.3956			
		T	98	29 (29.6)	69 (70.4)	0	1			
IL1B	rs12621220	CC	73	51 (69.9)	22 (30.1)					
		CT	61	42 (68.9)	19 (31.1)		0.8213*			
		TT	7	6 (85.7)	1 (14.3)					
		C	134	93 (69.4)	41 (30.6)		0.6741*			
		T	68	48 (70.6)	20 (29.4)	0.0089	0.9250			
	rs1143623	CC	7	6 (85.7)	1 (14.3)					
		CG	61	42 (68.9)	19 (31.1)		0.8213*			
		GG	73	51 (69.9)	22 (30.1)					
		C	68	48 (70.6)	20 (29.4)	0.0089	0.9250			
		G	134	93 (69.4)	41 (30.6)		0.6741*			

Table 32. (Continued).

Gene	SNPs	Genotype	N	Toxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
IL1B	rs16944	AA	14	11 (78.6)	3 (21.4)					
		AG	71	49 (69.0)	22 (31.0)	0.5252	0.7691			
		GG	56	39 (69.6)	17 (30.4)					
		A	85	60 (70.6)	25 (29.4)	0.0144	0.9044			
		G	127	88 (69.3)	39 (30.7)		0.5550*			
	rs1143627	CC	15	11 (73.3)	4 (26.7)					
		CT	70	49 (70.0)	21 (30.0)	0.0801	0.9608			
		TT	56	39 (69.6)	17 (30.4)					
		C	85	60 (70.6)	25 (29.4)	0.0144	0.9044			
		T	126	88 (69.8)	38 (30.2)		1*			
IL6	rs1800795	CC	17	5 (29.4)	12 (70.6)					
		CG	69	20 (29.0)	49 (71.0)	0.0554	0.9727			
		GG	55	17 (30.9)	38 (69.1)					
		C	86	25 (29.1)	61 (70.9)	0.002	0.9648			
		G	124	37 (29.8)	87 (70.2)	0	1			
IL16	rs7170924	GG	79	19 (24.1)	60 (75.9)					
		GT	53	20 (37.7)	33 (62.3)	2.8982	0.2348			
		TT	9	3 (33.3)	6 (66.7)					
		G	132	39 (29.5)	93 (70.5)		1*			
		T	62	23 (37.1)	39 (62.9)	2.2376	0.1347			
MDM2	rs1470383	CC	7	1 (14.3)	6 (85.7)					
		CT	35	10 (28.6)	25 (71.4)		0.7600*			
		TT	99	31 (31.3)	68 (68.7)					
		C	42	11 (26.2)	31 (73.8)	0.1656	0.6840			
		T	134	41 (30.6)	93 (69.4)		0.6741*			
	rs1690924	AA	56	18 (32.1)	38 (67.9)					
		AG	65	19 (29.2)	46 (70.8)	0.2636	0.8765			
		GG	19	5 (26.3)	14 (73.7)					
		A	121	37 (30.6)	84 (69.4)	0.0116	0.9142			
		G	84	24 (28.6)	60 (71.4)	0.0694	0.7921			
MTHFR	rs1801131	AA	75	21 (28.0)	54 (72.0)					
		AC	53	17 (32.1)	36 (67.9)	0.2532	0.8811			
		CC	13	4 (30.8)	9 (69.2)					
		A	128	38 (29.7)	90 (70.3)		1*			
		C	66	21 (31.8)	54 (68.2)	0.0962	0.7564			
	rs1801133	CC	44	13 (29.5)	31 (70.5)					
		CT	73	22 (30.1)	51 (69.9)	0.0099	0.9951			
		TT	24	7 (29.2)	17 (70.8)					
		C	117	35 (29.9)	82 (70.1)	0	1			
		T	97	29 (29.5)	68 (70.1)	0	1			
MTR	rs1805087	AA	99	73 (73.7)	26 (26.3)					
		AG	37	23 (62.2)	14 (37.8)		0.3619*			
		GG	5	3 (60.0)	2 (40.0)					
		A	136	96 (70.6)	40 (29.4)		0.6343*			
		G	42	26 (61.9)	16 (38.1)	1.4489	0.2287			
SLC19A1	rs1051266	AA	35	9 (25.7)	26 (74.3)					
		AG	64	19 (29.7)	45 (70.3)	0.5304	0.7670			
		GG	42	14 (33.3)	28 (66.7)					
		A	99	28 (28.3)	71 (71.7)	0.1587	0.6903			
		G	106	33 (31.1)	73 (68.9)	0.1557	0.6932			
XRCC1	rs1799782	CC	20	5 (25.0)	15 (75.0)					
		CT	71	21 (29.6)	50 (70.4)	0.3377	0.8446			
		TT	50	16 (32.0)	34 (68.0)					
		C	91	26 (28.6)	65 (71.4)	0.0545	0.8154			
		T	121	37 (30.6)	84 (69.4)	0.0583	0.8092			
	rs25487	AA	120	39 (32.5)	81 (67.5)					
		AG	20	3 (15.0)	17 (85.0)		0.2586*			
		GG	1	0 (0.0)	1 (100.0)					
		A	140	42 (30.0)	98 (70.0)		1*			
		G	21	3 (14.3)	18 (85.7)	2.031	0.1541			

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 33. Influence of clinical characteristic and gene polymorphisms on general toxicity.

	General Toxicity	
	OR ($CI_{95\%}$)	**p-value**
Chemotherapy agents		
Gemcitabine (Reference)		
Paclitaxel	0.13 (0.02-0.52)	0.00587
Pemetrexed	0.10 (0.02-0.38)	0.00147
Vinorelbine	0.27 (0.07-0.91)	0.04098
Personal history of cancer (No)	11.57 (2.09-217.62)	0.02245
ERCC1 C118T-CT/TT	26.05 (4.33-515.77)	0.00345
ERCC2 rs50872-CC	4.06 (1.66-10.65)	0.00291

OR: odds ratio; $CI_{95\%}$: 95% confidence interval

5.4.3.3 Other toxicity subtypes

The bivariate analysis showed that higher grade of asthenia was associated to IL16 rs7170924 (TT>GT>GG) polymorphism (p=0.0377; Table 39). No association was found for other genetic or clinical variables. ERCC5 His46His and SLC19A1 Arg27His were associated with gastrointestinal toxicity. Particularly, patients carrying the CC genotype for ERCC5 His46His (p=0.0269; Table 40) or AA for SLC19A1 Arg27His (p=0.0143; Table 40) showed greater risk of gastrointestinal toxicity. No association was reached on multivariate logistic regression model. ERCC2 rs238416 was the only polymorphism that showed influence on grade 3-4 infection. Particularly, carriers of AA genotype showed greater risk of grade 3-4 infection to those carrying the G-allele (p=0.0177; RR=9.63; $CI_{95\%}$=1.56-59.6; Table 41). No multivariate regression analysis was investigated. The bivariate analysis revealed that IL6 rs1800795-CC, MTR rs1805087-AG/GG and XRCC1 Gln399Arg-GG genotypes were associated with severe nephrotoxicity (p=0.0384 for IL6 rs1800795, p=0.0251 for MTR rs1805087 and p=0.0429 for XRCC1 Gln399Arg) (Table 42). However, multivariate logistic regression analysis did not revealed influence of these polymorphisms on severe nephrotoxicity. The only polymorphism associated with grade 3-4 neurotoxicity was MDM2 rs1470383, particularly the CC genotype (p=0.0497; Table 43). No multivariate regression analysis was investigated.

Table 34. Association of gene polymorphisms with more than one adverse events.

Gene	SNPs	Genotype	N	Toxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				>1 N (%)	0-1 N (%)					
ABCB1	rs1045642	CC	49	32 (65.3)	17 (34.7)					
		CT	62	41 (66.1)	21 (33.9)	0.6237	0.7321			
		TT	30	22 (73.3)	8 (26.7)					
		C	111	73 (65.8)	38 (34.2)	0.3192	0.5721			
		T	92	63 (68.5)	29 (31.5)	0.0376	0.8462			
	rs1128503	CC	53	31 (58.5)	22 (41.5)					
		CT	63	46 (73.0)	17 (27.0)	3.0585	0.2167			
		TT	25	18 (72.0)	7 (28.0)					
		C	116	77 (66.4)	39 (33.6)	0.0952	0.7577			
		T	88	64 (72.7)	24 (27.3)	2.4368	0.1185			
	rs2032582	GG	47	30 (63.8)	17 (36.2)					
		G (GT/AG)	69	49 (71.0)	20 (29.0)	0.8141	0.6656			
		T (TT/AT)	25	16 (64.0)	9 (36.0)					
		G vs TT/AT	116	79 (68.1)	37 (31.9)	0.0262	0.8715			
		G/T vs GG	94	65 (69.1)	29 (30.9)	0.1976	0.6566			
ERCC1	rs11615	CC	20	12 (60.0)	8 (40.0)					
		CT	72	47 (65.3)	25 (34.7)	1.4669	0.4802			
		TT	49	36 (73.5)	13 (26.5)					
		C	92	59 (64.1)	33 (35.9)	0.8793	0.3484			
		T	121	83 (68.6)	38 (31.4)	0.2521	0.6156			
	rs3212986	GG	74	52 (70.3)	22 (29.7)					
		GT	56	35 (62.5)	21 (37.5)	1.0310	0.5972			
		TT	11	8 (72.7)	3 (27.3)					
		G	130	87 (66.9)	43 (33.1)		1*			
		T	67	43 (64.2)	24 (35.8)	0.3488	0.5548			
ERCC2	rs13181	GG	25	14 (56.0)	11 (44.0)					
		GT	55	40 (72.7)	15 (27.3)	2.1892	0.3347			
		TT	61	41 (67.2)	20 (32.8)					
		G	80	54 (67.5)	26 (32.5)	0	1			
		T	116	81 (69.8)	35 (30.2)	1.2153	0.2703			
	rs1799793	AA	24	14 (58.3)	10 (41.7)					
		AG	55	44 (80.0)	11 (20.0)	6.5522	0.0378			
		GG	62	37 (59.7)	25 (40.3)					
		A	79	58 (73.4)	21 (26.6)	2.3913	0.1220			
		G	117	81 (69.2)	36 (30.8)	0.6373	0.4247			
	rs50872	CC	82	55 (67.1)	27 (32.9)					
		CT	54	37 (68.5)	17 (31.5)		0.9501*			
		TT	5	3 (60.0)	2 (40.0)					
		C	136	92 (67.6)	44 (32.4)		0.6612*			
		T	59	40 (67.8)	19 (32.2)	0	1			
	rs238416	AA	19	12 (63.2)	7 (36.8)					
		AG	67	46 (68.7)	21 (31.3)	0.2041	0.9030			
		GG	55	37 (67.3)	18 (32.7)					
		A	86	58 (67.4)	28 (32.6)	0	1			
		G	122	83 (68.0)	39 (32.0)	0.0251	0.8740			
ERCC5	rs17655	CC	79	50 (63.3)	29 (36.7)					
		CG	52	37 (71.2)	15 (28.8)	1.6624	0.4355			
		GG	10	8 (80.0)	2(20.0)					
		C	131	87 (66.4)	44 (33.6)		0.4986*			
		G	62	45 (72.6)	17 (27.4)	0.9739	0.3237			
	rs1047768	CC	43	28 (65.1)	15 (34.9)					
		CT	70	49 (70.0)	21 (30.0)	0.4408	0.8022			
		TT	28	18 (64.3)	10 (35.7)					
		C	113	77 (68.1)	36 (31.9)	0.0270	0.8694			
		T	98	67 (68.4)	31 (31.6)	0.0339	0.8540			
IL1B	rs12621220	CC	73	42 (57.5)	31 (42.5)					
		CT	61	48 (78.7)	13 (21.3)		0.0241*			
		TT	7	5 (71.4)	2 (28.6)					
		C	134	90 (67.2)	44 (32.8)		1*			
		T	68	53 (77.9)	15 (22.1)	5.7739	0.0163	CC	1.35	1.06- 1.72
	rs1143623	CC	7	5 (71.4)	2 (28.6)					
		CG	61	48 (78.7)	13 (21.3)	6.8206	0.0330			
		GG	73	42 (57.5)	31 (42.5)					
		C	68	53 (77.9)	15 (22.1)	5.7739	0.0163	GG	1.35	1.06- 1.72
		G	134	90 (67.2)	44 (32.8)		1*			

Table 34. (Continued).

Gene	SNPs	Genotype	N	Toxicity		χ^2	p-value	Cat Ref	RR	95% CI
				>1 N (%)	0-1 N (%)					
IL1B	rs16944	AA	14	7 (50.0)	7 (50.0)					
		AG	71	55 (77.5)	16 (22.5)	7.0287	0.0298			
		GG	56	33 (58.9)	23 (41.1)					
		A	85	62 (72.9)	23 (27.1)	2.4119	0.1204			
		G	127	88 (69.3)	39 (30.7)		0.2274*			
	rs1143627	CC	15	8 (53.3)	7 (46.7)					
		CT	70	54 (77.1)	16 (22.9)	6.2015	0.0450			
		TT	56	33 (58.9)	23 (41.1)					
		C	85	62 (72.9)	23 (27.1)	2.4119	0.1204			
		T	126	87 (69.0)	39 (31.0)		0.2499*			
IL6	rs1800795	CC	17	14 (82.4)	3 (17.6)					
		CG	69	45 (65.2)	24 (34.8)	1.9735	0.3728			
		GG	55	36 (65.5)	19 (34.5)					
		C	86	59 (68.6)	27 (31.4)	0.042	0.8376			
		G	124	81 (65.3)	43 (34.7)	1.274	0.2590			
IL16	rs7170924	GG	79	52 (65.8)	27 (34.2)					
		GT	53	36 (67.9)	17 (32.1)	0.537	0.7645			
		TT	9	7 (77.8)	2 (22.2)					
		G	132	88 (66.7)	44 (33.3)		0.7180*			
		T	62	43 (69.4)	19 (30.6)	0.0692	0.7925			
MDM2	rs1470383	CC	7	6 (85.7)	1 (14.3)					
		CT	35	20 (57.1)	15 (42.9)		0.2545*			
		TT	99	69 (69.7)	30 (30.3)					
		C	42	26 (61.9)	16 (38.1)	0.4987	0.4801			
		T	134	89 (66.4)	45 (33.6)		0.4269*			
	rs1690924	AA	56	39 (69.6)	17 (30.4)					
		AG	65	42 (64.6)	23 (35.4)	0.6908	0.7079			
		GG	19	14 (73.7)	5 (26.3)					
		A	121	81 (66.9)	40 (33.1)	0.102	0.7484			
		G	84	56 (66.7)	28 (33.3)	0.0341	0.8535			
MTHFR	rs1801131	AA	75	51 (68.0)	24 (32.0)					
		AC	53	36 (67.9)	17 (32.1)	0.2221	0.8949			
		CC	13	8 (61.5)	5 (38.5)					
		A	128	87 (68.0)	41 (32.0)		0.7574*			
		C	66	44 (66.7)	22 (33.3)	0	1			
	rs1801133	CC	44	30 (68.2)	14 (31.8)					
		CT	73	48 (65.8)	25 (34.2)	0.2309	0.8909			
		TT	24	17 (70.8)	7 (29.2)					
		C	117	78 (66.7)	39 (33.3)	0.0248	0.8748			
		T	97	65 (67.0)	32 (33.0)	0	1			
MTR	rs1805087	AA	99	67 (67.7)	32 (32.3)					
		AG	37	26 (70.3)	11 (29.7)		0.4537*			
		GG	5	2 (40.0)	3 (60.0)					
		A	136	93 (68.4)	43 (31.6)		0.3298*			
		G	42	28 (66.7)	14 (33.3)	0	1			
SLC19A1	rs1051266	AA	35	24 (68.6)	11 (31.4)					
		AG	64	40 (62.5)	24 (37.5)	1.5059	0.4710			
		GG	42	31 (73.8)	11 (26.2)					
		A	99	64 (64.6)	35 (35.4)	0.7481	0.3871			
		G	106	71 (67.0)	35 (33.0)	0	1			
XRCC1	rs1799782	CC	120	81 (67.5)	39 (32.5)					
		CT	20	13 (65.0)	7 (35.0)		0.8670*			
		TT	1	1 (100.0)	0 (0.0)					
		C	140	94 (67.1)	46 (32.9)		1*			
		T	21	14 (66.7)	7 (33.3)	0	1			
	rs25487	AA	20	13 (65.0)	7 (35.0)					
		AG	71	44 (62.0)	27 (38.0)	2.6865	0.2610			
		GG	50	38 (76.0)	12 (24.0)					
		A	91	57 (62.6)	34 (37.4)	2.0487	0.1523			
		G	121	82 (67.8)	39 (32.2)	0	1			

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 35. Association of gene polymorphisms with more than two adverse events.

Gene	SNPs	Genotype	N	Toxicity >2 N (%)	Toxicity 0-2 N (%)	χ^2	p-value	Ref. Cat.	RR	95% CI
ABCB1	rs1045642	CC	49	9 (18.4)	40 (81.6)					
		CT	62	17 (27.4)	45 (72.6)	2.3994	0.3013			
		TT	30	10 (33.3)	20 (66.7)					
		C	111	26 (23.4)	85 (76.6)	0.7543	0.3851			
		T	92	27 (29.3)	65 (70.7)	1.4911	0.2221			
	rs1128503	CC	53	9 (17.0)	44 (83.0)					
		CT	63	15 (23.8)	48 (76.2)	8.7742	0.0124			
		TT	25	12 (48.0)	13 (52.0)					
		C	116	24 (20.7)	92 (79.3)	6.6958	0.0097	C	2.32	1.23-4.39
		T	88	27 (30.7)	61 (69.3)	2.5848	0.1079			
	rs2032582	GG	47	8 (17.0)	39 (83.0)					
		G (GT/AG)	69	19 (27.5)	50 (72.5)	3.3771	0.1848			
		T (TT/AT)	25	9 (36.0)	16 (64.0)					
		G vs TT/AT	116	27 (23.3)	89 (76.7)	1.1461	0.2844			
		G/T vs GG	94	28 (29.8)	66 (70.2)	2.0562	0.1516			
ERCC1	rs11615	CC	20	3 (15.0)	17 (85.0)					
		CT	72	21 (29.2)	51 (70.8)	1.6951	0.4285			
		TT	49	12 (24.5)	37 (75.5)					
		C	92	24 (26.1)	68 (73.9)	0	0.9966			
		T	121	33 (27.3)	88 (72.7)	0.7908	0.3739			
	rs3212986	GG	74	17 (23.0)	57 (77.0)					
		GT	56	17 (30.4)	39 (69.6)	1.2532	0.5344			
		TT	11	2 (18.2)	9 (81.8)					
		G	130	34 (26.2)	96 (73.8)		0.7293*			
		T	67	19 (28.4)	48 (71.6)	0.2905	0.5899			
ERCC2	rs13181	GG	25	3 (12.0)	22 (88.0)					
		GT	55	14 (25.5)	41 (74.5)	3.4196	0.1809			
		TT	61	19 (31.1)	42 (68.9)					
		G	80	17 (21.2)	63 (78.8)	1.3006	0.2541			
		T	116	33 (28.4)	83 (71.6)	2.1254	0.1449			
	rs1799793	AA	24	2 (8.3)	22 (91.7)					
		AG	55	19 (34.5)	36 (65.5)	6.1423	0.0464			
		GG	62	15 (24.2)	47 (75.8)					
		A	79	21 (26.6)	58 (73.4)	0.0165	0.8979			
		G	117	34 (29.1)	83 (70.9)	3.4755	0.0623			
	rs50872	CC	82	25 (30.5)	57 (69.5)					
		CT	54	10 (18.5)	44 (81.5)		0.2830*			
		TT	5	1 (20.0)	4 (80.0)					
		C	136	35 (25.7)	101 (74.3)		1*			
		T	59	11 (18.6)	48 (81.4)	1.9469	0.1629			
	rs238416	AA	19	5 (26.3)	14 (73.7)					
		AG	67	19 (28.4)	48 (71.6)	0.6866	0.7094			
		GG	55	12 (21.8)	43 (78.2)					
		A	86	24 (27.9)	62 (72.1)	0.3731	0.5413			
		G	122	31 (25.4)	91 (74.6)		1*			
ERCC5	rs17655	CC	79	21 (26.6)	58 (73.4)					
		CG	52	13 (25.0)	39 (75.0)	0.2145	0.8983			
		GG	10	2 (20.0)	8 (80.0)					
		C	131	34 (26.0)	97 (74.0)		1*			
		G	62	15 (24.2)	47 (75.8)	0.0165	0.8979			
	rs1047768	CC	43	12 (27.9)	31 (72.1)					
		CT	70	19 (27.1)	51 (72.9)	1.0906	0.5797			
		TT	28	5 (17.9)	23 (82.1)					
		C	113	31 (27.4)	82 (72.6)	0.6373	0.4247			
		T	98	24 (24.5)	74 (75.5)	0.0478	0.8269			
IL1B	rs12621220	CC	73	22 (30.1)	51 (69.9)					
		CT	61	13 (21.3)	48 (78.7)	1.8513	0.3963			
		TT	7	1 (14.3)	6 (85.7)					
		C	134	35 (26.1)	99 (73.9)		0.6781*			
		T	68	14 (20.6)	54 (79.4)	1.2234	0.2687			
	rs1143623	CC	7	1 (14.3)	6 (85.7)					
		CG	61	13 (21.3)	48 (78.7)	1.8513	0.3963			
		GG	73	22 (30.1)	51 (69.9)					
		C	68	14 (20.6)	54 (79.4)	1.2234	0.2687			
		G	134	35 (26.1)	99 (73.9)		0.6781*			

Table 35. (Continued).

Gene	SNPs	Genotype	N	Toxicity		χ^2	p-value	Cat Ref	RR	95% CI
				>2 N (%)	0-2 N (%)					
IL1B	rs16944	AA	14	1 (7.1)	13 (92.9)					
		AG	71	17 (23.9)	54 (76.1)	3.8714	0.1443			
		GG	56	18 (32.1)	38 (67.9)					
		A	85	18 (21.2)	67 (78.8)	1.5975	0.2063			
		G	127	35 (27.6)	92 (72.4)		0.1167*			
	rs1143627	CC	15	2 (13.3)	13 (86.7)					
		CT	70	16 (22.9)	54 (77.1)	2.7246	0.2561			
		TT	56	18 (32.1)	38 (67.9)					
		C	85	18 (21.2)	67 (78.8)	1.5975	0.2063			
		T	126	34 (27.0)	92 (73.0)		0.3547*			
IL6	rs1800795	CC	17	8 (47.1)	9 (52.9)					
		CG	69	16 (23.2)	53 (76.8)	4.7417	0.0934			
		GG	55	12 (21.8)	43 (78.2)					
		C	86	24 (27.9)	62 (72.1)	0.3731	0.5413			
		G	124	28 (22.6)	96 (77.4)	3.512	0.0609			
IL16	rs7170924	GG	79	17 (21.5)	62 (78.5)					
		GT	53	15 (28.3)	38 (71.7)	2.5761	0.2758			
		TT	9	4 (44.4)	5 (55.6)					
		G	132	32 (24.2)	100 (75.8)		0.2332*			
		T	62	19 (30.6)	43 (69.4)	1.0795	0.2988			
MDM2	rs1470383	CC	7	1 (14.3)	6 (85.7)					
		CT	35	7 (20.0)	28 (80.0)	1.423	0.4909			
		TT	99	28 (28.3)	71 (71.7)					
		C	42	8 (19.0)	34 (81.0)	0.8817	0.3477			
		T	134	35 (26.1)	99 (73.9)		0.6781*			
	rs1690924	AA	56	11 (19.6)	45 (80.4)					
		AG	65	21 (32.3)	44 (67.7)	2.7761	0.2496			
		GG	19	4 (21.1)	15 (78.9)					
		A	121	32 (26.4)	89 (73.6)		0.7807*			
		G	84	25 (29.8)	59 (70.2)	1.3103	0.2523			
MTHFR	rs1801131	AA	75	15 (20.0)	60 (80.0)					
		AC	53	17 (32.1)	36 (67.9)	2.5883	0.2741			
		CC	13	4 (30.8)	9 (69.2)					
		A	128	32 (25.0)	96 (75.0)		0.7394*			
		C	66	21 (31.8)	45 (68.2)	1.9948	0.1578			
	rs1801133	CC	44	12 (27.3)	32 (72.7)					
		CT	73	19 (26.0)	54 (74.0)	0.3582	0.8360			
		TT	24	5 (20.8)	19 (79.2)					
		C	117	31 (26.5)	86 (73.5)	0.104	0.7470			
		T	97	24 (24.7)	73 (75.3)	0.0123	0.9117			
MTR	rs1805087	AA	99	26 (26.3)	73 (73.7)					
		AG	37	10 (27.0)	27 (73.0)		0.5781*			
		GG	5	0 (0.0)	5 (100.0)					
		A	136	36 (26.5)	100 (73.5)		0.3287*			
		G	42	10 (23.8)	32 (76.2)	0.0089	0.9248			
SLC19A1	rs1051266	AA	35	6 (17.1)	29 (82.9)					
		AG	64	18 (28.1)	46 (71.9)	1.7259	0.4219			
		GG	42	12 (28.6)	30 (71.4)					
		A	99	24 (24.2)	75 (75.8)	0.1076	0.7429			
		G	106	30 (28.3)	76 (71.7)	1.1863	0.2761			
XRCC1	rs1799782	CC	120	30 (25.0)	90 (75.0)					
		CT	20	6 (30.0)	14 (70.0)		0.6973*			
		TT	1	0 (0.0)	1 (100.0)					
		C	140	36 (25.7)	104 (74.3)		1*			
		T	21	6 (28.6)	15 (71.4)	0.0056	0.9402			
	rs25487	AA	20	1 (5.0)	19 (95.0)					
		AG	71	18 (25.4)	53 (74.6)	6.3214	0.0424			
		GG	50	17 (34.0)	33 (66.0)					
		A	91	19 (20.9)	72 (79.1)	2.2725	0.1317			
		G	121	35 (28.9)	86 (71.1)	3.9856	0.0459	AA	5.79	1.03-32.47

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 36. Influence of gene polymorphisms on number of adverse events.

	Number of adverse events	
	OR ($CI_{95\%}$)	**p-value**
		>1 adverse events
ERCC2 Asp312Asn		
AA	0.23 (0.07-0.71)	0.01140
(Reference) AG		
GG	0.31 (0.13-0.73)	0.00891
IL1B rs12621220-CT/TT	3.37 (1.55-7.69)	0.00280
		>2 adverse events
ERCC2 Asp312Asn-AG/GG	5.92 (1.52-40.18)	0.026040
ABCB1 C1236T-TT	4.35 (1.67-11.76)	0.002842

Table 37. Association of gene polymorphisms with hematological toxicity.

Gene	SNPs	Genotype	N	Hematological toxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
ABCB1	rs1045642	CC	49	15 (30.6)	34 (69.4)					
		CT	62	11 (17.7)	51 (82.3)	2.6133	0.2707			
		TT	30	8 (26.7)	22 (73.3)					
		C	111	26 (23.4)	85 (76.6)	0.0164	0.8982			
		T	92	19 (20.7)	73 (79.3)	1.2317	0.2671			
	rs1128503	CC	53	14 (26.4)	39 (73.6)					
		CT	63	10 (15.9)	53 (84.1)	5.9393	0.0513			
		TT	25	10 (40.0)	15 (60.0)					
		C	116	24 (20.7)	92 (79.3)	3.2023	0.0735			
		T	88	20 (22.7)	68 (77.3)	0.0856	0.7698			
	rs2032582	GG	47	14 (29.8)	33 (70.2)					
		G (GT/AG)	69	11 (15.9)	58 (84.1)	5.2749	0.0715			
		T (TT/AT)	25	9 (36.0)	16 (64.0)					
		G vs TT/AT	116	25 (21.6)	91 (78.4)	1.6232	0.2027			
		G/T vs GG	94	20 (21.3)	74 (78.7)	0.8188	0.3655			
ERCC1	rs11615	CC	20	1 (5.0)	19 (95.0)					
		CT	72	21 (29.2)	51 (70.8)	5.0014	0.0820			
		TT	49	12 (24.5)	37 (75.5)					
		C	92	22 (23.9)	70 (76.1)	0.0058	0.9392			
		T	121	33 (27.3)	88 (72.7)	3.5153	0.0608			
	rs3212986	GG	74	16 (21.6)	58 (78.4)					
		GT	56	16 (28.6)	40 (71.4)	1.0708	0.5854			
		TT	11	2 (18.2)	9 (81.8)					
		G	130	32 (24.6)	98 (75.4)		1*			
		T	67	18 (26.9)	49 (73.1)	0.2807	0.5962			
ERCC2	rs13181	GG	25	5 (20.0)	20 (80.0)					
		GT	55	12 (21.8)	43 (78.2)	0.8596	0.6506			
		TT	61	17 (27.9)	44 (72.1)					
		G	80	17 (21.2)	63 (78.8)	0.5064	0.4767			
		T	116	29 (25.0)	87 (75.0)	0.0742	0.7853			
	rs1799793	AA	24	6 (25.0)	18 (75.0)					
		AG	55	12 (21.8)	43 (78.2)	0.2658	0.8756			
		GG	62	16 (25.8)	46 (74.2)					
		A	79	18 (22.8)	61 (77.2)	0.0475	0.8274			
		G	117	28 (23.9)	89 (76.1)	0	1			
	rs50872	CC	82	26 (31.7)	56 (68.3)					
		CT	54	8 (14.8)	46 (85.2)		0.0381*			
		TT	5	0 (0.0)	5 (100.0)					
		C	136	34 (25.0)	102 (75.0)		0.3365*			
		T	59	8 (13.6)	51 (86.4)	5.2237	0.0223	T	2.34	1.13-4.85
	rs238416	AA	19	5 (26.3)	14 (73.7)					
		AG	67	15 (22.4)	52 (77.6)	0.2134	0.8988			
		GG	55	14 825.5)	41 (74.5)					
		A	86	20 (23.3)	66 (76.7)	0.0092	0.9236			
		G	122	29 (23.8)	93 (76.2)		0.7787*			
ERCC5	rs17655	CC	79	21 (26.6)	58 (73.4)					
		CG	52	12 (23.1)	40 (76.9)	1.3822	0.5010			
		GG	10	1 (10.0)	9 (90.0)					
		C	131	33 (25.2)	98 (74.8)		0.4511*			
		G	62	13 (21.0)	49 (79.0)	0.3309	0.5651			
	rs1047768	CC	43	9 (20.9)	34 (79.1)					
		CT	70	21 (30.0)	49 (70.0)	3.0415	0.2185			
		TT	28	4 (14.3)	24 (85.7)					
		C	113	30 (26.5)	83 (73.5)	1.2348	0.2665			
		T	98	25 (25.5)	73 (74.5)	0.1380	0.7103			
IL1B	rs12621220	CC	73	56 (76.7)	17 (23.3)					
		CT	61	45 (73.8)	16 (26.2)	0.5459	0.7611			
		TT	7	6 (85.7)	1 (14.3)					
		C	134	101 (75.4)	33 (24.6)		1*			
		T	68	51 (75.0)	17 (25.0)	0.0564	0.8123			
	rs1143623	CC	7	6 (85.7)	1 (14.3)					
		CG	61	45 (73.8)	16 (26.2)	0.5459	0.7611			
		GG	73	56 (76.7)	17 (23.3)					
		C	68	51 (75.0)	17 (25.0)	0.0564	0.8123			
		G	134	101 (75.4)	33 (24.6)		1*			

Table 37. (Continued).

Gene	SNPs	Genotype	N	Hematological toxicity		χ^2	p-value	Cat Ref	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
IL1B	rs16944	AA	14	12 (85.7)	2 (14.3)					
		AG	71	52 (73.2)	19 (26.8)	1.0356	0.5958			
		GG	56	43 (76.8)	13 (23.2)					
		A	85	64 (75.3)	21 (24.7)	0.0410	0.8395			
		G	127	95 (74.8)	32 (25.2)		0.5180*			
	rs1143627	CC	15	12 (80.0)	3 (20.0)	0.2615	0.8774			
		CT	70	52 (74.3)	18 (25.7)	0.0410	0.8395			
		TT	56	43 (76.8)	13 (23.2)					
		C	85	64 (75.3)	21 (24.7)	0.0144	0.9044			
		T	126	95 (75.4)	31 (24.6)		1*			
IL6	rs1800795	CC	17	3 (17.6)	14 (82.4)					
		CG	69	16 (23.2)	53 (76.8)	0.7207	0.6974			
		GG	55	15 (27.3)	40 (72.7)					
		C	86	19 (22.1)	67 (77.9)	0.2495	0.6174			
		G	124	31 (25.0)	93 (75.0)		0.7629*			
IL16	rs7170924	GG	79	13 (16.5)	66 (83.5)					
		GT	53	19 (35.8)	34 (64.2)	6.5382	0.0380			
		TT	9	2 (22.2)	7 (77.8)					
		G	132	32 (24.2)	100 (75.8)		1*			
		T	62	21 (33.9)	41 (66.1)	4.8451	0.0277	GG	2.06	1.08-3.92
MDM2	rs1470383	CC	7	0 (0.0)	7 (100.0)					
		CT	35	8 (22.9)	27 (77.1)	2.5044	0.2859			
		TT	99	26 (26.3)	73 (73.7)					
		C	42	8 (19.0)	34 (81.0)	0.4910	0.4835			
		T	134	34 (25.4)	100 (74.6)		0.1958*			
	rs1690924	AA	56	14 (25.0)	42 (75.0)					
		AG	65	16 (24.6)	49 (75.4)	0.1274	0.9383			
		GG	19	4 (21.1)	15 (78.9)					
		A	121	30 (24.8)	91 (75.2)		1*			
		G	84	20 (23.8)	64 (76.2)	0	1*			
MTHFR	rs1801131	AA	75	16 (21.3)	59 (78.7)					
		AC	53	15 (28.3)	38 (71.7)	0.8325	0.6595			
		CC	13	3 (23.1)	10 (76.9)					
		A	128	31 (24.2)	97 (75.8)		1*			
		C	66	18 (27.3)	48 (72.7)	0.6768	0.4107			
	rs1801133	CC	44	12 (27.3)	32 (72.7)					
		CT	73	18 (24.7)	55 (75.3)	0.9791	0.6129			
		TT	24	4 (16.7)	20 (83.3)					
		C	117	30 (25.6)	87 (74.4)	0.4547	0.5001			
		T	97	22 (22.7)	75 (77.3)	0.1430	0.7053			
MTR	rs1805087	AA	99	79 (79.8)	20 (20.2)					
		AG	37	25 (67.6)	12 (32.4)		0.2109*			
		GG	5	3 (60.0)	2 (40.0)					
		A	136	104 (76.5)	32 (23.5)		0.5941*			
		G	42	28 (66.7)	14 (33.3)	2.1075	0.1466			
SLC19A1	rs1051266	AA	35	7 (20.0)	28 (80.0)					
		AG	64	16 (25.0)	48 (75.0)	0.4501	0.7985			
		GG	42	11 (26.2)	31 (73.8)					
		A	99	23 (23.2)	76 (76.8)	0.0257	0.8727			
		G	106	27 (25.5)	79 (74.5)	0.1834	0.6685			
XRCC1	rs1799782	CC	120	31 (25.8)	89 (74.2)					
		CT	20	3 (15.0)	17 (85.0)		0.5472*			
		TT	1	0 (0.0)	1 (100.0)					
		C	140	34 (24.3)	106 (75.7)		1*			
		T	21	3 (14.3)	18 (85.7)	0.7478	0.3872			
	rs25487	AA	20	4 (20.0)	16 (80.0)					
		AG	71	18 (25.4)	53 (74.6)	0.2448	0.8848			
		GG	50	12 (24.0)	38 (76.0)					
		A	91	22 (24.2)	69 (75.8)	0	1			
		G	121	30 (24.8)	91 (75.2)		0.7823*			

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 38. Influence of clinical characteristic and gene polymorphisms on hematological toxicity.

	Hematological Toxicity	
	OR ($CI_{95\%}$)	**p-value**
Chemotherapy agents		
Gemcitabine (Reference)		
Paclitaxel	0.15 (0.03, 0.54)	0.00559
Pemetrexed	0.11 (0.02, 0.42)	0.00218
Vinorelbine	0.36 (0.11, 1.10)	0.07525
ERCC2 rs50872-CC	3.23 (1.29, 8.82)	0.01562
IL16 rs7170924- GT/TT	3.19 (1.36, 7.97)	0.01007

Table 39. Association of gene polymorphisms with asthenia.

Gene	SNPs	Genotype	N	Asthenia		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
ABCB1	rs1045642	CC	49	1 (2.0)	48 (98.0)					
		CT	62	2 (3.2)	60 (96.8)		1*			
		TT	30	0 (0.0)	30 (100.0)					
		C	111	3 (2.7)	108 (97.3)		1*			
		T	92	2 (2.2)	90 (97.8)		1*			
	rs1128503	CC	53	1 (1.9)	52 (98.1)					
		CT	63	1 (1.6)	62 (98.4)		0.5838*			
		TT	25	1 (4.0)	24 (96.0)					
		C	116	2 (1.7)	114 (98.3)		0.4458*			
		T	88	2 (2.3)	86 (97.7)		1*			
	rs2032582	GG	69	2 (2.9)	67 (97.1)					
		G (GT/AG)	47	1 (2.1)	46 (97.9)		1*			
		T (TT/AT)	25	0 (0.0)	25 (100.0)					
		G vs TT/AT	116	3 (2.6)	113 (97.4)		1*			
		G/T vs GG	94	2 (2.1)	92 (97.9)		1*			
ERCC1	rs11615	CC	20	0 (0.0)	20 (100.0)					
		CT	72	2 (2.8)	70 (97.2)		1*			
		TT	49	1 (2.0)	48 (98.0)					
		C	92	2 (2.2)	90 (97.8)		1*			
		T	121	3 (2.5)	118 (97.5)		1*			
	rs3212986	GG	74	1 (1.4)	73 (98.6)					
		GT	56	2 (3.6)	54 (96.4)		0.6692*			
		TT	11	0 (0.0)	11 (100.0)					
		G	130	3 (2.3)	127 (97.7)		1*			
		T	67	2 (3.0)	65 (97.0)		0.6043*			
ERCC2	rs13181	GG	25	0 (0.0)	25 (100.0)					
		GT	55	2 (3.6)	53 (96.4)		0.7799*			
		TT	61	1 (1.6)	60 (98.4)					
		G	80	2 (2.5)	78 (97.5)		1*			
		T	116	3 (2.6)	113 (97.4)		1*			
	rs1799793	AA	24	0 (0.0)	24 (100.0)					
		AG	55	2 (3.6)	53 (96.4)		0.7726*			
		GG	62	1 (1.6)	61 (98.4)					
		A	79	2 (2.5)	77 (97.5)		1*			
		G	117	3 (2.6)	114 (97.4)		1*			
	rs50872	CC	82	3 (3.7)	79 (96.3)					
		CT	54	0 (0.0)	54 (100.0)		0.3513*			
		TT	5	0 (0.0)	5 (100.0)					
		C	136	3 (2.2)	133 (97.8)		1*			
		T	59	0 (0.0)	59 (100.0)		0.2647*			
	rs238416	AA	19	1 (5.3)	18 (94.7)					
		AG	67	2 (3.0)	65 (97.0)		0.2587*			
		GG	55	0 (0.0)	55 (100.0)					
		A	86	3 (3.5)	83 (96.5)		0.2812*			
		G	122	2 (1.6)	120 (98.4)		0.3544*			
ERCC5	rs17655	CC	79	1 (1.3)	78 (98.7)					
		CG	52	2 (3.8)	50 (96.2)		0.6497*			
		GG	10	0 (0.0)	10 (100.0)					
		C	131	3 (2.3)	128 (97.7)		1*			
		G	62	2 (3.2)	60 (96.8)		0.5823*			
	rs1047768	CC	43	0 (0.0)	43 (100.0)					
		CT	70	3 (4.3)	67 (95.7)		0.3025*			
		TT	28	0 (0.0)	28 (100.0)					
		C	113	3 (2.7)	110 (97.3)		1*			
		T	98	3 (3.1)	95 (96.9)		0.5531*			
IL1B	rs12621220	CC	73	71 (97.3)	2 (2.7)					
		CT	61	60 (98.4)	1 (1.6)		1*			
		TT	7	7 (100.0)	0 (0.0)					
		C	134	131 (97.8)	3 (2.2)		1*			
		T	68	67 (98.5)	1 (1.5)		1*			
	rs1143623	CC	7	7 (100.0)	0 (0.0)					
		CG	61	60 (98.4)	1 (1.6)		1*			
		GG	73	71 (97.3)	2 (2.7)					
		C	68	67 (98.5)	1 (1.5)		1*			
		G	134	131 (100.0)	3 (0.0)		1*			

Table 39. (Continued).

Gene	SNPs	Genotype	N	Asthenia		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
IL1B	rs16944	AA	14	14 (100.0)	0 (0.0)					
		AG	71	69 (97.2)	2 (2.8)		1*			
		GG	56	55 (98.2)	1 (1.8)					
		A	85	83 (97.6)	2 (2.4)		1*			
		G	127	124 (97.6)	3 (2.4)		1*			
	rs1143627	CC	15	15 (100.0)	0 (0.0)					
		CT	70	68 (97.1)	2 (2.9)		1*			
		TT	56	55 (98.2)	1 (1.8)					
		C	85	83 (97.6)	2 (2.4)		1*			
		T	126	123 (97.6)	3 (2.4)		1*			
IL6	rs1800795	CC	17	0 (0.0)	17 (100.0)					
		CG	69	1 (1.4)	68 (98.6)		0.7179*			
		GG	55	2 (3.6)	53 (96.4)					
		C	86	1 (1.2)	85 (98.8)		0.5604*			
		G	124	3 (2.4)	121 (97.6)		1*			
IL16	rs7170924	GG	79	0 (0.0)	79 (100.0)					
		GT	53	2 (3.8)	51 (96.2)		0.0377*			
		TT	9	1 (11.1)	8 (88.9)					
		G	132	2 (1.5)	130 (98.5)		0.1807*			
		T	62	3 (4.8)	59 (95.2)		0.0827*			
MDM2	rs1470383	CC	7	1 (14.3)	6 (85.7)					
		CT	35	0 (0.0)	35 (100.0)		0.1569*			
		TT	99	2 (2.0)	97 (98.0)					
		C	42	1 (2.4)	41 (97.6)		1*			
		T	134	2 (1.5)	132 (98.5)		0.1426*			
	rs1690924	AA	56	2 (3.6)	54 (96.4)					
		AG	65	1 (1.5)	64 (98.5)		0.7398*			
		GG	19	0 (0.0)	19 (100.0)					
		A	121	3 (2.5)	118 (97.5)		1*			
		G	84	1 (1.2)	83 (98.8)		0.5638*			
MTHFR	rs1801131	AA	75	1 (1.3)	74 (98.7)					
		AC	53	1 (1.9)	52 (98.1)		0.3047*			
		CC	13	1 (7.7)	12 (92.3)					
		A	128	2 (1.6)	126 (98.4)		0.2535*			
		C	66	2 (3.0)	64 (97.0)		0.5995*			
	rs1801133	CC	44	1 (2.3)	43 (97.7)					
		CT	72	2 (2.7)	71 (97.3)		1*			
		TT	24	0 (0.0)	24 (100.0)					
		C	117	3 (2.6)	114 (97.4)		1*			
		T	97	2 (2.1)	95 (97.9)		1*			
MTR	rs1805087	AA	99	96 (97.0)	3 (3.0)					
		AG	37	37 (100.0)	0 (0.0)		0.6075*			
		GG	5	5 (100.0)	0 (0.0)					
		A	136	133 (97.8)	3 (2.2)		1*			
		G	42	42 (100.0)	0 (0.0)		0.5545*			
SLC19A1	rs1051266	AA	35	0 (0.0)	35 (100.0)					
		AG	64	1 (1.6)	63 (98.4)		0.4548*			
		GG	42	2 (4.8)	40 (95.2)					
		A	99	1 (1.0)	98 (99.0)		0.2115*			
		G	106	3 (2.8)	103 (97.2)		0.5741*			
XRCC1	rs1799782	CC	120	3 (2.5)	117 (97.5)					
		CT	20	0 (0.0)	20 (100.0)		1*			
		TT	1	0 (0.0)	1 (100.0)					
		C	140	3 (2.1)	137 (97.9)		1*			
		T	21	0 (0.0)	21 (100.0)		1*			
	rs25487	AA	20	0 (0.0)	20 (100.0)					
		AG	71	3 (4.2)	68 (95.8)		0.3829*			
		GG	50	0 (0.0)	50 (100.0)					
		A	91	3 (3.3)	88 (96.7)		0.5523*			
		G	121	3 (2.5)	118 (97.5)		1*			

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 40. Association of gene polymorphisms with gastrointestinal toxicity.

Gene	SNPs	Genotype	N	Gastrointestinal toxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
ABCB1	rs1045642	CC	49	2 (4.1)	47 (95.9)					
		CT	62	1 (1.6)	61 (98.4)		0.5981*			
		TT	30	0 (0.0)	30 (100.0)					
		C	111	3 (2.7)	108 (97.3)		1*			
		T	92	1 (1.1)	91 (98.9)		0.2769*			
	rs1128503	CC	53	2 (3.8)	51 (96.2)					
		CT	63	1 (1.6)	62 (98.4)		0.7737*			
		TT	25	0 (0.0)	25 (100.0)					
		C	116	3 (2.6)	113 (97.4)		1*			
		T	88	1 (1.1)	87 (98.9)		0.5564*			
	rs2032582	GG	69	1 (1.4)	68 (98.6)					
		G (GT/AG)	47	2 (4.3)	45 (95.7)		0.5816*			
		T (TT/AT)	25	0 (0.0)	25 (100.0)					
		G vs TT/AT	116	3 (2.6)	113 (97.4)		1*			
		G/T vs GG	94	1 (1.1)	93 (98.9)		0.2577*			
ERCC1	rs11615	CC	70	0 (0.0)	20 (100.0)					
		CT	72	2 (2.8)	70 (97.2)		1*			
		TT	49	1 (2.0)	48 (98.0)					
		C	92	2 (2.2)	90 (97.8)		1*			
		T	121	3 (2.5)	118 (97.5)		1*			
	rs3212986	GG	74	2 (2.7)	72 (97.3)					
		GT	56	1 (1.8)	55 (98.2)		1*			
		TT	11	0 (0.0)	11 (100.0)					
		G	130	3 (2.3)	127 (97.7)		1*			
		T	67	1 (1.5)	66 (98.5)		1*			
ERCC2	rs13181	GG	25	0 (0.0)	25 (100.0)					
		GT	55	1 (1.8)	54 (98.2)		1*			
		TT	61	2 (3.3)	59 (96.7)					
		G	80	1 (1.2)	79 (98.8)		0.5785*			
		T	116	3 (2.6)	113 (97.4)		1*			
	rs1799793	AA	24	0 (0.0)	24 (100.0)					
		AG	55	2 (3.6)	53 (96.4)		0.7726*			
		GG	62	1 (1.6)	61 (98.4)					
		A	79	2 (2.5)	77 (97.5)		1*			
		G	117	3 (2.6)	114 (97.4)		1*			
	rs50872	CC	82	2 (2.4)	80 (97.6)					
		CT	54	1 (1.9)	53 (98.1)		1*			
		TT	5	0 (0.0)	5 (100.0)					
		C	136	3 (2.2)	133 (97.8)		1*			
		T	59	1 (1.7)	58 (98.3)		1*			
	rs238416	AA	19	0 (0.0)	19 (100.0)					
		AG	67	3 (4.5)	64 (95.5)		0.3634*			
		GG	55	0 (0.0)	55 (100.0)					
		A	86	3 (3.5)	83 (96.5)		0.2812*			
		G	122	3 (2.5)	119 (97.5)		1*			
ERCC5	rs17655	CC	79	2 (2.5)	77 (97.5)					
		CG	52	1 (1.9)	51 (98.1)		1*			
		GG	10	0 (0.0)	10 (100.0)					
		C	131	3 (2.3)	128 (97.7)		1*			
		G	62	1 (1.6)	61 (98.4)		1*			
	rs1047768	CC	43	3 (7.0)	40 (93.0)					
		CT	70	0 (0.0)	70 (100.0)		0.0342*			
		TT	28	0 (0.0)	28 (100.0)					
		C	113	3 (2.7)	110 (97.3)		1*			
		T	98	0 (0.0)	98 (100.0)		0.0269*	T	-	-
IL1B	rs12621220	CC	73	72 (98.6)	1 (1.4)					
		CT	61	59 (96.7)	2 (3.3)		0.6495*			
		TT	7	7 (100.0)	0 (0.0)					
		C	134	131 (97.8)	3 (2.2)		1*			
		T	68	66 (97.1)	2 (2.9)		0.6092*			
	rs1143623	CC	7	7 (100.0)	0 (0.0)					
		CG	61	59 (96.7)	2 (3.3)		0.6495*			
		GG	73	72 (98.6)	1 (1.4)					
		C	68	66 (97.1)	2 (2.9)		0.6092*			
		G	134	131 (97.8)	3 (2.2)		1*			

Table 40. (Continued).

Gene	SNPs	Genotype	N	Gastrointestinal toxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
IL1B	rs16944	AA	14	14 (100.0)	0 (0.0)					
		AG	71	69 (97.2)	2 (2.8)		1*			
		GG	56	55 (98.2)	1 (1.8)					
		A	85	83 (97.6)	2 (2.4)		1*			
		G	127	124 (97.6)	3 (2.4)		1*			
	rs1143627	CC	15	15 (100.0)	0 (0.0)					
		CT	70	68 (97.1)	2 (2.9)		1*			
		TT	56	55 (98.2)	1 (1.8)					
		C	85	83 (97.6)	2 (2.4)		1*			
		T	126	123 (97.6)	3 (2.4)		1*			
IL6	rs1800795	CC	17	1 (5.9)	16 (94.1)					
		CG	69	1 (1.4)	68 (98.6)		0.4938*			
		GG	55	1 (1.8)	54 (98.2)					
		C	86	2 (2.3)	84 (97.7)		1*			
		G	124	2 (1.6)	122 (98.4)		0.3219*			
IL16	rs7170924	GG	79	2 (2.5)	77 (97.5)					
		GT	53	0 (0.0)	53 (100.0)		0.1496*			
		TT	9	1 (11.1)	8 (88.9)					
		G	132	2 (1.5)	130 (98.5)		0.1807*			
		T	62	1 (1.6)	61 (98.4)		1*			
MDM2	rs1470383	CC	7	0 (0.0)	7 (100.0)					
		CT	35	0 (0.0)	35 (100.0)		0.6287*			
		TT	99	3 (3.0)	96 (97.0)					
		C	42	0 (0.0)	42 (100.0)		0.5545*			
		T	134	3 (2.2)	131 (97.8)		1*			
	rs1690924	AA	56	2 (3.6)	54 (96.4)					
		AG	65	1 (1.5)	64 (98.5)		0.7398*			
		GG	19	0 (0.0)	19 (100.0)					
		A	121	3 (2.5)	118 (97.5)		1*			
		G	84	1 (1.2)	83 (98.8)		0.5638*			
MTHFR	rs1801131	AA	75	1 (1.3)	74 (98.7)					
		AC	53	1 (1.9)	52 (98.1)		0.3047*			
		CC	13	1 (7.7)	12 (92.3)					
		A	128	2 (1.6)	126 (98.4)		0.2535*			
		C	66	2 (3.0)	64 (97.0)		0.5995*			
	rs1801133	CC	44	2 (4.5)	42 (95.5)					
		CT	73	1 (1.4)	72 (98.6)		0.5786*			
		TT	24	0 (0.0)	24 (100.0)					
		C	117	3 (2.6)	114 (97.4)		1*			
		T	97	1 (1.0)	96 (99.0)		0.2296*			
MTR	rs1805087	AA	99	97 (98.0)	2 (2.0)					
		AG	37	37 (100.0)	0 (0.0)		0.1204*			
		GG	5	4 (80.0)	1 (20.0)					
		A	136	134 (98.5)	2 (1.5)		0.1034*			
		G	42	41 (97.6)	1 (2.4)		1*			
SLC19A1	rs1051266	AA	35	3 (8.6)	32 (91.4)					
		AG	64	0 (0.0)	64 (100.0)		0.0143*			
		GG	42	0 (0.0)	42 (100.0)					
		A	99	3 (3.0)	96 (97.0)		0.5545*			
		G	106	0 (0.0)	106 (100.0)		0.0143*	G	-	-
XRCC1	rs1799782	CC	120	3 (2.5)	117 (97.5)					
		CT	20	0 (0.0)	20 (100.0)		1*			
		TT	1	0 (0.0)	1 (100.0)					
		C	140	3 (2.1)	137 (97.9)		1*			
		T	21	0 (0.0)	21 (100.0)		1*			
	rs25487	AA	20	1 (5.0)	19 (95.0)					
		AG	71	1 (1.4)	70 (98.6)		0.5381*			
		GG	50	1 (2.0)	49 (98.0)					
		A	91	2 (2.2)	89 (97.8)		1*			
		G	121	2 (1.7)	119 (98.3)		0.3703*			

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 41. Association of gene polymorphisms with infection.

Gene	SNPs	Genotype	N	Infection		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
ABCB1	rs1045642	CC	49	2 (4.1)	47 (95.9)					
		CT	62	2 (3.2)	60 (96.8)		1*			
		TT	30	1 (3.3)	29 (96.7)					
		C	111	4 (3.6)	107 (96.4)		1*			
		T	92	3 (3.3)	89 (96.7)		1*			
	rs1128503	CC	53	2 (3.8)	51 (96.2)					
		CT	63	1 (1.6)	62 (98.4)		0.26*			
		TT	25	2 (8.0)	23 (92.0)					
		C	116	3 (2.6)	113 (97.4)		0.2149*			
		T	88	3 (3.4)	85 (96.6)		1*			
	rs2032582	GG	47	1 (2.1)	46 (97.9)					
		G (GT/AG)	69	2 (2.9)	67 (97.1)		0.3979*			
		T (TT/AT)	25	2 (8.0)	23 (92.0)					
		G vs TT/AT	116	3 (2.6)	113 (97.4)		0.2149*			
		G/T vs GG	94	4 (4.3)	90 (95.7)		0.6648*			
ERCC1	rs11615	CC	20	0 (0.0)	20 (100.0)					
		CT	72	3 (4.2)	69 (95.8)		1*			
		TT	49	2 (4.1)	47 (95.9)					
		C	92	3 (3.3)	89 (96.7)		1*			
		T	121	5 (4.1)	116 (95.9)		1*			
	rs3212986	GG	74	2 (2.7)	72 (97.3)					
		GT	56	3 (5.4)	53 (94.6)		0.7691*			
		TT	11	0 (0.0)	11 (100.0)					
		G	130	5 (3.8)	125 (96.2)		1*			
		T	67	3 (4.5)	64 (95.5)		0.6685*			
ERCC2	rs13181	GG	25	0 (0.0)	25 (100.0)					
		GT	55	1 (1.8)	54 (98.2)		0.3313*			
		TT	61	4 (6.6)	57 (93.4)					
		G	80	1 (1.2)	79 (98.8)		0.1659*			
		T	116	5 (4.3)	111 (95.7)		0.5859*			
	rs1799793	AA	24	0 (0.0)	24 (100.0)					
		AG	55	3 (5.5)	52 (94.5)		0.5987*			
		GG	62	2 (3.2)	60 (96.8)					
		A	79	3 (3.8)	76 (96.2)		1*			
		G	117	5 (4.3)	112 (95.7)		0.5884*			
	rs50872	CC	82	3 (3.7)	79 (96.3)					
		CT	54	2 (3.7)	52 (96.3)		1*			
		TT	5	0 (0.0)	5 (100.0)					
		C	136	5 (3.7)	131 (96.3)		1*			
		T	59	2 (3.4)	57 (96.6)		1*			
	rs238416	AA	19	3 (15.8)	16 (84.2)					
		AG	67	2 (3.0)	65 (97.0)		0.0094*			
		GG	55	0 (0.0)	55 (100.0)					
		A	86	5 (5.8)	81 (94.2)		0.1565*			
		G	122	2 (1.6)	120 (98.4)		0.0177*	G	9.63	1.56-59.6
ERCC5	rs17655	CC	79	2 (2.5)	77 (97.5)					
		CG	52	3 (5.8)	49 (94.2)		0.5767*			
		GG	10	0 (0.0)	10 (100.0)					
		C	131	5 (3.8)	126 (96.2)		1*			
		G	62	3 (4.8)	59 (95.2)		0.6541*			
	rs1047768	CC	43	2 (4.7)	41 (95.3)					
		CT	70	2 (2.9)	68 (97.1)		0.8475*			
		TT	28	1 (3.6)	27 (96.4)					
		C	113	4 (3.5)	109 (96.5)		1*			
		T	98	3 (3.1)	95 (96.9)		0.6407*			
IL1B	rs12621220	CC	73	71 (97.3)	2 (2.7)					
		CT	61	58 (95.1)	3 (4.9)		0.7367*			
		TT	8	7 (100.0)	0 (0.0)					
		C	134	129 (96.3)	5 (3.7)		1*			
		T	68	65 (95.6)	3 (4.4)		0.6723*			
	rs1143623	CC	7	7 (100.0)	0 (0.0)					
		CG	61	58 (95.1)	3 (4.9)		0.7367*			
		GG	73	71 (97.3)	2 (2.7)					
		C	68	65 (95.6)	3 (4.4)		0.6723*			
		G	134	129 (96.3)	5 (3.7)		1*			

Table 41. (Continued).

Gene	SNPs	Genotype	N	Infection		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
IL1B	rs16944	AA	14	13 (92.9)	1 (7.1)					
		AG	71	69 (97.2)	2 (2.8)		0.5112*			
		GG	56	54 (96.4)	2 (3.6)					
		A	85	82 (96.5)	3 (3.5)		1*			
		G	127	123 (96.9)	4 (3.1)		0.4119*			
	rs1143627	CC	15	14 (93.3)	1 (6.7)					
		CT	70	68 (97.1)	2 (2.9)		0.6501*			
		TT	56	54 (96.4)	2 (3.6)					
		C	85	82 (96.5)	3 (3.5)		1*			
		T	126	122 (96.8)	4 (3.2)		0.4351*			
IL6	rs1800795	CC	17	0 (0.0)	17 (100.0)					
		CG	69	4 (5.8)	65 (94.2)		0.4273*			
		GG	55	1 (1.8)	54 (98.2)					
		C	86	4 (4.7)	82 (95.3)		0.6484*			
		G	124	5 (4.0)	119 (96.0)		1*			
IL16	rs7170924	GG	79	5 (6.3)	74 (93.7)					
		GT	53	0 (0.0)	53 (100.0)		0.1676*			
		TT	9	0 (0.0)	9 (100.0)					
		G	132	5 (3.8)	127 (96.2)		1*			
		T	62	0 (0.0)	62 (100.0)		0.0671*			
MDM2	rs1470383	CC	7	0 (0.0)	7 (100.0)					
		CT	35	1 (2.9)	34 (97.1)		1*			
		TT	99	4 (4.0)	95 (96.0)					
		C	42	1 (2.4)	41 (97.6)		1*			
		T	134	5 (3.7)	129 (96.3)		1*			
	rs1690924	AA	56	1 (1.8)	55 (98.2)					
		AG	65	4 (6.2)	61 (93.8)		0.443*			
		GG	19	0 (0.0)	19 (100.0)					
		A	121	5 (4.1)	116 (95.9)		1*			
		G	84	3 (4.8)	81 (95.2)		0.6481*			
MTHFR	rs1801131	AA	75	2 (2.7)	73 (97.3)					
		AC	53	3 (5.7)	50 (94.3)		0.7848*			
		CC	13	0 (0.0)	13 (100.0)					
		A	128	5 (3.9)	123 (96.1)		1*			
		C	66	3 (4.5)	63 (95.5)		0.6649*			
	rs1801133	CC	44	1 (2.3)	43 (97.7)					
		CT	73	2 (2.7)	71 (97.3)		0.3826			
		TT	24	2 (8.3)	22 (91.7)					
		C	117	3 (2.6)	114 (97.4)		0.2008*			
		T	97	4 (4.1)	93 (95.9)		1*			
MTR	rs1805087	AA	99	94 (94.9)	5 (5.1)					
		AG	37	37 (100.0)	0 (0.0)		0.4362*			
		GG	5	5 (100.0)	0 (0.0)					
		A	136	131 (96.3)	5 (3.7)		1*			
		G	42	42 (100.0)	0 (0.0)		0.3219*			
SLC19A1	rs1051266	AA	35	0 (0.0)	35 (100.0)					
		AG	64	2 (3.1)	62 (96.9)		0.2639*			
		GG	42	3 (7.1)	39 (92.9)					
		A	99	2 (2.0)	97 (98.0)		0.1564*			
		G	106	5 (4.7)	101 (95.3)		0.3323*			
XRCC1	rs1799782	CC	120	5 (4.2)	115 (95.8)					
		CT	20	0 (0.0)	20 (100.0)		1*			
		TT	1	0 (0.0)	1 (100.0)					
		C	140	5 (3.6)	135 (96.4)		1*			
		T	21	0 (0.0)	21 (100.0)		1*			
	rs25487	AA	20	1 (5.0)	19 (95.0)					
		AG	71	3 (4.2)	68 (95.8)		0.6972*			
		GG	50	1 (2.0)	49 (98.0)					
		A	91	4 (4.4)	87 (95.6)		0.6557*			
		G	121	4 (3.3)	117 (96.7)		0.5401*			

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 42. Association of gene polymorphisms with nephrotoxicity.

Gene	SNPs	Genotype	N	Nephrotoxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
ABCB1	rs1045642	CC	49	1 (2.0)	48 (98.0)					
		CT	62	0 (0.0)	62 (100.0)		0.0958*			
		TT	30	2 (6.7)	28 (93.3)					
		C	111	1 (0.9)	110 (99.1)		0.1145*			
		T	92	2 (2.2)	90 (97.8)		1*			
	rs1128503	CC	53	1 (1.9)	52 (98.1)					
		CT	63	1 (1.6)	62 (98.4)		0.5838*			
		TT	25	1 (4.0)	24 (96.0)					
		C	116	2 (1.7)	114 (98.3)		0.4458*			
		T	88	2 (2.3)	86 (97.7)		1*			
	rs2032582	GG	47	1 (2.1)	46 (97.9)					
		G (GT/AG)	69	1 (1.4)	68 (98.6)		0.7589*			
		T (TT/AT)	25	1 (4.0)	24 (96.0)					
		G vs TT/AT	116	2 (1.7)	114 (98.3)		0.4458*			
		G/T vs GG	94	2 (2.1)	92 (97.9)		1*			
ERCC1	rs11615	CC	20	0 (0.0)	20 (100.0)					
		CT	72	1 (1.4)	71 (98.6)		0.7261*			
		TT	49	2 (4.1)	47 (95.9)					
		C	92	1 (1.1)	91 (98.9)		0.2769*			
		T	121	3 (2.5)	118 (97.5)		1*			
	rs3212986	GG	74	2 (2.7)	72 (97.3)					
		GT	56	1 (1.8)	55 (98.2)		1*			
		TT	11	0 (0.0)	11 (100.0)					
		G	130	3 (2.3)	127 (97.7)		1*			
		T	67	1 (1.5)	66 (98.5)		1*			
ERCC2	rs13181	GG	25	0 (0.0)	25 (100.0)					
		GT	55	2 (3.6)	53 (96.4)		0.7799*			
		TT	61	1 (1.6)	60 (98.4)					
		G	80	2 (2.5)	78 (97.5)		1*			
		T	116	3 (2.6)	113 (97.4)		1*			
	rs1799793	AA	24	0 (0.0)	24 (100.0)					
		AG	55	2 (3.6)	53 (96.4)		0.7726*			
		GG	62	1 (1.6)	61 (98.4)					
		A	79	2 (2.5)	77 (97.5)		1*			
		G	117	3 (2.6)	114 (97.4)		1*			
	rs50872	CC	82	3 (3.7)	79 (96.3)					
		CT	54	0 (0.0)	54 (100.0)		0.3513*			
		TT	5	0 (0.0)	5 (100.0)					
		C	136	3 (2.2)	133 (97.8)		1*			
		T	59	0 (0.0)	59 (100.0)		0.2647*			
	rs238416	AA	19	0 (0.0)	19 (100.0)					
		AG	67	2 (3.0)	65 (97.0)		1*			
		GG	55	1 (1.8)	54 (98.2)					
		A	86	2 (2.3)	84 (97.7)		1*			
		G	122	3 (2.5)	119 (97.5)		1*			
ERCC5	rs17655	CC	79	3 (3.8)	76 (96.2)					
		CG	52	0 (0.0)	52 (100.0)		0.4206*			
		GG	10	0 (0.0)	10 (100.0)					
		C	131	3 (2.3)	128 (97.7)		1*			
		G	62	0 (0.0)	62 (100.0)		0.2556*			
	rs1047768	CC	43	1 (2.3)	42 (97.7)					
		CT	70	1 (1.4)	69 (98.6)		0.7729*			
		TT	28	1 (3.6)	27 (96.4)					
		C	113	2 (1.8)	111 (98.2)		0.488*			
		T	98	2 (2.0)	96 (98)		1*			
IL1B	rs12621220	CC	73	72 (98.6)	1 (1.4)					
		CT	61	60 (98.4)	1 (1.6)		0.1426*			
		TT	7	6 (85.7)	1 (14.3)					
		C	134	132 (98.5)	2 (1.5)		0.1426*			
		T	68	66 (97.1)	2 (2.9)		0.6092*			
	rs1143623	CC	7	6 (85.7)	1 (14.3)					
		CG	61	60 (98.4)	1 (1.6)		0.1426*			
		GG	73	72 (98.6)	1 (1.4)					
		C	68	66 (97.1)	2 (2.9)		0.6092*			
		G	134	132 (98.5)	2 (1.5)		0.1426*			

Table 42. (Continued).

Gene	SNPs	Genotype	N	Nephrotoxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
IL1B	rs16944	AA	14	14 (100.0)	0 (0.0)					
		AG	71	68 (95.8)	3 (4.2)		0.4566*			
		GG	56	56 (100.0)	0 (0.0)					
		A	85	82 (96.5)	3 (3.5)		0.2766*			
		G	127	124 (97.6)	3 (2.4)		1*			
	rs1143627	CC	15	14 (93.3)	1 (6.7)					
		CT	70	68 (97.1)	2 (2.9)		0.2203*			
		TT	56	56 (100.0)	0 (0.0)					
		C	85	82 (96.5)	3 (3.5)		0.2766*			
		T	126	124 (98.4)	2 (1.6)		0.2882*			
IL6	rs1800795	CC	17	2 (11.8)	15 (88.2)					
		CG	69	1 (1.4)	68 (98.6)		0.0384*			
		GG	55	0 (0.0)	55 (100.0)					
		C	86	3 (3.5)	83 (96.5)		0.2812*			
		G	124	1 (0.8)	123 (99.2)		0.0384*	G	14.59	1.11-191.6
IL16	rs7170924	GG	79	2 (2.5)	77 (97.5)					
		GT	53	1 (1.9)	52 (98.1)		1*			
		TT	9	0 (0.0)	9 (100.0)					
		G	132	3 (2.3)	129 (97.7)		1*			
		T	62	1 (1.6)	61 (98.4)		1*			
MDM2	rs1470383	CC	7	0 (0.0)	7 (100.0)					
		CT	35	2 (5.7)	33 (94.3)		0.2857*			
		TT	99	1 (1.0)	98 (99.0)					
		C	42	2 (4.8)	40 (95.2)		0.2115*			
		T	134	3 (2.2)	131 (97.8)		1*			
	rs1690924	AA	56	1 (1.8)	55 (98.2)					
		AG	65	1 (1.5)	64 (98.5)		0.5161*			
		GG	19	1 (5.3)	18 (94.7)					
		A	121	2 (1.7)	119 (98.3)		0.3566*			
		G	84	2 (2.4)	82 (97.6)		1*			
MTHFR	rs1801131	AA	75	2 (2.7)	73 (97.3)					
		AC	53	1 (1.9)	52 (98.1)		1*			
		CC	13	0 (0.0)	13 (100.0)					
		A	128	3 (2.3)	125 (97.7)		1*			
		C	66	1 (1.5)	65 (98.5)		1*			
	rs1801133	CC	44	1 (2.3)	43 (97.7)					
		CT	73	1 (1.4)	72 (98.6)		0.7471*			
		TT	24	1 (4.2)	23 (95.8)					
		C	117	2 (1.7)	115 (98.3)		0.4312*			
		T	97	2 (2.1)	95 (97.9)		1*			
MTR	rs1805087	AA	99	0 (0.0)	99 (100.0)					
		AG	37	3 (8.1)	34 (91.9)		0.0273*			
		GG	5	0 (0.0)	5 (100.0)					
		A	136	3 (2.2)	133 (97.8)		1*			
		G	42	3 (7.1)	39 (92.9)		0.0251*	AA	-	-
SLC19A1	rs1051266	AA	35	0 (0.0)	35 (100.0)					
		AG	64	3 (4.7)	61 (95.3)		0.3343*			
		GG	42	0 (0.0)	42 (100.0)					
		A	99	3 (3.0)	96 (97.0)		0.5545*			
		G	106	3 (2.8)	103 (97.2)		0.5741*			
XRCC1	rs1799782	CC	120	3 (2.5)	117 (97.5)					
		CT	20	0 (0.0)	20 (100.0)		1*			
		TT	1	0 (0.0)	1 (100.0)					
		C	140	3 (2.1)	137 (97.9)		1*			
		T	21	0 (0.0)	21 (100.0)		1*			
	rs25487	AA	20	0 (0.0)	20 (100.0)					
		AG	71	0 (0.0)	71 (100.0)		0.0956*			
		GG	50	3 (6.0)	47 (94.0)					
		A	91	0 (0.0)	91 (100.0)		0.0429*	A	-	-
		G	121	3 (2.5)	118 (97.5)		1*			

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

Table 43. Association of gene polymorphisms with neurotoxicity.

Gene	SNPs	Genotype	N	Neurotoxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
ABCB1	rs1045642	CC	49	0 (0.0)	49 (100.0)					
		CT	62	1 (1.6)	61 (98.4)		1*			
		TT	30	0 (0.0)	30 (100.0)					
		C	111	1 (0.0)	110 (99.1)		1*			
		T	92	1 (1.1)	91 (98.9)		1*			
	rs1128503	CC	53	0 (0.0)	53 (100.0)					
		CT	63	0 (0.0)	63 (100.0)		0.1773*			
		TT	25	1 (4.0)	24 (96.0)					
		C	116	0 (0.0)	116 (100.0)		0.1773*			
		T	88	1 (1.1)	87 (98.9)		1*			
	rs2032582	GG	47	0 (0.0)	47 (100.0)					
		G (GT/AG)	69	1 (1.4)	68 (98.6)		1*			
		T (TT/AT)	25	0 (0.0)	25 (100.0)					
		G vs TT/AT	116	1 (0.9)	115 (99.1)		1*			
		G/T vs GG	94	1 (1.1)	93 (98.9)		1*			
ERCC1	rs11615	CC	20	0 (0.0)	20 (100.0)					
		CT	72	1 (1.4)	71 (98.6)		1*			
		TT	49	0 (0.0)	49 (100.0)					
		C	92	1 (1.1)	91 (98.9)		1*			
		T	121	1 (0.8)	120 (99.2)		1*			
	rs3212986	GG	74	0 (0.0)	74 (100.0)					
		GT	55	1 (1.8)	55 (98.2)		0.4752*			
		TT	11	0 (0.0)	11 (100.0)					
		G	130	1 (0.8)	129 (99.2)		1*			
		T	67	1 (1.5)	66 (98.5)		0.4752*			
ERCC2	rs13181	GG	25	0 (0.0)	25 (100.0)					
		GT	55	1 (1.8)	54 (98.2)		0.5674*			
		TT	61	0 (0.0)	61 (100.0)					
		G	80	1 (1.2)	79 (98.8)		1*			
		T	116	1 (0.9)	115 (99.1)		1*			
	rs1799793	AA	24	0 (0.0)	24 (100.0)					
		AG	55	1 (1.8)	54 (98.2)		0.5603*			
		GG	62	0 (0.0)	62 (100.0)					
		A	79	1 (1.3)	78 (98.7)		1*			
		G	117	1 (0.9)	116 (99.1)		1*			
	rs50872	CC	82	1 (1.2)	81 (98.8)					
		CT	54	0 (0.0)	54 (100.0)		1*			
		TT	5	0 (0.0)	5 (100.0)					
		C	136	1 (0.7)	135 (99.3)		1*			
		T	59	0 (0.0)	59 (100.0)		1*			
	rs238416	AA	19	0 (0.0)	19 (100.0)					
		AG	67	1 (1.5)	66 (98.5)		1*			
		GG	55	0 (0.0)	55 (100.0)					
		A	86	1 (1.2)	85 (98.8)		1*			
		G	122	1 (0.8)	121 (99.2)		1*			
ERCC5	rs17655	CC	79	0 (0.0)	79 (100.0)					
		CG	52	1 (1.9)	51 (98.1)		0.4397*			
		GG	10	0 (0.0)	10 (100.0)					
		C	131	1 (0.8)	130 (99.2)		1*			
		G	62	1 (1.6)	61 (98.4)		0.4397*			
	rs1047768	CC	43	0 (0.0)	43 (100.0)					
		CT	70	1 (1.4)	69 (98.6)		1*			
		TT	28	0 (0.0)	28 (100.0)					
		C	113	1 (0.9)	112 (99.1)		1*			
		T	98	1 (1.0)	97 (99.0)		1*			
IL1B	rs12621220	CC	73	72 (98.6)	1 (1.4)					
		CT	61	61 (100.0)	0 (0.0)		1*			
		TT	7	7 (100.0)	0 (0.0)					
		C	134	133 (99.3)	1 (0.7)		1*			
		T	68	68 (100.0)	0 (0.0)		1*			
	rs1143623	CC	7	7 (100.0)	0 (0.0)					
		CG	61	61 (100.0)	0 (0.0)		1*			
		GG	73	72 (98.6)	1 (1.4)					
		C	68	68 (100.0)	0 (0.0)		1*			
		G	134	133 (99.3)	1 (0.7)		1*			

Table 43. (Continued)

Gene	SNPs	Genotype	N	Neurotoxicity		χ^2	p-value	Ref. Cat.	RR	95% CI
				Grade 3-4 N (%)	Grade 0-2 N (%)					
IL1B	rs16944	AA	14	14 (100.0)	0 (0.0)					
		AG	71	71 (100.0)	0 (0.0)		0.4965*			
		GG	56	55 (98.2)	1 (1.8)					
		A	85	85 (100.0)	0 (0.0)		0.3972*			
		G	127	126 (99.2)	1 (0.8)		1*			
	rs1143627	CC	15	15 (100.0)	0 (0.0)					
		CT	79	70 (100.0)	0 (0.0)		0.5035*			
		TT	56	55 (98.2)	1 81.8)					
		C	85	85 (100.0)	0 (0.0)		0.3972*			
		T	126	125 (99.2)	1 (0.8)		1*			
IL6	rs1800795	CC	17	0 (0.0)	17 (100.0)					
		CG	69	0 (0.0)	69 (100.0)		0.5106*			
		GG	55	1 (1.8)	54 (98.2)					
		C	86	0 (0.0)	86 (100.0)		0.3901*			
		G	124	1 (0.8)	123 (99.2)		1*			
IL16	rs7170924	GG	79	0 (0.0)	79 (100.0)					
		GT	52	1 (1.9)	52 (98.1)		0.4397*			
		TT	9	0 (0.0)	9 (100.0)					
		G	132	1 (0.8)	131 (99.2)		1*			
		T	62	1 (1.6)	61 (98.4)		0.4397*			
MDM2	rs1470383	CC	7	1 (14.3)	6 (85.7)					
		CT	35	0 (0.0)	35 (100.0)		0.0497*			
		TT	99	0 (0.0)	99 (100.0)					
		C	42	1 (2.4)	41 (97.6)		0.2979*			
		T	134	0 (0.0)	134 (100.0)		0.0497*	T	-	-
	rs1690924	AA	56	1 (1.8)	55 (98.2)					
		AG	65	0 (0.0)	65 (100.0)		0.5357*			
		GG	19	0 (0.0)	19 (100.0)					
		A	121	1 (0.8)	120 (99.2)		1*			
		G	84	0 (0.0)	84 (100.0)		0.4*			
MTHFR	rs1801131	AA	75	0 (0.0)	75 (100.0)					
		AC	52	1 (1.9)	52 (98.1)		0.4681*			
		CC	13	0 (0.0)	13 (100.0)					
		A	128	1 (0.8)	127 (99.2)		1*			
		C	66	1 (1.5)	65 (98.5)		0.4681*			
	rs1801133	CC	44	0 (0.0)	44 (100.0)					
		CT	73	1 (1.4)	72 (98.6)		1*			
		TT	24	0 (0.0)	24 (100.0)					
		C	117	1 (0.9)	116 (99.1)		1*			
		T	97	1 (1.0)	96 (99.0)		1*			
MTR	rs1805087	AA	99	1 (1)	98 (99)					
		AG	37	0 (0.0)	37 (100.0)		1*			
		GG	5	0 (0.0)	5 (100.0)					
		A	136	1 (0.7)	135 (99.3)		1*			
		G	42	0 (0.0)	42 (100.0)		1*			
SLC19A1	rs1051266	AA	35	0 (0.0)	35 (100.0)					
		AG	64	0 (0.0)	64 (100.0)		0.5461*			
		GG	42	1 (2.4)	41 (97.6)					
		A	99	0 (0.0)	99 (100.0)		0.2979*			
		G	106	1 (0.9)	105 (99.1)		1*			
XRCC1	rs1799782	CC	120	1 (0.8)	119 (99.2)					
		CT	20	0 (0.0)	20 (100.0)		1*			
		TT	1	0 (0.0)	1 (100.0)					
		C	140	1 (0.7)	139 (99.3)		1*			
		T	21	0 (0.0)	21 (100.0)		1*			
	rs25487	AA	20	0 (0.0)	20 (100.0)					
		AG	71	1 (1.4)	70 (98.6)		1*			
		GG	50	0 (0.0)	50 (100.0)					
		A	91	1 (1.1)	90 (98.9)		1*			
		G	121	1 (0.8)	120 (99.2)		1*			

N: number of patients; Ref Cat: reference category; RR: relative risk; 95% CI: 95% confidence interval.
*p-value for Fisher's Exact Test.

DISCUSSION

6 DISCUSSION

Chemotherapy based on platinum compounds, used as the standard treatment for NSCLC patients with wild-type EGFR or ALK translocation negative, and also as second line in NSCLC patients with ALK-positive or EGFR-mutant, presents poor clinical outcomes and is generally accompanied by severe adverse events (asthenia, gastrointestinal toxicity, hematological toxicity, neurotoxicity and nephrotoxicity) [19,45-47]. The inter-individual variability described among patients with the same clinic-pathologic characteristics may be partly explained by genetic factors. Several SNPS in different genes involved in platinum pharmacodynamics, metabolism and mechanism of action have been proposed as potential causes of this variability . In this study, 141 NSCLC patients from a single institution treated with cisplatin or carboplatin in combination with gemcitabine, paclitaxel, pemetrexed or vinorelbine were recruited to investigate the potential role of ERCC1, ERCC2, ERCC5, XRCC1, MDM2, ABCB1, MTHFR, MTR, SLC19A1, IL1B, IL6 and IL16 gene polymorphisms in chemotherapy clinical outcomes and toxicity.

ERCC1 C8092-GG genotype was associated with better response in our patients (Table 27). Previous studies have reported similar results. In Asian population, two studies with 115 and 163 patients have reported worse ORR to platinum-based chemotherapy in patients carrying the A-allele (OR=0.23; $CI_{95\%}$=0.10, 0.57 for AC/AA vs CC and OR=0.44; $CI_{95\%}$=0.27-0.74 for A vs C allele, respectively) [128,129]. In our patients, the GG genotype for XRCC1 Gln399Arg was associated with better ORR compared to those with AG/AA genotypes (Table 27). This result is in consonance with a recent meta-analysis, which evaluated 13 studies and 1.334 cases from Asian population (OR=2.05; $CI_{95\%}$=1.62-2.60; I^2=26%; $P_{heterogeneity}$=0.18; GG vs AG/AA) [62]. However, no significant association had previously been reported in Caucasian patients [110,132,142,143,154,176]. We also found that patients carrying MDM2 rs1690924-GG genotype were in higher risk to death (Table 29). To date, no other studies have found association between this polymorphism and OS [65]. However, the GG genotype for MDM2-rs1690924 has been related to lower gastrointestinal toxicity (OR=2.32; $CI_{95\%}$=1.30-4.14 for AG vs AA) in 663 Chinese NSCLC patients [65]. In our patients, we also observed that those carrying the IL1B rs16944-A allele, MTR rs1805087-A allele or SLC19A1 Arg27His-AA genotype were associated with higher risk of progression (Table 31). To date, no other studies have explored the effect of IL1B rs16944 and MTR rs1805087 on PFS in NSCLC patients treated with platinum-based. However, two studies in 101 IIIB/IV and 465 I-IV NSCLC patients failed to find an association between MTR rs1805087 and response (OR=0.66; $CI_{95\%}$=0.23-1.89 for AG/GG vs AA)

and OS (HR=0.99; $CI_{95\%}$=0.23-1.89 for AG/GG vs AA) [67,68]. The influence of SLC19A1 Arg27His on clinical outcomes of platinum-based chemotherapy has also been explored, showing no association [69-73].

The effect of ERCC1 C118T on chemotherapy outcomes in NSCLC patients has been extensively investigated, with conflicting results. Some studies have reported better ORR, OS and PFS in patients carrying the CC genotype [80,105,110,125,126,128,133-137,151], whereas others have described higher ORR, OS and PFS in patients with T-allele [105,127,129-133,138,139]. In our study, ERCC1 C118T showed no association neither with response or survival, which is in consonance with the two meta-analysis which have analyzed the compiled results of most of the other studies [60,157]. Previous results for XRCC1 Arg194Trp have reported better ORR for T-allele in Asian population, but not in Caucasian patients [81,132,186,192-195]. A recent meta-analysis, which involved 11 studies and compiled 1.329 cases, has reported similar results in Asian population (OR=0.38; $CI_{95\%}$=0.30-0.48; I^2=0%; $P_{heterogeneity}$=0.830; CT/TT vs CC) [62]. No associations between OS, PFS and XRCC1 Arg194Trp SNPs have been found [81,104,132,174,184,197-199]. In our study, this polymorphism, along with ERCC2 Lys751Gln or Asp312Asn, was not associated with platinum based chemotherapy outcomes. This lack of association of ERCC2 Lys751Gln or Asp312Asn with ORR and PFS is in consonance with a previous meta-analysis including 22 studies/3240 patients [60], which reported no association between ORR (OR=0.93; $CI_{95\%}$=0.78-1.12; I^2=0.0%; $P_{heterogeneity}$=0.707; CC/AC vs AA for ERCC2 Lys751Gln and OR=0.87; $CI_{95\%}$=0.70-1.08; I^2=44.8%; $P_{heterogeneity}$=0.041; AA/AG vs GG for ERCC2 Asp312Asn) or PFS (HR=1.08; $CI_{95\%}$=0.93-1.25; I^2=28%; $P_{heterogeneity}$=0.187; AA/AG vs GG for ERCC2 Lys751Gln and HR=1.15; $CI_{95\%}$=0.93-1.41; I^2=24.2%; $P_{heterogeneity}$=0.266; AA/AG vs GG for ERCC2 Asp312Asn). However, both SNPs have been associated with OS in several studies [110,153,173,175]. In our study, the MDM2 rs1470383 gene polymorphism was not associated with clinical outcomes of platinum-based chemotherapy. To date, the association between this SNP and response has not been evaluated, being only related to hematological toxicity to chemotherapy in an Asian study with 663 Chinese NSCLC patients (OR=4.10; $CI_{95\%}$=1.73-9.71); no association with OS and PFS was found [65]. MTHFR A1298C and C677T were not associated with clinical outcomes of platinum-based chemotherapy in our patients. Nevertheless, a previous study conducted in 1004 Chinese stage III/IV NSCLC patients has reported that carriers of MTHFR A1298C-AA genotype presented lower ORR (OR=1.52; $CI_{95\%}$=1.04-2.23 for AC vs AA) and PFS (p=0.03) [79]. A meta-analysis compiling data from 3 studies and 147 patients, both in Asian and Caucasian populations, has also shown better response in individuals with MTHFR C677T-TT genotype (OR=1.72; $CI_{95\%}$=1.01-2.93; I^2=16%; $P_{heterogeneity}$=0.31; TT vs CT/CC) [66]. Additionally, the MTHFR

C677T-TT genotype has also been associated with higher OS (p=0.026) and PFS (p=0.012) in 208 Italian stage IIIB/IV NSCLC patients [70]. The influence of IL1B rs12621220, IL1B rs1123623, IL1B rs1143627, IL16 rs7170924 on platinum-based chemotherapy clinical outcomes, which showed no significant association in the present study, has not been previously evaluated.

On the other hand, in our patients, ERCC1 C118T-T allele and ERCC2 rs50872-CC genotype acted as predictors of occurrence of at least one case of grade 3-4 adverse events, along with gemcitabine as combination agent and non-history of previous non-lung cancer (Table 33). Despite the influence of ERCC1 gene polymorphisms on toxicity has been widely investigated in NSCLC, no association has been previously reported [80,81,110,132,142,143,146,158-165]. Only a study of 81 Caucasian patients diagnosed with late-stage solid tumors (55.6% lung cancer) has reported a role of ERCC1 gene polymorphisms as predictors of cisplatin-induced nephrotoxicity [249]. In particular, patients carrying the T-allele of ERCC1 C118T polymorphism showed a 12.8% mean decrease in estimated glomerular filtration rates (p=0.047) [249]. The effect of ERCC2 rs50872 polymorphism on platinum-based chemotherapy toxicity had not been previously investigated in NSCLC, although C-allele was associated to higher median overall survival (OS) and progression-free survival (PFS) (p=0.009 and p=0.032, respectively) in 129 unresectable Asian NSCLC patients [181]. In our patients, the occurrence of multiple adverse events was influenced by ERCC2 Asp312Asn G-alelle, ABCB1 C1236T-TT genotype and IL1B rs12621220 C-allele (Table 36). Studies with large samples have not reported association between ERCC2 Asp312Asn and toxicity [110,132,142,143,146,163,164,172,173]. However, the G-allele for ERCC2 Asp312Asn was associated with lower frequency of severe hematological toxicity in 55 Polish stage IIIB-IV NSCLC patients (OR=0.08; $CI_{95\%}$=0.01-0.40; p=0.0005; AG/GG vs AA) [81]. The effect of ABCB1 C1236T polymorphism on toxicity has not previously described, although the CC genotype has been associated with lower OS and PFS in previous studies [96,97]. The influence of IL1B-rs12621220 on platinum-based chemotherapy toxicity has not either been previously investigated, although the heterozygous variant has recently been associated with decreased risk of NSCLC in patients over age 63 from a study including 889 lung cancer and 1005 controls (OR=0.71; $CI_{95\%}$=0.52-0.98; p=0.04) [250].

Since platinum based chemotherapy present a wide toxicity profile, we analyzed different subtypes of toxicity: asthenia, gastrointestinal toxicity, infection, hematological toxicity, nephrotoxicity and neurotoxicity. According to our results, patients carrying IL16 rs7170924 T-allele were in higher risk of severe hematological toxicity (OR=3.19; $CI_{95\%}$=1.35-7.97; Table 38) and asthenia (p=0.0377; Table 39). No studies have evaluated the impact of IL16 rs7170924

polymorphism on these toxicity subtypes. However, a protective effect of the IL16 rs7170924 T-allele on PFS has been reported in 651 Caucasian stage I-IV NSCLC patients (HR=0.65; $CI_{95\%}$=0.50-0.83 for GT/TT vs GG) [78]. In our patients, hematological toxicity adjusted by chemotherapy reagents was also associated with ERCC2 rs50872 polymorphism. Particularly, the CC genotype was associated with higher risk of hematological toxicity (OR=3.23; $CI_{95\%}$=1.29-8.82; Table 38). No studies have been published related to the effect of ERCC2 rs50872 polymorphism on hematological toxicity. In our patients, the CC genotypes for ERCC5 His46His or AA for SLC19A1 Arg27His demonstrated to be good prognostic factors for gastrointestinal toxicity (Table 40). However, the independency of these associations could not be analyzed, since the only three patients who presented gastrointestinal toxicity had the same genotype profile for these gene polymorphisms. The role of ERCC5 His46His on gastrointestinal toxicity has been previously investigated in 74 Spanish stage IIIA-IV NSCLC patients, but no significant association was found [132]. Nevertheless, higher ORR (OR=1.90; $CI_{95\%}$=1.10-3.28; TT vs CC), and PFS (HR=0.52; $CI_{95\%}$=0.31-0.96; TT vs CC) have been described for patients carrying the TT genotype [184]. SLC19A1 Arg27His was not associated with severe gastrointestinal toxicity in 94 Caucasian stage IIIB-IV NSCLC patients, although the GG genotype was associated with better OS (HR=1.76; $CI_{95\%}$=1.11-2.78 for AG/AA vs GG) [71]. Our patients carrying AA genotype for ERCC2 rs238416 showed greater risk of grade 3-4 infection (p=0.0177; RR=9.63; $CI_{95\%}$=1.56-59.6; Table 41). The effect of ERCC2 rs238416 polymorphism on infection has not been previously investigated, but its influence on survival has been revealed in 129 Asian unresectable NSCLC patients. In a subgroup analysis based on gemcitabine-platinum doublets, the GT genotype for ERCC2 rs238416 polymorphism showed greater OS and PFS (p=0.004 and p=0.033, respectively) [181]. Nephrotoxicity showed association with IL6 rs1800795, MTR rs1805087 and XRCC1 Gln399Arg. Particularly, carriers of IL6 rs1800795-CC, MTR rs1805087-AG/GG and XRCC1 Gln399Arg-GG genotypes had higher risk of severe nephrotoxicity (p=0.0384 for IL6 rs1800795, p=0.0251 for MTR rs1805087 and p=0.0429 for XRCC1 Gln399Arg; Table 42). No association with nephrotoxicity has been reported for IL6 rs1800795, although the CC genotype has been related to higher OS (HR=1.68; $CI_{95\%}$=1.08-2.63 for CG/GG vs CC) in 414 Portuguese stage I-IV NSCLC patients [77]. The function of MTR rs1805087 polymorphism on platinum based chemotherapy toxicity has not been previously studied and no significant association has been observed on response and survival in two studies with 101 IIIB/IV and 465 I-IV NSCLC patients [67,68]. The role of XRCC1 Gln399Arg on nephrotoxicity has been previously investigated in 55 Caucasian stage IIIB-IV NSCLC patients, but no association was found [75,76,81]. However, in consonance with our data, the GG genotype has been associated with other subtypes of toxicity, specifically, increased hematological toxicity (OR=0.32; $CI_{95\%}$=0.12-0.86 for AG/AA vs GG and OR=0.22;

$CI_{95\%}$=0.06-0.82 for AG/AA vs GG) and gastrointestinal toxicity (OR=0.29; $CI_{95\%}$=0.11-0.83 for AG/AA vs GG) [75,76,81]. On the contrary, other study in 487 Chinese stage IIIA-IV NSCLC patients has shown a protective effect of GG genotype for hematological toxicity (OR=2.14; $CI_{95\%}$=1.21-3.78 for AG/GG vs GG) [76]. MDM2 rs1470383 polymorphism was the only associated with neurotoxicity in our patients. Particularly, carriers of CC genotype had greater risk of grade 3-4 neurotoxicity (Table 43). This effect of MDM2 rs1470383 on neurotoxicity has not been previously described, although the CC genotype has previously been proposed as a risk factor for hematological chemotherapy-related toxicity (OR=4.10; $CI_{95\%}$=1.73-9.71 for CC vs TT) [65].

In our study, ERCC2 Lys751Gln polymorphism showed no influence on toxicity, in consonance with other studies developed in larger samples (up to 493 patients) [110,132,142,143,146,159,160,163,164,172,173,178,180]. However, small studies developed have reported an association with hematological toxicity and nephrotoxicity [80,81]. The A-allele for ERCC2 Lys751Gln polymorphism was associated with increased grade 2-3 neutropenia (p=0.04) in 62 Spanish stage IIIB-IV NSCLC patients [80] and severe nephrotoxicity in 55 Polish late-stage NSCLC patients (OR=0.07; $CI_{95\%}$=0.02-0.31 for AC/CC vs AA) [81]. ERCC5 Asp1104His was not either associated with toxicity in our patients, despite a previous study with 388 Chinese NSCLC patients had proposed ERCC5 rs17655 as a risk factor for infection (p=0.017) [74]. Similarly, no association was found between ABCB1 C3435T, ABCB1 Ala893Ser/Thr and XRCC1 Arg194Trp in our patients, which is in consonance with previous studies [81,97,104,132,158,163,174,184,197-199]. MDM2 rs1690924-GG genotype has been related to lower gastrointestinal toxicity in 663 Asian stage IIIA-IV NSCLC patients (OR=2.32; $CI_{95\%}$=1.30-4.14 for AG vs AA) [65]. However, MDM2 rs1690924 did not influence toxicity in our study. A similar outcome was observed for polymorphisms in MTHFR, which did not demonstrate an association with toxicity in our patients, despite a previous study in 1.004 Chinese stage III-IV NSCLC patients reported that MTHFR C677T-CC and MTHFR A1298C-AA genotype were predictors of higher hematological toxicity (OR=0.40; $CI_{95\%}$=0.19-0.85 for CT vs CC and OR=0.40; $CI_{95\%}$=0.22-2.23 for AC vs AA, respectively) [79]. The effect of IL1B rs1143623, rs16944 and rs1143627 on platinum-based chemotherapy toxicity, which showed no significant association in the present study, has not been previously evaluated.

The limitations of our study include the limited sample size, which may have been responsible from the lack of association between some polymorphisms, particularly when investigating those toxicity subtypes which occurred in a very low number of patients. However, the recruitment of a single hospital cohort, following the same therapeutic protocols by the same

team of oncologists ensured its homogeneity and reliability of the response and toxicity variables. Despite the limited sample size, the effects observed in these patients were evident. Further studies in larger cohorts will be necessary to confirm the prognostic value of some of the biomarkers, particularly ERCC1, ERCC2, XRCC1, MDM2, ABCB1, MTR, SLC19A1, IL1B and IL16 gene polymorphisms in the management of NSCLC patients.

Our results suggested that ERCC1 C118T, ERCC1 C8092A, ERCC2 Asp312Asn, ERCC2 rs50872, XRCC1 Gln399Arg, MDM2 rs1690924, ABCB1 C1236T, MTR rs1805087, SLC19A1 Arg27His, IL1B rs12621220, IL1B rs16944 and IL16 rs7170924 polymorphisms may substantially act as prognostic factors in NSCLC patients treated with platinum-based chemotherapy. These associations should be validated in a new, independent cohort of patients. Furthermore, the influence of ERCC2 rs238416, ERCC5 His46His, XRCC1 Gln399Arg, MDM2 rs1470383, MTR rs1805087, SLC19A1 Arg27His, IL6 rs1800795 gene polymorphisms on toxicity to chemotherapy in NSCLC patients should be elucidated in further studies with larger samples.

CONCLUSIONS

7 CONCLUSIONS

I. ERCC1 C8092A-GG and XRCC1 Gln399Arg-GG genotypes were associated with better ORR in NSCLC patients.

II. NSCLC patients carrying the MDM2 rs1690924-GG genotype were in higher risk of death.

III. The IL1B rs16944-A, MTR rs1805087-A alleles and the SLC19A1 rs1051266-AA genotype were associated with greater risk of progression in NSCLC.

IV. No association between ERCC1 C118T, ERCC2 Lys751Gln, ERCC2 Asp312Asn, ERCC2 rs50872, ERCC2 rs238416, ERCC5 Asp1104His, ERCC5 His46His, XRCC1 Arg194Trp, MDM2 rs1470383, ABCB1 C3435T, ABCB1 C1236T, ABCB1 Ale893Ser/Ther, MTHFR A1298C, MTHFR C677T, IL1B rs12621220, IL1B rs1143623, IL1B rs1143627, IL6 rs1800795, IL16 rs7170924 and clinical outcomes of platinum-based chemotherapy was found in our patients.

V. Patients with ERCC1 C118T-T allele and ERCC2 rs50872-CC genotype had higher risk of general toxicity for platinum-based chemotherapy.

VI. The G-allele of ERCC2 Asp312Asn, ABCB1 C1236T-TT and the IL1B rs12621220-CT/TT genotypes conferred a higher risk to present multiple adverse events in NSCLC.

VII. ERCC2 rs50872-CC genotype and IL16 rs7170924-T allele were associated with grade 3-4 hematological toxicity in NSCLC.

VIII. No influence of ERCC1 C8092A, ERCC2 Lys751Gln, ERCC5 Asp1104His, XRCC1 Arg194Trp, MDM2 rs1690924, ABCB1 C3435T, ABCB1 Ala893Ser/Thr, MTHFR A1298C, MTHFR C677T, IL1B rs1143623, IL1B rs16944, and IL1B rs1143627 on platinum-based chemotherapy toxicity was found in our patients.

REFERENCES

8 REFERENCES

1 Siegel RL, Miller KD, Jemal A. Cancer statistics, 2016. CA: A cancer journal for clinicians 2016;66:7-30.

2 Alberg AJ, Ford JG, Samet JM. Epidemiology of lung cancer: ACCP evidence-based clinical practice guidelines (2nd edition). Chest 2007;132:29s-55s.

3 Taylor R, Najafi F, Dobson A. Meta-analysis of studies of passive smoking and lung cancer: Effects of study type and continent. International Journal of Epidemiology 2007;36:1048-1059.

4 Straif K, Benbrahim-Tallaa L, Baan R, Grosse Y, Secretan B, El Ghissassi F, Bouvard V, Guha N, Freeman C, Galichet L, Cogliano V. A review of human carcinogens--Part c: Metals, arsenic, dusts, and fibres. The Lancet Oncology 2009;10:453-454.

5 Cao J, Yang C, Li J, Chen R, Chen B, Gu D, Kan H. Association between long-term exposure to outdoor air pollution and mortality in China: A cohort study. Journal of Hazardous Materials 2011;186:1594-1600.

6 Hales S, Blakely T, Woodward A. Air pollution and mortality in New Zealand: Cohort study. Journal of Epidemiology and Community Health 2012;66:468-473.

7 Katanoda K, Sobue T, Satoh H, Tajima K, Suzuki T, Nakatsuka H, Takezaki T, Nakayama T, Nitta H, Tanabe K, Tominaga S. An association between long-term exposure to ambient air pollution and mortality from lung cancer and respiratory diseases in Japan. Journal of Epidemiology 2011;21:132-143.

8 Lantz PM, Mendez D, Philbert MA. Radon, smoking, and lung cancer: The need to refocus radon control policy. American Journal of Public Health 2013;103:443-447.

9 Freudenheim JL, Ritz J, Smith-Warner SA, Albanes D, Bandera EV, van den Brandt PA, Colditz G, Feskanich D, Goldbohm RA, Harnack L, Miller AB, Rimm E, Rohan TE, Sellers TA, Virtamo J, Willett WC, Hunter DJ. Alcohol consumption and risk of lung cancer: A pooled analysis of cohort studies. The American Journal of Clinical Nutrition 2005;82:657-667.

10 The effect of vitamin e and beta carotene on the incidence of lung cancer and other cancers in male smokers. The alpha-tocopherol, beta carotene cancer prevention study group. The New England journal of medicine 1994;330:1029-1035.

11 Omenn GS, Goodman GE, Thornquist MD, Balmes J, Cullen MR, Glass A, Keogh JP, Meyskens FL, Valanis B, Williams JH, Barnhart S, Hammar S. Effects of a combination of beta carotene and vitamin a on lung cancer and cardiovascular disease. The New England Journal of Medicine 1996;334:1150-1155.

12 Yang IA, Holloway JW, Fong KM. Genetic susceptibility to lung cancer and co-morbidities. Journal of Thoracic Disease 2013;5 Suppl 5:S454-462.

13 Li J, Yang M, Li P, Su Z, Gao P, Zhang J. Idiopathic pulmonary fibrosis will increase the risk of lung cancer. Chinese Medical Journal 2014;127:3142-3149.

14 Powell HA, Iyen-Omofoman B, Baldwin DR, Hubbard RB, Tata LJ. Chronic obstructive pulmonary disease and risk of lung cancer: The importance of smoking and timing of diagnosis. Journal of Thoracic Oncology 2013;8:6-11.

15 Fauci AS. Harrison. Manual de medicina. McGraw-Hill, 2010.

16 Herbst RS HJ, Lippman SM. Lung cancer. The New England Journal of Medicine 2008;359:1367-1380.

17 Molina JR, Yang P, Cassivi SD, Schild SE, Adjei AA. Non-small cell lung cancer: Epidemiology, risk factors, treatment, and survivorship. Mayo Clinic Proceedings 2008;83:584-594.

18 Edge SB BR, Compton CC, Fritz AG, Green FL, Trotti A, editors. AJCC cancer staging manual. 7 ed. Springer-Verlag New York 2010:pp. XV–648.

19 Ettinger DS, Wood DE, Akerley W, Bazhenova LA, Borghaei H, Camidge DR, Cheney RT, Chirieac LR, D'Amico TA, Dilling TJ, Dobelbower MC, Govindan R, Hennon M, Horn L, Jahan TM, Komaki R, Lackner RP, Lanuti M, Lilenbaum R, Lin J, Loo BW, Jr., Martins R, Otterson GA, Patel JD, Pisters KM, Reckamp K, Riely GJ, Schild SE, Shapiro TA, Sharma N, Stevenson J, Swanson SJ, Tauer K, Yang SC, Gregory K, Hughes M. NCCN guidelines insights: Non-small cell lung cancer, version 4.2016. Journal of the National Comprehensive Cancer Network : JNCCN 2016;14:255-264.

20 Jordan MA, Wilson L. Microtubules as a target for anticancer drugs. Nature Reviews Cancer 2004;4:253-265.

21 Adjei AA. Pharmacology and mechanism of action of pemetrexed. Clinical Lung Cancer 2004;5 Suppl 2:S51-55.

22 Huang P, Chubb S, Hertel LW, Grindey GB, Plunkett W. Action of 2',2'-difluorodeoxycytidine on DNA synthesis. Cancer Research 1991;51:6110-6117.

23 Scagliotti GV, Parikh P, von Pawel J, Biesma B, Vansteenkiste J, Manegold C, Serwatowski P, Gatzemeier U, Digumarti R, Zukin M, Lee JS, Mellemgaard A, Park K, Patil S, Rolski J, Goksel T, de Marinis F, Simms L, Sugarman KP, Gandara D. Phase III study comparing cisplatin plus gemcitabine with cisplatin plus pemetrexed in chemotherapy-naive patients with advanced-stage non-small-cell lung cancer. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 2008;26:3543-3551.

24 Schiller JH, Harrington D, Belani CP, Langer C, Sandler A, Krook J, Zhu J, Johnson DH. Comparison of four chemotherapy regimens for advanced non-small-cell lung cancer. The New England Journal of Medicine 2002;346:92-98.

25 Zhong C, Liu H, Jiang L, Zhang W, Yao F. Chemotherapy plus best supportive care versus best supportive care in patients with non-small cell lung cancer: A meta-analysis of randomized controlled trials. PloS One 2013;8:e58466.

26 Blumenthal GM, Karuri SW, Zhang H, Zhang L, Khozin S, Kazandjian D, Tang S, Sridhara R, Keegan P, Pazdur R. Overall response rate, progression-free survival, and overall survival with targeted and standard therapies in advanced non-small-cell lung cancer: Us food and drug administration trial-level and patient-level analyses. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 2015;33:1008-1014.

27 Cardenal F, Lopez-Cabrerizo MP, Anton A, Alberola V, Massuti B, Carrato A, Barneto I, Lomas M, Garcia M, Lianes P, Montalar J, Vadell C, Gonzalez-Larriba JL, Nguyen B, Artal A, Rosell R. Randomized phase III study of gemcitabine-cisplatin versus etoposide-cisplatin in the treatment of locally advanced or metastatic non-small-cell lung cancer. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 1999;17:12-18.

28 Choi MK, Hong JY, Chang W, Kim M, Kim S, Jung HA, Lee SJ, Park S, Chung MP, Sun JM, Park K, Ahn MJ, Ahn JS. Safety and efficacy of gemcitabine or pemetrexed in combination with a platinum in patients with non-small-cell lung cancer and prior interstitial lung disease. Cancer Chemotherapy and Pharmacology 2014;73:1217-1225.

29 Crino L, Weder W, van Meerbeeck J, Felip E. Early stage and locally advanced (non-metastatic) non-small-cell lung cancer: Esmo clinical practice guidelines for diagnosis, treatment and follow-up. Annals of Oncology : official journal of the European Society for Medical Oncology / ESMO 2010;21 Suppl 5:v103-115.

30 Fenchel K, Sellmann L, Dempke WC. Overall survival in non-small cell lung cancer-what is clinically meaningful? Translational Lung Cancer Research 2016;5:115-119.

31 Gervais R, Robinet G, Clement-Duchene C, Denis F, El Kouri C, Martin P, Chouaki N, Bourayou N, Morere JF. Pemetrexed and carboplatin, an active option in first-line treatment of elderly patients with advanced non-small cell lung cancer (NSCLC): A phase II trial. Lung Cancer 2013;80:185-190.

32 Hasegawa Y, Miura D, Kitamura C, Suzuki T, Kobayashi K, Sato N. A randomized phase II trial of gemcitabine plus carboplatin: Biweekly versus standard schedules in patients with advanced non-small cell lung cancer. Chemotherapy 2013;59:346-353.

33 Ichiki M, Kawasaki M, Takayama K, Ninomiya K, Kuba M, Iwami F, Miyazaki N, Oishi K, Takeo S, Aizawa H, Nakanishi Y. A multicenter phase II study of carboplatin and paclitaxel with a biweekly schedule in patients with advanced non-small-cell lung cancer: Kyushu thoracic oncology group trial. Cancer Chemotherapy and Pharmacology 2006;58:368-373.

34 Karampeazis A, Vamvakas L, Kentepozidis N, Polyzos A, Chandrinos V, Rigas G, Christofyllakis C, Kotsakis A, Hatzidaki D, Pallis AG, Georgoulias V. Biweekly carboplatin plus gemcitabine as first-line treatment of elderly patients with advanced squamous non-small-cell lung cancer: A multicenter phase I-II trial by the hellenic oncology research group. Clinical Lung Cancer 2016

35 Kim HJ, Kim TG, Lee HJ, Kim JH, Lim BH, Seo JW, Kang EM, Lee BU, Ahn YM, Roh YH, Nam SH, Kim BS. A phase II study of combination chemotherapy with docetaxel and carboplatin for elderly patients with advanced non-small cell lung cancer. Lung Cancer 2010;68:248-252.

36 Maemondo M, Inoue A, Kobayashi K, Sugawara S, Oizumi S, Isobe H, Gemma A, Harada M, Yoshizawa H, Kinoshita I, Fujita Y, Okinaga S, Hirano H, Yoshimori K, Harada T, Ogura T, Ando M, Miyazawa H, Tanaka T, Saijo Y, Hagiwara K, Morita S, Nukiwa T. Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR. The New England Journal of Medicine 2010;362:2380-2388.

37 Schuette WH, Groschel A, Sebastian M, Andreas S, Muller T, Schneller F, Guetz S, Eschbach C, Bohnet S, Leschinger MI, Reck M. A randomized phase II study of pemetrexed in combination with cisplatin or carboplatin as first-line therapy for patients with locally advanced or metastatic non-small-cell lung cancer. Clinical Lung Cancer 2013;14:215-223.

38 Soejima K, Naoki K, Ishioka K, Nakamura M, Nakatani M, Kawada I, Watanabe H, Nakachi I, Yasuda H, Satomi R, Nakayama S, Yoda S, Ikemura S, Terai H, Sato T, Ohgino K, Arai D, Tani T, Kuroda A, Nishino M, Betsuyaku T. A phase II study of biweekly paclitaxel and carboplatin in elderly patients with advanced non-small cell lung cancer. Cancer Chemotherapy and Pharmacology 2015;75:513-519.

39 Takatani H, Nakamura Y, Nagashima S, Soda H, Kinoshita A, Fukuda M, Fukuda M, Soejima Y, Kasai T, Nakatomi K, Iida T, Oka M, Tsukamoto K, Kohno S. Phase I and II trials of vinorelbine with carboplatin for patients 75 years of age or older with previously untreated non-small-cell lung cancer. Clinical Lung Cancer 2012;13:347-351.

40 Belani CP, Einzig A, Bonomi P, Dobbs T, Capozzoli MJ, Earhart R, Cohen LJ, Luketich JD. Multicenter phase II trial of docetaxel and carboplatin in patients with stage IIIB and IV non-small-cell lung cancer. Annals of oncology : official journal of the European Society for Medical Oncology / ESMO 2000;11:673-678.

41 Ozkaya S, Findik S, Atici AG, Dirica A. Cisplatin-based chemotherapy in elderly patients with advanced stage (IIIB and IV) non-small cell lung cancer patients. Neoplasma 2011;58:348-351.

42 Ozkaya S, Findik S, Dirican A, Atici AG. Long-term survival rates of patients with stage IIIB and IV non-small cell lung cancer treated with cisplatin plus vinorelbine or gemcitabine. Experimental and Therapeutic Medicine 2012;4:1035-1038.

43 Choi PJ, Jeong SS, Yoon SS. Prognosis of recurrence after complete resection in early-stage non-small cell lung cancer. The Korean Journal of Thoracic and Cardiovascular Surgery 2013;46:449-456.

44 Xiao HQ, Tian RH, Zhang ZH, Du KQ, Ni YM. Efficacy of pemetrexed plus platinum doublet chemotherapy as first-line treatment for advanced nonsquamous non-small-cell-lung cancer: A systematic review and meta-analysis. OncoTargets and Therapy 2016;9:1471-1476.

45 Mok TS WY, Thongprasert S, Yang CH, Chu DT, Saijo N, Sunpaweravong P, Han B, Margono B, Ichinose Y, Nishiwaki Y, Ohe Y, Yang JJ, Chewaskulyong B, Jiang H, Duffield EL, Watkins CL, Armour AA, Fukuoka M. Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. The New England Journal of Medicine 2009;361:947-957.

46 Madias NE, Harrington JT. Platinum nephrotoxicity. American Journal of Medicine 1978;65:307-314.

47 Goldstein RS, Mayor GH. Minireview. The nephrotoxicity of cisplatin. Life Sciences 1983;32:685-690.

48 Albain KS, Crowley JJ, LeBlanc M, Livingston RB. Survival determinants in extensive-stage non-small-cell lung cancer: The southwest oncology group experience. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 1991;9:1618-1626.

49 Fontanini G, Bigini D, Vignati S, Basolo F, Mussi A, Lucchi M, Chine S, Angeletti CA, Harris AL, Bevilacqua G. Microvessel count predicts metastatic disease and survival in non-small cell lung cancer. The Journal of Pathology 1995;177:57-63.

50 Ichinose Y, Kato H, Koike T, Tsuchiya R, Fujisawa T, Shimizu N, Watanabe Y, Mitsudomi T, Yoshimura M. Overall survival and local recurrence of 406 completely resected stage IIIA-N2

non-small cell lung cancer patients: Questionnaire survey of the Japan Clinical Oncology Group to plan for clinical trials. Lung cancer 2001;34:29-36.

51 Ichinose Y, Yano T, Asoh H, Yokoyama H, Yoshino I, Katsuda Y. Prognostic factors obtained by a pathologic examination in completely resected non-small-cell lung cancer. An analysis in each pathologic stage. The Journal of Thoracic and Cardiovascular Surgery 1995;110:601-605.

52 Izbicki JR, Passlick B, Karg O, Bloechle C, Pantel K, Knoefel WT, Thetter O. Impact of radical systematic mediastinal lymphadenectomy on tumor staging in lung cancer. The Annals of Thoracic Surgery 1995;59:209-214.

53 Macchiarini P, Fontanini G, Hardin MJ, Chuanchieh H, Bigini D, Vignati S, Pingitore R, Angeletti CA. Blood vessel invasion by tumor cells predicts recurrence in completely resected t1 n0 m0 non-small-cell lung cancer. The Journal of Thoracic and Cardiovascular Surgery 1993;106:80-89.

54 Osaki T, Nagashima A, Yoshimatsu T, Tashima Y, Yasumoto K. Survival and characteristics of lymph node involvement in patients with N1 non-small cell lung cancer. Lung Cancer 2004;43:151-157.

55 Tanaka F, Yanagihara K, Otake Y, Yamada T, Shoji T, Miyahara R, Inui K, Wada H. Prognostic factors in patients with resected pathologic (p-) t1-2n1m0 non-small cell lung cancer (NSCLC). European Journal of Cardio-Thoracic Surgery : official journal of the European Association for Cardio-Thoracic Surgery 2001;19:555-561.

56 Thomas P, Doddoli C, Thirion X, Ghez O, Payan-Defais MJ, Giudicelli R, Fuentes P. Stage I non-small cell lung cancer: A pragmatic approach to prognosis after complete resection. The Annals of Thoracic Surgery 2002;73:1065-1070.

57 Li T, Kung HJ, Mack PC, Gandara DR. Genotyping and genomic profiling of non-small-cell lung cancer: Implications for current and future therapies. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 2013;31:1039-1049.

58 Wei HB, Lu XS, Shang LH, Xu G, Hu J, Che DH, Liu F, Wu Y, Zhang GM, Yu Y. Polymorphisms of ercc1 c118t/c8092a and mdr1 c3435t predict outcome of platinum-based chemotherapies in advanced non-small cell lung cancer: A meta-analysis. Archives of Medical Research 2011;42:412-420.

59 Wei SZ, Zhan P, Shi MQ, Shi Y, Qian Q, Yu LK, Song Y. Predictive value of ercc1 and xpd polymorphism in patients with advanced non-small cell lung cancer receiving platinum-based chemotherapy: A systematic review and meta-analysis. Medical Oncology 2011;28:315-321.

60 Yang Y, Xian L. The association between the ercc1/2 polymorphisms and the clinical outcomes of the platinum-based chemotherapy in non-small cell lung cancer (NSCLC): A systematic review and meta-analysis. Tumour Biology : the journal of the International Society for Oncodevelopmental Biology and Medicine 2014;35:2905-2921.

61 Huang SJ, Wang YF, Jin ZY, Sun JY, Guo ZL. Role of ercc1 variants in response to chemotherapy and clinical outcome of advanced non-small cell lung cancer. Tumour Biology : the journal of the International Society for Oncodevelopmental Biology and Medicine 2014;35:4023-4029.

62 Li L, Wan C, Wen FQ. Polymorphisms in the XRCC1 gene are associated with treatment response to platinum chemotherapy in advanced non-small cell lung cancer patients based on meta-analysis. Genetics and Molecular Research : GMR 2014;13:3772-3786.

63 Shen XY, Lu FZ, Wu Y, Zhao LT, Lin ZF. XRCC3 Thr241Met polymorphism and clinical outcomes of NSCLC patients receiving platinum-based chemotherapy: A systematic review and meta-analysis. PloS One 2013;8:e69553.

64 Shiraishi K, Kohno T, Tanai C, Goto Y, Kuchiba A, Yamamoto S, Tsuta K, Nokihara H, Yamamoto N, Sekine I, Ohe Y, Tamura T, Yokota J, Kunitoh H. Association of DNA repair gene polymorphisms with response to platinum-based doublet chemotherapy in patients with non-small-cell lung cancer. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 2010;28:4945-4952.

65 Qian J, Liu H, Gu S, Wu Q, Zhao X, Wu W, Wang H, Wang J, Chen H, Zhang W, Wei Q, Jin L, Lu D. Genetic variants of the MDM2 gene are predictive of treatment-related toxicities and overall survival in patients with advanced NSCLC. Clinical Lung Cancer 2015;16:e37-53.

66 Zhu N, Gong Y, He J, Xia J, Chen X. Influence of methylenetetrahydrofolate reductase C677T polymorphism on the risk of lung cancer and the clinical response to platinum-based chemotherapy for advanced non-small cell lung cancer: An updated meta-analysis. Yonsei Medical Journal 2013;54:1384-1393.

67 Cui LH YZ, Zhang TT, Shin MH, Kim HN, Choi JS. Influence of polymorphisms in MTHFR 677 C→T, TYMS 3R→2R and MTR 2756 A→G on NSCLC risk and response to platinum-based chemotherapy in advanced NSCLC. Pharmacogenomics 2011;12:797-808.

68 Matakidou A, El Galta R, Rudd MF, Webb EL, Bridle H, Eisen T, Houlston RS. Prognostic significance of folate metabolism polymorphisms for lung cancer. British Journal of Cancer 2007;97:247-252.

69 Li WJ, Jiang H, Fang XJ, Ye HL, Liu MH, Liu YW, Chen Q, Zhang L, Zhang JY, Yuan CL, Zhang QY. Polymorphisms in thymidylate synthase and reduced folate carrier (SLC19A1) genes predict survival outcome in advanced non-small cell lung cancer patients treated with pemetrexed-based chemotherapy. Oncology Letters 2013;5:1165-1170.

70 Tiseo M, Giovannetti E, Tibaldi C, Camerini A, Di Costanzo F, Barbieri F, Burgers JA, Vincent A, Peters GJ, Smit EF, Ardizzoni A. Pharmacogenetic study of patients with advanced non-small cell lung cancer (NSCLC) treated with second-line pemetrexed or pemetrexed-carboplatin. Lung Cancer 2012;78:92-99.

71 Corrigan A, Walker JL, Wickramasinghe S, Hernandez MA, Newhouse SJ, Folarin AA, Lewis CM, Sanderson JD, Spicer J, Marinaki AM. Pharmacogenetics of pemetrexed combination therapy in lung cancer: Pathway analysis reveals novel toxicity associations. The Pharmacogenomics Journal 2014;14:411-417.

72 Smit EF, Burgers SA, Biesma B, Smit HJ, Eppinga P, Dingemans AM, Joerger M, Schellens JH, Vincent A, van Zandwijk N, Groen HJ. Randomized phase II and pharmacogenetic study of pemetrexed compared with pemetrexed plus carboplatin in pretreated patients with advanced non-small-cell lung cancer. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 2009;27:2038-2045.

73 Adjei AA, Salavaggione OE, Mandrekar SJ, Dy GK, Ziegler KL, Endo C, Molina JR, Schild SE, Adjei AA. Correlation between polymorphisms of the reduced folate carrier gene (SLC19A1) and survival after pemetrexed-based therapy in non-small cell lung cancer: A north central cancer treatment group-based exploratory study. Journal of Thoracic Oncology 2010;5:1346-1353.

74 Zhang L, Gao G, Li X, Ren S, Li A, Xu J, Zhang J, Zhou C. Association between single nucleotide polymorphisms (snps) and toxicity of advanced non-small-cell lung cancer patients treated with chemotherapy. PloS One 2012;7:e48350.

75 Deng JH, Deng J, Shi DH, Ouyang XN, Niu PG. Clinical outcome of cisplatin-based chemotherapy is associated with the polymorphisms of GSTP1 and XRCC1 in advanced non-small cell lung cancer patients. Clinical & Translational Oncology : official publication of the Federation of Spanish Oncology Societies and of the National Cancer Institute of Mexico 2015;17:720-726.

76 Peng Y, Li Z, Zhang S, Xiong Y, Cun Y, Qian C, Li M, Ren T, Xia L, Cheng Y, Wang D. Association of DNA base excision repair genes (OGG1, APE1 and XRCC1) polymorphisms with outcome to platinum-based chemotherapy in advanced nonsmall-cell lung cancer patients. International Journal of Cancer 2014;135:2687-2696.

77 Gomes M, Coelho A, Araujo A, Azevedo A, Teixeira AL, Catarino R, Medeiros R. IL-6 polymorphism in non-small cell lung cancer: A prognostic value? Tumour Biology : the journal of the International Society for Oncodevelopmental Biology and Medicine 2015; 36(5):3679-84.

78 Woods NT, Monteiro AN, Thompson ZJ, Amankwah EK, Naas N, Haura EB, Beg AA, Schabath MB. Interleukin polymorphisms associated with overall survival, disease-free survival, and recurrence in non-small cell lung cancer patients. Molecular Carcinogenesis 2015; 54 Suppl 1:E172-84

79 Li X, Shao M, Wang S, Zhao X, Chen H, Qian J, Song X, Wang J, Jin L, Wu J, Li Q, Bai C, Han B, Gao Z, Lu D. Heterozygote advantage of methylenetetrahydrofolate reductase polymorphisms on clinical outcomes in advanced non-small cell lung cancer (NSCLC) patients treated with platinum-based chemotherapy. Tumour biology : the journal of the International Society for Oncodevelopmental Biology and Medicine 2014;35:11159-11170.

80 Isla D, Sarries C, Rosell R, Alonso G, Domine M, Taron M, Lopez-Vivanco G, Camps C, Botia M, Nunez L, Sanchez-Ronco M, Sanchez JJ, Lopez-Brea M, Barneto I, Paredes A, Medina B, Artal A, Lianes P. Single nucleotide polymorphisms and outcome in docetaxel-cisplatin-treated advanced non-small-cell lung cancer. Annals of oncology : official journal of the European Society for Medical Oncology / ESMO 2004;15:1194-1203.

81 Powrozek T, Mlak R, Krawczyk P, Homa I, Ciesielka M, Koziol P, Prendecka M, Milanowski J, Malecka-Massalska T. The relationship between polymorphisms of genes regulating DNA repair or cell division and the toxicity of platinum and vinorelbine chemotherapy in advanced NSCLC patients. Clinical & Translational Oncology : official publication of the Federation of Spanish Oncology Societies and of the National Cancer Institute of Mexico 2016;18:125-131.

82 Kartalou M, Essigmann JM. Recognition of cisplatin adducts by cellular proteins. Mutation Research 2001;478:1-21.

83 Jordan P, Carmo-Fonseca M. Molecular mechanisms involved in cisplatin cytotoxicity. Cell and Molecular Life Sciences 2000;57:1229-1235.

84 Sancar A. DNA repair in humans. Annual Revew of Genetics 1995;29:69-105.

85 Garcia-Campelo R, Alonso-Curbera G, Anton Aparicio LM, Rosell R. Pharmacogenomics in lung cancer: An analysis of DNA repair gene expression in patients treated with platinum-based chemotherapy. Expert Opinion on Pharmacotherapy 2005;6:2015-2026.

86 Jin S, Levine AJ. The p53 functional circuit. Journal of Cell Science 2001;114:4139-4140.

87 Michael D, Oren M. The p53-MDM2 module and the ubiquitin system. Seminars in Cancer Biology 2003;13:49-58.

88 Zheng D, Chen Y, Gao C, Wei Y, Cao G, Lu N, Hou Y, Jiang X, Wang J. Polymorphisms of p53 and MDM2 genes are associated with severe toxicities in patients with non-small cell lung cancer. Cancer Biology & Therapy 2014;15:1542-1551.

89 Dong J, Ren B, Hu Z, Chen J, Hu L, Dai J, Jin G, Xu L, Shen H. MDM2 SNP309 contributes to non-small cell lung cancer survival in Chinese. Molecular Carcinogenesis 2011;50:433-438.

90 Pu X, Hildebrandt MA, Lu C, Lin J, Stewart DJ, Ye Y, Gu J, Spitz MR, Wu X. PI3K/PTEN/AKT/mTOR pathway genetic variation predicts toxicity and distant progression in lung cancer patients receiving platinum-based chemotherapy. Lung Cancer 2011;71:82-88.

91 Lin M, Stewart DJ, Spitz MR, Hildebrandt MA, Lu C, Lin J, Gu J, Huang M, Lippman SM, Wu X. Genetic variations in the transforming growth factor-beta pathway as predictors of survival in advanced non-small cell lung cancer. Carcinogenesis 2011;32:1050-1056.

92 Manning BD, Cantley LC. AKT/PKB signaling: Navigating downstream. Cell 2007;129:1261-1274.

93 Nicholson KM, Anderson NG. The protein kinase B/AKT signalling pathway in human malignancy. Cellular Signalling 2002;14:381-395.

94 Ikushima H, Miyazono K. TGFbeta signalling: A complex web in cancer progression. Nature Reviews Cancer 2010;10:415-424.

95 Yin JY, Huang Q, Zhao YC, Zhou HH, Liu ZQ. Meta-analysis on pharmacogenetics of platinum-based chemotherapy in non small cell lung cancer (NSCLC) patients. PloS One 2012;7:e38150.

96 Weissfeld JL, Diergaarde B, Nukui T, Buch S, Pennathur A, Socinski MA, Siegfried JM, Romkes M. Inherited variation in the atp-binding cassette transporter abcb1 and survival after chemotherapy for stage III-IV lung cancer. Journal of Thoracic Oncology 2014;9:1264-1271.

97 Lamba JK, Fridley BL, Ghosh TM, Yu Q, Mehta G, Gupta P. Genetic variation in platinating agent and taxane pathway genes as predictors of outcome and toxicity in advanced non-small-cell lung cancer. Pharmacogenomics 2014;15:1565-1574.

98 Thiebaut F, Tsuruo T, Hamada H, Gottesman MM, Pastan I, Willingham MC. Cellular localization of the multidrug-resistance gene product P-glycoprotein in normal human tissues. Proceedings of the National Academy of Sciences USA 1987;84:7735-7738.

99 Ambudkar SV, Dey S, Hrycyna CA, Ramachandra M, Pastan I, Gottesman MM. Biochemical, cellular, and pharmacological aspects of the multidrug transporter. Annual Review of Pharmacology and Toxicology 1999;39:361-398.

100 Borst P, Elferink RO. Mammalian abc transporters in health and disease. Annual Review of Biochemistry 2002;71:537-592.

101 Jones PM, George AM. The abc transporter structure and mechanism: Perspectives on recent research. Cellular and Molecular Life Sciences 2004;61:682-699.

102 Sakaeda T, Nakamura T, Okumura K. MDR1 genotype-related pharmacokinetics and pharmacodynamics. Biological and Pharmaceutical Bulletin 2002;25:1391-1400.

103 Takara K, Sakaeda T, Okumura K. An update on overcoming mdr1-mediated multidrug resistance in cancer chemotherapy. Current Pharmaceutical Design 2006;12:273-286.

104 Han B, Guo Z, Ma Y, Kang S, Wang Y, Wei Q, Wu X. Association of GSTP1 and XRCC1 gene polymorphisms with clinical outcome of advanced non-small cell lung cancer patients with cisplatin-based chemotherapy. International Journal of Clinical and Experimental Pathology 2015;8:4113-4119.

105 Lv H, Han T, Shi X, Yao Y, Yao Y, Qiu W, Yue L, Liang J. Genetic polymorphism of gstp1 and ercc1 correlated with response to platinum-based chemotherapy in non-small cell lung cancer. Medical Oncology 2014;31:86.

106 Sun N, Sun X, Chen B, Cheng H, Feng J, Cheng L, Lu Z. MRP2 and GSTP1 polymorphisms and chemotherapy response in advanced non-small cell lung cancer. Cancer Chemotherapy and Pharmacology 2010;65:437-446.

107 Wu G, Jiang B, Liu X, Shen Y, Yang S. Association of GSTs gene polymorphisms with treatment outcome of advanced non-small cell lung cancer patients with cisplatin-based chemotherapy. International Journal of Clinical and Experimental Pathology 2015;8:13346-13352.

108 Zhou F, Yu Z, Jiang T, Lv H, Yao R, Liang J. Genetic polymorphisms of GSTP1 and XRCC1: Prediction of clinical outcome of platinum-based chemotherapy in advanced non-small cell lung cancer (NSCLC) patients. Swiss Medical Weekly 2011;141:w13275.

109 Booton R, Ward T, Heighway J, Ashcroft L, Morris J, Thatcher N. Glutathione-S-transferase P1 isoenzyme polymorphisms, platinum-based chemotherapy, and non-small cell lung cancer. Journal of Thoracic Oncology 2006;1:679-683.

110 Joerger M, Burgers SA, Baas P, Smit EF, Haitjema TJ, Bard MP, Doodeman VD, Smits PH, Vincent A, Huitema AD, Beijnen JH, Schellens JH. Germline polymorphisms in patients with advanced nonsmall cell lung cancer receiving first-line platinum-gemcitabine chemotherapy: A prospective clinical study. Cancer 2012;118:2466-2475.

111 Li YN, Gulati S, Baker PJ, Brody LC, Banerjee R, Kruger WD. Cloning, mapping and RNA analysis of the human methionine synthase gene. Hum Mol Genet 1996;5:1851-1858.

112 Goyette P, Sumner JS, Milos R, Duncan AM, Rosenblatt DS, Matthews RG, Rozen R. Human methylenetetrahydrofolate reductase: Isolation of cDNA, mapping and mutation identification. Nature Genetics 1994;7:195-200.

113 Chu J, Dolnick BJ. Natural antisense (rTSalpha) rna induces site-specific cleavage of thymidylate synthase mRNA. Biochimica et Biophysica Acta 2002;1587:183-193.

114 Hum DW, Bell AW, Rozen R, MacKenzie RE. Primary structure of a human trifunctional enzyme. Isolation of a cDNA encoding methylenetetrahydrofolate dehydrogenase-methenyltetrahydrofolate cyclohydrolase-formyltetrahydrofolate synthetase. The Journal of Biological Chemistry 1988;263:15946-15950.

115 Chiao JH, Roy K, Tolner B, Yang CH, Sirotnak FM. RFC-1 gene expression regulates folate absorption in mouse small intestine. The Journal of Biological Chemistry 1997;272:11165-11170.

116 Pollard JW. Tumour-educated macrophages promote tumour progression and metastasis. Nat Reviews Cancer 2004;4:71-78.

117 Topping RP, Wilkinson JC, Scarpinato KD. Mismatch repair protein deficiency compromises cisplatin-induced apoptotic signaling. The Journal of Biological Chemistry 2009;284:14029-14039.

118 Akyuz N, Boehden GS, Susse S, Rimek A, Preuss U, Scheidtmann KH, Wiesmuller L. DNA substrate dependence of p53-mediated regulation of double-strand break repair. Molecular and Cellular Biology 2002;22:6306-6317.

119 Offer H, Zurer I, Banfalvi G, Reha'k M, Falcovitz A, Milyavsky M, Goldfinger N, Rotter V. p53 modulates base excision repair activity in a cell cycle-specific manner after genotoxic stress. Cancer Research 2001;61:88-96.

120 Yamane K, Katayama E, Tsuruo T. P53 contains a DNA break-binding motif similar to the functional part of BRCT-related region of Rb. Oncogene 2001;20:2859-2867.

121 Zhu Q, Wani MA, El-Mahdy M, Wani AA. Decreased DNA repair efficiency by loss or disruption of p53 function preferentially affects removal of cyclobutane pyrimidine dimers from non-transcribed strand and slow repair sites in transcribed strand. The Journal of Biological Chemistry 2000;275:11492-11497.

122 de Laat WL, Appeldoorn E, Jaspers NG, Hoeijmakers JH. DNA structural elements required for ERCC1-XPF endonuclease activity. The Journal of Biological Chemistry 1998;273:7835-7842.

123 Coin F, Marinoni JC, Rodolfo C, Fribourg S, Pedrini AM, Egly JM. Mutations in the XPD helicase gene result in XP and TTD phenotypes, preventing interaction between XPD and the p44 subunit of TFIIH. Nature Genetics 1998;20:184-188.

124 Sijbers AM, de Laat WL, Ariza RR, Biggerstaff M, Wei YF, Moggs JG, Carter KC, Shell BK, Evans E, de Jong MC, Rademakers S, de Rooij J, Jaspers NG, Hoeijmakers JH, Wood RD. Xeroderma pigmentosum group F caused by a defect in a structure-specific DNA repair endonuclease. Cell 1996;86:811-822.

125 Su D, Ma S, Liu P, Jiang Z, Lv W, Zhang Y, Deng Q, Smith S, Yu H. Genetic polymorphisms and treatment response in advanced non-small cell lung cancer. Lung Cancer 2007;56:281-288.

126 Cheng J, Ha M, Wang Y, Sun J, Chen J, Wang Y, Tong C. A C118T polymorphism of ERCC1 and response to cisplatin chemotherapy in patients with late-stage non-small cell lung cancer. Journal of Cancer Research and Clinical Oncology 2012;138:231-238.

127 Wang J, Zhang Q, Zhang H, Wang Q, Yang X, Gu Y, Zhang S. [association between polymorphisms of ERCC1 and response in patients with advanced non-small cell lung cancer

receiving cisplatin-based chemotherapy]. Zhongguo Fei Ai Za Zhi = Chinese Journal of Lung Cancer 2010;13:337-341.

128 Gao H, Ge RC, Liu HY, Wang Y, Yan S. Effect of ERCC1 polymorphism on the response to chemotherapy and clinical outcome of non-small cell lung cancer. Genetics and Molecular Research : GMR 2014;13:8997-9004.

129 Li F, Sun X, Sun N, Qin S, Cheng H, Feng J, Chen B, Cheng L, Lu Z, Ji J, Zhou Y. Association between polymorphisms of ERCC1 and XPD and clinical response to platinum-based chemotherapy in advanced non-small cell lung cancer. American Journal of Clinical Oncology 2010;33:489-494.

130 Du Y, Su T, Zhao L, Tan X, Chang W, Zhang H, Cao G. Associations of polymorphisms in DNA repair genes and MDR1 gene with chemotherapy response and survival of non-small cell lung cancer. PloS One 2014;9:e99843.

131 Shi ZH, Shi GY, Liu LG. Polymorphisms in ERCC1 and XPF gene and response to chemotherapy and overall survival of non-small cell lung cancer. International Journal of Clinical and Experimental Pathology 2015;8:3132-3137.

132 Sullivan I, Salazar J, Majem M, Pallares C, Del Rio E, Paez D, Baiget M, Barnadas A. Pharmacogenetics of the DNA repair pathways in advanced non-small cell lung cancer patients treated with platinum-based chemotherapy. Cancer Letters 2014;353:160-166.

133 Zhao X, Zhang Z, Yuan Y, Yuan X. Polymorphisms in ERCC1 gene could predict clinical outcome of platinum-based chemotherapy for non-small cell lung cancer patients. Tumour Biology : the journal of the International Society for Oncodevelopmental Biology and Medicine 2014;35:8335-8341.

134 Dong J, Hu Z, Shu Y, Pan S, Chen W, Wang Y, Hu L, Jiang Y, Dai J, Ma H, Jin G, Shen H. Potentially functional polymorphisms in DNA repair genes and non-small-cell lung cancer survival: A pathway-based analysis. Molecular Carcinogenesis 2012;51:546-552.

135 Okuda K, Sasaki H, Hikosaka Y, Kawano O, Yukiue H, Yano M, Fujii Y. Excision repair cross complementation group 1 polymorphisms predict overall survival after platinum-based chemotherapy for completely resected non-small-cell lung cancer. The Journal of Surgical Research 2011;168:206-212.

136 Park SY, Hong YC, Kim JH, Kwak SM, Cho JH, Lee HL, Ryu JS. Effect of ERCC1 polymorphisms and the modification by smoking on the survival of non-small cell lung cancer patients. Medical Oncology 2006;23:489-498.

137 Ryu JS, Hong YC, Han HS, Lee JE, Kim S, Park YM, Kim YC, Hwang TS. Association between polymorphisms of ERCC1 and XPD and survival in non-small-cell lung cancer patients treated with cisplatin combination chemotherapy. Lung Cancer 2004;44:311-316.

138 Zhou C, Ren S, Zhou S, Zhang L, Su C, Zhang Z, Deng Q, Zhang J. Predictive effects of ercc1 and xrcc3 snp on efficacy of platinum-based chemotherapy in advanced NSCLC patients. Japanese Journal of Clinical Oncology 2010;40:954-960.

139 Ren S, Zhou S, Wu F, Zhang L, Li X, Zhang J, Xu J, Lv M, Zhang J, Zhou C. Association between polymorphisms of DNA repair genes and survival of advanced NSCLC patients treated with platinum-based chemotherapy. Lung Cancer 2012;75:102-109.

140 Takenaka T, Yano T, Kiyohara C, Miura N, Kouso H, Ohba T, Kometani T, Shoji F, Yoshino I, Maehara Y. Effects of excision repair cross-complementation group 1 (ERCC1) single nucleotide polymorphisms on the prognosis of non-small cell lung cancer patients. Lung Cancer 2010;67:101-107.

141 Zhou W, Gurubhagavatula S, Liu G, Park S, Neuberg DS, Wain JC, Lynch TJ, Su L, Christiani DC. Excision repair cross-complementation group 1 polymorphism predicts overall survival in advanced non-small cell lung cancer patients treated with platinum-based chemotherapy. Clinical Cancer Research : an official journal of the American Association for Cancer Research 2004;10:4939-4943.

142 Kalikaki A, Kanaki M, Vassalou H, Souglakos J, Voutsina A, Georgoulias V, Mavroudis D. DNA repair gene polymorphisms predict favorable clinical outcome in advanced non-small-cell lung cancer. Clinical Lung Cancer 2009;10:118-123.

143 Kalikaki A, Voutsina A, Koutsopoulos A, Papadaki C, Sfakianaki M, Yachnakis E, Xyrafas A, Kotsakis A, Agelaki S, Souglakos J, Mavroudis D, Georgoulias V. ERCC1 SNPs as potential predictive biomarkers in non-small cell lung cancer patients treated with platinum-based chemotherapy. Cancer Investigation 2015;33:107-113.

144 Olaussen KA, Dunant A, Fouret P, Brambilla E, Andre F, Haddad V, Taranchon E, Filipits M, Pirker R, Popper HH, Stahel R, Sabatier L, Pignon JP, Tursz T, Le Chevalier T, Soria JC. DNA repair by ERCC1 in non-small-cell lung cancer and cisplatin-based adjuvant chemotherapy. The New England Journal of Medicine 2006;355:983-991.

145 Park DJ, Zhang W, Stoehlmacher J, Tsao-Wei D, Groshen S, Gil J, Yun J, Sones E, Mallik N, Lenz HJ. ERCC1 gene polymorphism as a predictor for clinical outcome in advanced colorectal cancer patients treated with platinum-based chemotherapy. Clinical Advances in Hematology & Oncology 2003;1:162-166.

146 Tibaldi C, Giovannetti E, Vasile E, Mey V, Laan AC, Nannizzi S, Di Marsico R, Antonuzzo A, Orlandini C, Ricciardi S, Del Tacca M, Peters GJ, Falcone A, Danesi R. Correlation of CDA, ERCC1, and XPD polymorphisms with response and survival in gemcitabine/cisplatin-treated advanced non-small cell lung cancer patients. Clinical Cancer Research : an official journal of the American Association for Cancer Research 2008;14:1797-1803.

147 de las Penas R, Sanchez-Ronco M, Alberola V, Taron M, Camps C, Garcia-Carbonero R, Massuti B, Queralt C, Botia M, Garcia-Gomez R, Isla D, Cobo M, Santarpia M, Cecere F, Mendez P, Sanchez JJ, Rosell R. Polymorphisms in DNA repair genes modulate survival in cisplatin/gemcitabine-treated non-small-cell lung cancer patients. Annals of oncology : official journal of the European Society for Medical Oncology / ESMO 2006;17:668-675.

148 Mathiaux J, Le Morvan V, Pulido M, Jougon J, Begueret H, Robert J. Role of DNA repair gene polymorphisms in the efficiency of platinum-based adjuvant chemotherapy for non-small cell lung cancer. Molecular Diagnosis & Therapy 2011;15:159-166.

149 Hong W, Wang K, Zhang YP, Kou JY, Hong D, Su D, Mao WM, Yu XM, Xie FJ, Wang XJ. Methylenetetrahydrofolate reductase C677T polymorphism predicts response and time to progression to gemcitabine-based chemotherapy for advanced non-small cell lung cancer in a Chinese Han population. Journal of Zhejiang University Science B 2013;14:207-215.

150 Krawczyk P, Kucharczyk T, Kowalski DM, Powrozek T, Ramlau R, Kalinka-Warzocha E, Winiarczyk K, Knetki-Wroblewska M, Wojas-Krawczyk K, Kalakucka K, Dyszkiewicz W, Krzakowski

M, Milanowski J. Polymorphisms in TS, MTHFR and ERCC1 genes as predictive markers in first-line platinum and pemetrexed therapy in NSCLC patients. Journal of Cancer Research and Clinical Oncology 2014;140:2047-2057.

151 Krawczyk P, Wojas-Krawczyk K, Mlak R, Kucharczyk T, Biernacka B, Milanowski J. Predictive value of ERCC1 single-nucleotide polymorphism in patients receiving platinum-based chemotherapy for locally-advanced and advanced non-small cell lung cancer--a pilot study. Folia Histochemica et Cytobiologica / Polish Academy of Sciences, Polish Histochemical and Cytochemical Society 2012;50:80-86.

152 Li D, Zhou Q, Liu Y, Yang Y, Li Q. DNA repair gene polymorphism associated with sensitivity of lung cancer to therapy. Medical Oncology 2012;29:1622-1628.

153 Liao WY, Shih JY, Chang GC, Cheng YK, Yang JC, Chen YM, Yu CJ. Genetic polymorphism of XRCC1 Arg399Gln is associated with survival in non-small-cell lung cancer patients treated with gemcitabine/platinum. Journal of Thoracic Oncology 2012;7:973-981.

154 Tiseo M, Bordi P, Bortesi B, Boni L, Boni C, Baldini E, Grossi F, Recchia F, Zanelli F, Fontanini G, Naldi N, Campanini N, Azzoni C, Bordi C, Ardizzoni A. ERCC1/BRCA1 expression and gene polymorphisms as prognostic and predictive factors in advanced NSCLC treated with or without cisplatin. British journal of cancer 2013;108:1695-1703.

155 Zhang ZY, Tian X, Wu R, Liang Y, Jin XY. Predictive role of ERCC1 and XPD genetic polymorphisms in survival of Chinese non-small cell lung cancer patients receiving chemotherapy. Asian Pacific Journal of Cancer Prevention : APJCP 2012;13:2583-2586.

156 Wei HB, Hu J, Shang LH, Zhang YY, Lu FF, Wei M, Yu Y. A meta-analytic review of ERCC1/MDR1 polymorphism and chemosensitivity to platinum in patients with advanced non-small cell lung cancer. Chinese Medical Journal 2012;125:2902-2907.

157 Huang D, Zhou Y. Nucleotide excision repair gene polymorphisms and prognosis of non-small cell lung cancer patients receiving platinum-based chemotherapy: A meta-analysis based on 44 studies. Biomedical Reports 2014;2:452-462.

158 Chen S, Huo X, Lin Y, Ban H, Lin Y, Li W, Zhang B, Au WW, Xu X. Association of MDR1 and ERCC1 polymorphisms with response and toxicity to cisplatin-based chemotherapy in non-small-cell lung cancer patients. International Journal of Hygiene and Environmental Health 2010;213:140-145.

159 Gandara DR, Kawaguchi T, Crowley J, Moon J, Furuse K, Kawahara M, Teramukai S, Ohe Y, Kubota K, Williamson SK, Gautschi O, Lenz HJ, McLeod HL, Lara PN, Jr., Coltman CA, Jr., Fukuoka M, Saijo N, Fukushima M, Mack PC. Japanese-US common-arm analysis of paclitaxel plus carboplatin in advanced non-small-cell lung cancer: A model for assessing population-related pharmacogenomics. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 2009;27:3540-3546.

160 Ludovini V, Floriani I, Pistola L, Minotti V, Meacci M, Chiari R, Garavaglia D, Tofanetti FR, Flacco A, Siggillino A, Baldelli E, Tonato M, Crino L. Association of cytidine deaminase and xeroderma pigmentosum group D polymorphisms with response, toxicity, and survival in cisplatin/gemcitabine-treated advanced non-small cell lung cancer patients. Journal of Thoracic Oncology 2011;6:2018-2026.

161 Metro G, Chiari R, Mare M, Giannarelli D, Tofanetti FR, Minotti V, Ferraldeschi M, Giuffrida D, Marcomigni L, Bennati C, Fischer MJ, Meacci M, Bellavita R, Pistola L, Ludovini V, Crino L. Carboplatin plus pemetrexed for platinum-pretreated, advanced non-small cell lung cancer: A retrospective study with pharmacogenetic evaluation. Cancer Chemotherapy and Pharmacology 2011;68:1405-1412.

162 Suk R, Gurubhagavatula S, Park S, Zhou W, Su L, Lynch TJ, Wain JC, Neuberg D, Liu G, Christiani DC. Polymorphisms in ERCC1 and grade 3 or 4 toxicity in non-small cell lung cancer patients. Clinical Cancer Research : an official journal of the American Association for Cancer Research 2005;11:1534-1538.

163 Vinolas N, Provencio M, Reguart N, Cardenal F, Alberola V, Sanchez-Torres JM, Baron FJ, Cobo M, Maestu I, Moreno I, Mesia C, Izquierdo A, Felip E, Lopez-Brea M, Marquez A, Sanchez-Ronco M, Taron M, Santarpia MC, Rosell R. Single nucleotide polymorphisms in MDR1 gen correlates with outcome in advanced non-small-cell lung cancer patients treated with cisplatin plus vinorelbine. Lung Cancer 2011;71:191-198.

164 Iranzo V, Sirera R, Bremnes RM, Blasco A, Jantus-Lewintre E, Taron M, Berrocal A, Blasco S, Caballero C, Del Pozo N, Rosell R, Camps C. Chemotherapy-induced neutropenia does not correlate with DNA repair gene polymorphisms and treatment efficacy in advanced non-small-cell lung cancer patients. Clinical Lung Cancer 2011;12:224-230.

165 KimCurran V, Zhou C, Schmid-Bindert G, Shengxiang R, Zhou S, Zhang L, Zhang J. Lack of correlation between ERCC1 (C8092A) single nucleotide polymorphism and efficacy/toxicity of platinum based chemotherapy in Chinese patients with advanced non-small cell lung cancer. Advances in Medical Sciences 2011;56:30-38.

166 Wang Z, Xu Y, Zhu S. Interleukin-16 rs4778889 polymorphism contributes to the development of renal cell cancer in a Chinese population. International Journal of Clinical and Experimental Pathology 2015;8:15228-15233.

167 Duell EJ, Wiencke JK, Cheng TJ, Varkonyi A, Zuo ZF, Ashok TD, Mark EJ, Wain JC, Christiani DC, Kelsey KT. Polymorphisms in the DNA repair genes XRCC1 and ERCC2 and biomarkers of DNA damage in human blood mononuclear cells. Carcinogenesis 2000;21:965-971.

168 Lunn RM, Helzlsouer KJ, Parshad R, Umbach DM, Harris EL, Sanford KK, Bell DA. XPD polymorphisms: Effects on DNA repair proficiency. Carcinogenesis 2000;21:551-555.

169 Booton R, Ward T, Heighway J, Taylor P, Power F, Ashcroft L, Morris J, Thatcher N. Xeroderma pigmentosum group D haplotype predicts for response, survival, and toxicity after platinum-based chemotherapy in advanced non-small cell lung cancer. Cancer 2006;106:2421-2427.

170 Camps C, Sarries C, Roig B, Sanchez JJ, Queralt C, Sancho E, Martinez N, Taron M, Rosell R. Assessment of nucleotide excision repair XPD polymorphisms in the peripheral blood of gemcitabine/cisplatin-treated advanced non-small-cell lung cancer patients. Clinical Lung Cancer 2003;4:237-241.

171 Li XD, Han JC, Zhang YJ, Li HB, Wu XY. Common variations of DNA repair genes are associated with response to platinum-based chemotherapy in NSCLCs. Asian Pacific Journal of Cancer Prevention : APJCP 2013;14:145-148.

172 Provencio M, Camps C, Cobo M, De las Penas R, Massuti B, Blanco R, Alberola V, Jimenez U, Delgado JR, Cardenal F, Taron M, Ramirez JL, Sanchez A, Rosell R. Prospective assessment of XRCC3, XPD and Aurora kinase A single-nucleotide polymorphisms in advanced lung cancer. Cancer Chemotherapy and Pharmacology 2012;70:883-890.

173 Wu W, Li H, Wang H, Zhao X, Gao Z, Qiao R, Zhang W, Qian J, Wang J, Chen H, Wei Q, Han B, Lu D. Effect of polymorphisms in XPD on clinical outcomes of platinum-based chemotherapy for Chinese non-small cell lung cancer patients. PloS One 2012;7:e33200.

174 Zhang L, Ma W, Li Y, Wu J, Shi GY. Pharmacogenetics of DNA repair gene polymorphisms in non-small-cell lung carcinoma patients on platinum-based chemotherapy. Genetics and Molecular Research : GMR 2014;13:228-236.

175 Zhou M, Ding YJ, Feng Y, Zhang QR, Xiang Y, Wan HY. Association of xeroderma pigmentosum group D (Asp312Asn, Lys751Gln) and cytidine deaminase (Lys27gln, Ala70thr) polymorphisms with outcome in Chinese non-small cell lung cancer patients treated with cisplatin-gemcitabine. Genetics and Molecular Research : GMR 2014;13:3310-3318.

176 Giachino DF, Ghio P, Regazzoni S, Mandrile G, Novello S, Selvaggi G, Gregori D, DeMarchi M, Scagliotti GV. Prospective assessment of XPD Lys751Gln and XRCC1 Arg399Gln single nucleotide polymorphisms in lung cancer. Clinical Cancer Research : an official journal of the American Association for Cancer Research 2007;13:2876-2881.

177 Yao CY, Huang XE, Li C, Shen HB, Shi MQ, Feng JF, Pan LX, Tang JH. Lack of influence of XRCC1 and XPD gene polymorphisms on outcome of platinum-based chemotherapy for advanced non small cell lung cancers. Asian Pacific Journal of Cancer Prevention : APJCP 2009;10:859-864.

178 Cheng H, Qin Q, Sun X, Li F, Sun N, Cheng L, Lu Z, Chen B. Predictive effect of XPA and XPD polymorphisms on survival of advanced NSCLC patients treated with platinum-based chemotherapy: A three-dimensional (3-D), polyacrylamide gel-based DNA microarray method. Technology in Cancer Research & Treatment 2013;12:473-482.

179 Liu L, Yuan P, Wu C, Zhang X, Wang F, Guo H, Zhong R, Xu Y, Wu J, Yu D, Wu T, Zhang X, Nie S, Miao X, Lin D. Assessment of XPD Lys751Gln and XRCC1 T-77C polymorphisms in advanced non-small-cell lung cancer patients treated with platinum-based chemotherapy. Lung Cancer 2011;73:110-115.

180 Li Y, Huang XE, Jin GF, Shen HB, Xu L. Lack of any relationship between chemotherapy toxicity in non-small cell lung cancer cases and polymorphisms in XRCC1 codon 399 or XPD codon 751. Asian Pacific Journal of Cancer Prevention : APJCP 2011;12:739-742.

181 Kim SH, Lee GW, Lee MJ, Cho YJ, Jeong YY, Kim HC, Lee JD, Hwang YS, Kim IS, Lee S, Oh SY. Clinical significance of ERCC2 haplotype-tagging single nucleotide polymorphisms in patients with unresectable non-small cell lung cancer treated with first-line platinum-based chemotherapy. Lung Cancer 2012;77:578-584.

182 Chen X, Sun H, Ren S, Kim Curran V, Zhang L, Zhou S, Zhang J, Zhou C. Association of XRCC3 and XPD751 SNP with efficacy of platinum-based chemotherapy in advanced NSCLC patients. Clinical & Translational Oncology : official publication of the Federation of Spanish Oncology Societies and of the National Cancer Institute of Mexico 2012;14:207-213.

183 O'Donovan A, Davies AA, Moggs JG, West SC, Wood RD. XPG endonuclease makes the 3' incision in human DNA nucleotide excision repair. Nature 1994;371:432-435.

184 Liu D, Wu J, Shi GY, Zhou HF, Yu Y. Role of XRCC1 and ERCC5 polymorphisms on clinical outcomes in advanced non-small cell lung cancer. Genetics and Molecular Research : GMR 2014;13:3100-3107.

185 Hu W, Pan J, Zhao P, Yang G, Yang S. Genetic polymorphisms in XPG could predict clinical outcome of platinum-based chemotherapy for advanced non-small cell lung cancer. Tumour Biology : the journal of the International Society for Oncodevelopmental Biology and Medicine 2014;35:5561-5567.

186 Sun X, Li F, Sun N, Shukui Q, Baoan C, Jifeng F, Lu C, Zuhong L, Hongyan C, YuanDong C, Jiazhong J, Yingfeng Z. Polymorphisms in XRCC1 and XPG and response to platinum-based chemotherapy in advanced non-small cell lung cancer patients. Lung Cancer 2009;65:230-236.

187 Yuli Y, Zhe S, Xia W, Siqing L, Zhenxuan W, Yu-Hua Z, Bing S, Jun-Wei C. XPG is a novel biomarker of clinical outcome in advanced non-small-cell lung cancer. Pakistan Journal of Medical Sciences 2013;29:762-767.

188 Zharkov DO. Base excision DNA repair. Cellular and Molecular Life Sciences 2008;65:1544-1565.

189 Liu Y, Prasad R, Beard WA, Kedar PS, Hou EW, Shock DD, Wilson SH. Coordination of steps in single-nucleotide base excision repair mediated by apurinic/apyrimidinic endonuclease 1 and DNA polymerase beta. The Journal of Biological Chemistry 2007;282:13532-13541.

190 Masson M, Niedergang C, Schreiber V, Muller S, Menissier-de Murcia J, de Murcia G. XRCC1 is specifically associated with poly(ADP-ribose) polymerase and negatively regulates its activity following DNA damage. Molecular and Cellular Biology 1998;18:3563-3571.

191 Whitehouse CJ, Taylor RM, Thistlethwaite A, Zhang H, Karimi-Busheri F, Lasko DD, Weinfeld M, Caldecott KW. XRCC1 stimulates human polynucleotide kinase activity at damaged DNA termini and accelerates DNA single-strand break repair. Cell 2001;104:107-117.

192 Hong CY, Xu Q, Yue Z, Zhang Y, Yuan Y. [Correlation of the sensitivity of NP chemotherapy in non-small lung cancer with DNA repair gene XRCC1 polymorphism]. Ai zheng = Aizheng = Chinese Journal of Cancer 2009;28:1291-1297.

193 Wang ZH, Miao XP, Tan W, Zhang XR, Xu BH, Lin DX. [Single nucleotide polymorphisms in XRCC1 and clinical response to platin-based chemotherapy in advanced non-small cell lung cancer]. Ai Zheng = Aizheng = Chinese Journal of Cancer 2004;23:865-868.

194 Xu C, Wang X, Zhang Y, Li L. [Effect of the XRCC1 and XRCC3 genetic polymorphisms on the efficacy of platinum-based chemotherapy in patients with advanced non-small cell lung cancer]. Zhongguo Fei Ai Za Zhi = Chinese Journal of Lung Cancer 2011;14:912-917.

195 Yuan P, Miao XP, Zhang XM, Wang ZH, Tan W, Sun Y, Zhang XR, Xu BH, Lin DX. [XRCC1 and XPD genetic polymorphisms predict clinical responses to platinum-based chemotherapy in advanced non-small cell lung cancer]. Zhonghua Zhong Liu Za Zhi = Chinese Journal of Oncology 2006;28:196-199.

196 Lee SY, Kang HG, Yoo SS, Kang YR, Choi YY, Lee WK, Choi JE, Jeon HS, Shin KM, Oh IJ, Kim KS, Lee J, Cha SI, Kim CH, Kim YC, Park JY. Polymorphisms in DNA repair and apoptosis-related genes and clinical outcomes of patients with non-small cell lung cancer treated with first-line paclitaxel-cisplatin chemotherapy. Lung Cancer 2013;82:330-339.

197 Zhao W, Hu L, Xu J, Shen H, Hu Z, Ma H, Shu Y, Shao Y, Yin Y. Polymorphisms in the base excision repair pathway modulate prognosis of platinum-based chemotherapy in advanced non-small cell lung cancer. Cancer Chemotherapy and Pharmacology 2013;71:1287-1295.

198 Ke HG, Li J, Shen Y, You QS, Yan Y, Dong HX, Liu JH, Shen ZY. Prognostic significance of GSTP1, XRCC1 and XRCC3 polymorphisms in non-small cell lung cancer patients. Asian Pacific Journal of Cancer Prevention : APJCP 2012;13:4413-4416.

199 Liu JY, Liu QM, Li LR. Association of GSTP1 and XRCC1 gene polymorphisms with clinical outcomes of patients with advanced non-small cell lung cancer. Genetics and Molecular Research : GMR 2015;14:10331-10337.

200 Shrivastav M, De Haro LP, Nickoloff JA. Regulation of DNA double-strand break repair pathway choice. Cell Research 2008;18:134-147.

201 Brenneman MA, Wagener BM, Miller CA, Allen C, Nickoloff JA. Xrcc3 controls the fidelity of homologous recombination: Roles for XRCC3 in late stages of recombination. Molecular Cell 2002;10:387-395.

202 Butkiewicz D, Drosik A, Suwinski R, Krzesniak M, Rusin M, Kosarewicz A, Rachtan J, Matuszczyk I, Gawkowska-Suwinska M. Influence of DNA repair gene polymorphisms on prognosis in inoperable non-small cell lung cancer patients treated with radiotherapy and platinum-based chemotherapy. International Journal of Cancer Journal International Du Cancer 2012;131:E1100-1108.

203 Yin M, Liao Z, Huang YJ, Liu Z, Yuan X, Gomez D, Wang LE, Wei Q. Polymorphisms of homologous recombination genes and clinical outcomes of non-small cell lung cancer patients treated with definitive radiotherapy. PloS One 2011;6:e20055.

204 Jin ZY, Zhao XT, Zhang LN, Wang Y, Yue WT, Xu SF. Effects of polymorphisms in the XRCC1, XRCC3, and XPG genes on clinical outcomes of platinum-based chemotherapy for treatment of non-small cell lung cancer. Genetics and Molecular Research : GMR 2014;13:7617-7625.

205 Fitch ME, Cross IV, Ford JM. p53 responsive nucleotide excision repair gene products p48 and XPC, but not p53, localize to sites of UV-irradiation-induced DNA damage, in vivo. Carcinogenesis 2003;24:843-850.

206 Seo YR, Fishel ML, Amundson S, Kelley MR, Smith ML. Implication of p53 in base excision DNA repair: In vivo evidence. Oncogene 2002;21:731-737.

207 Amaral JD, Xavier JM, Steer CJ, Rodrigues CM. The role of p53 in apoptosis. Discovery Medicine 2010;9:145-152.

208 Vousden KH, Lane DP. p53 in health and disease. Nature Reviews Molecular Cell Biology 2007;8:275-283.

209 Weston A, Perrin LS, Forrester K, Hoover RN, Trump BF, Harris CC, Caporaso NE. Allelic frequency of a p53 polymorphism in human lung cancer. Cancer Epidemiology, Biomarkers & Prevention : a publication of the American Association for Cancer Research, cosponsored by the American Society of Preventive Oncology 1992;1:481-483.

210 Dumont P, Leu JI, Della Pietra AC, 3rd, George DL, Murphy M. The codon 72 polymorphic variants of p53 have markedly different apoptotic potential. Nature Genetics 2003;33:357-365.

211 Bergamaschi D, Gasco M, Hiller L, Sullivan A, Syed N, Trigiante G, Yulug I, Merlano M, Numico G, Comino A, Attard M, Reelfs O, Gusterson B, Bell AK, Heath V, Tavassoli M, Farrell PJ, Smith P, Lu X, Crook T. p53 polymorphism influences response in cancer chemotherapy via modulation of p73-dependent apoptosis. Cancer Cell 2003;3:387-402.

212 Vikhanskaya F, Siddique MM, Kei Lee M, Broggini M, Sabapathy K. Evaluation of the combined effect of p53 codon 72 polymorphism and hotspot mutations in response to anticancer drugs. Clinical Cancer Research : an official journal of the American Association for Cancer Research 2005;11:4348-4356.

213 Perez-Ramirez C, Canadas-Garre M, Molina MA, Faus-Dader MJ, Calleja-Hernandez MA. PTEN and PI3K/AKT in non-small-cell lung cancer. Pharmacogenomics 2015;16:1843-1862.

214 Stoyanov B, Volinia S, Hanck T, Rubio I, Loubtchenkov M, Malek D, Stoyanova S, Vanhaesebroeck B, Dhand R, Nurnberg B, et al. Cloning and characterization of a G protein-activated human phosphoinositide-3 kinase. Science (New York, NY) 1995;269:690-693.

215 Rameh LE, Cantley LC. The role of phosphoinositide 3-kinase lipid products in cell function. The Journal of Biological Chemistry 1999;274:8347-8350.

216 Sansal I, Sellers WR. The biology and clinical relevance of the pten tumor suppressor pathway. Journal of Clinical Oncology : official journal of the American Society of Clinical Oncology 2004;22:2954-2963.

217 Cantley LC. The phosphoinositide 3-kinase pathway. Science (New York, NY) 2002;296:1655-1657.

218 Duronio V. The life of a cell: Apoptosis regulation by the PI3K/PKB pathway. The Biochemical Journal 2008;415:333-344.

219 Engelman JA, Luo J, Cantley LC. The evolution of phosphatidylinositol 3-kinases as regulators of growth and metabolism. Nature Reviews Genetics 2006;7:606-619.

220 Kim MJ, Kang HG, Lee SY, Jeon HS, Lee WK, Park JY, Lee EB, Lee JH, Cha SI, Kim DS, Kim CH, Kam S, Jung TH, Park JY. AKT1 polymorphisms and survival of early stage non-small cell lung cancer. Journal of Surgical Oncology 2012;105:167-174.

221 Hoffmeyer S, Burk O, von Richter O, Arnold HP, Brockmoller J, Johne A, Cascorbi I, Gerloff T, Roots I, Eichelbaum M, Brinkmann U. Functional polymorphisms of the human multidrug-resistance gene: Multiple sequence variations and correlation of one allele with P-glycoprotein expression and activity in vivo. Proceedings of the National Academy of Sciences of the United States of America 2000;97:3473-3478.

222 Pan JH, Han JX, Wu JM, Huang HN, Yu QZ, Sheng LJ. MDR1 single nucleotide polymorphism G2677T/A and haplotype are correlated with response to docetaxel-cisplatin

chemotherapy in patients with non-small-cell lung cancer. Respiration; International Review of Thoracic Diseases 2009;78:49-55.

223 Pan JH, Han JX, Wu JM, Sheng LJ, Huang HN, Yu QZ. MDR1 single nucleotide polymorphisms predict response to vinorelbine-based chemotherapy in patients with non-small cell lung cancer. Respiration; International Review of Thoracic Diseases 2008;75:380-385.

224 Yan PW, Huang XE, Yan F, Xu L, Jiang Y. Influence of MDR1 gene codon 3435 polymorphisms on outcome of platinum-based chemotherapy for advanced non small cell lung cancer. Asian Pacific Journal of Cancer Prevention : APJCP 2011;12:2291-2294.

225 Peklak-Scott C, Smitherman PK, Townsend AJ, Morrow CS. Role of glutathione S-transferase P1-1 in the cellular detoxification of cisplatin. Molecular Cancer Therapeutics 2008;7:3247-3255.

226 Ekhart C, Rodenhuis S, Smits PH, Beijnen JH, Huitema AD. An overview of the relations between polymorphisms in drug metabolising enzymes and drug transporters and survival after cancer drug treatment. Cancer Treatment Reviews 2009;35:18-31.

227 Ou C, Zhu B, Zhao HL, Huang LS, Li PZ, Lao M, Li LQ. Association of glutathione S-transferase P1 gene polymorphism with the risk of small-cell carcinoma of lung cancer. Journal of Receptor and Signal Transduction Research 2015;35:165-169.

228 Singh S. Cytoprotective and regulatory functions of glutathione S-transferases in cancer cell proliferation and cell death. Cancer Chemotherapy and Pharmacology 2015;75:1-15.

229 Watson MA, Stewart RK, Smith GB, Massey TE, Bell DA. Human glutathione S-transferase P1 polymorphisms: Relationship to lung tissue enzyme activity and population frequency distribution. Carcinogenesis 1998;19:275-280.

230 Yang P, Mandrekar SJ, Hillman SH, Allen Ziegler KL, Sun Z, Wampfler JA, Cunningham JM, Sloan JA, Adjei AA, Perez E, Jett JR. Evaluation of glutathione metabolic genes on outcomes in advanced non-small cell lung cancer patients after initial treatment with platinum-based chemotherapy: An NCCTG-97-24-51 based study. Journal of Thoracic Oncology 2009;4:479-485.

231 Bailey LB, Gregory JF, 3rd. Folate metabolism and requirements. The Journal of Nutrition 1999;129:779-782.

232 Wang L, Shangguan S, Chang S, Yu X, Wang Z, Lu X, Wu L, Zhang T. Determining the association between methylenetetrahydrofolate reductase (MTHFR) gene polymorphisms and genomic DNA methylation level: A meta-analysis. Birth Defects Research Part A: Clinical and Molecular Teratology 2016

233 Hanks J, Ayed I, Kukreja N, Rogers C, Harris J, Gheorghiu A, Liu CL, Emery P, Pufulete M. The association between MTHFR 677C>T genotype and folate status and genomic and gene-specific DNA methylation in the colon of individuals without colorectal neoplasia. The American Journal of Clinical Nutrition 2013;98:1564-1574.

234 Yamada K, Chen Z, Rozen R, Matthews RG. Effects of common polymorphisms on the properties of recombinant human methylenetetrahydrofolate reductase. Proceedings of the National Academy of Sciences of the United States of America 2001;98:14853-14858.

235 Izmirli M. A literature review of MTHFR (C677T and A1298C polymorphisms) and cancer risk. Molecular Biology Reports 2013;40:625-637.

236 Song MA, Brasky TM, Marian C, Weng DY, Taslim C, Llanos AA, Dumitrescu RG, Liu Z, Mason JB, Spear SL, Kallakury BV, Freudenheim JL, Shields PG. Genetic variation in one-carbon metabolism in relation to genome-wide DNA methylation in breast tissue from heathy women. Carcinogenesis 2016

237 Leclerc D, Campeau E, Goyette P, Adjalla CE, Christensen B, Ross M, Eydoux P, Rosenblatt DS, Rozen R, Gravel RA. Human methionine synthase: cDNA cloning and identification of mutations in patients of the cblG complementation group of folate/cobalamin disorders. Human Molecular Genetics 1996;5:1867-1874.

238 Sharp L, Little J. Polymorphisms in genes involved in folate metabolism and colorectal neoplasia: A huge review. American Journal of Epidemiology 2004;159:423-443.

239 Azzoli CG, Baker S, Temin S, Pao W, Aliff T, Brahmer J, Johnson DH, Laskin JL, Masters G, Milton D. American society of clinical oncology clinical practice guideline update on chemotherapy for stage IV non–small-cell lung cancer. Journal of Clinical Oncology 2009;27:6251-6266.

240 Coussens LM, Werb Z. Inflammation and cancer. Nature 2002;420:860-867.

241 Lin WW, Karin M. A cytokine-mediated link between innate immunity, inflammation, and cancer. The Journal of Clinical Investigation 2007;117:1175-1183.

242 Aggarwal BB, Vijayalekshmi RV, Sung B. Targeting inflammatory pathways for prevention and therapy of cancer: Short-term friend, long-term foe. Clinical Cancer Research : an official journal of the American Association for Cancer Research 2009;15:425-430.

243 Dranoff G. Cytokines in cancer pathogenesis and cancer therapy. Nature Reviews Cancer 2004;4:11-22.

244 Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, Rubinstein L, Shankar L, Dodd L, Kaplan R, Lacombe D, Verweij J. New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). European Journal of Cancer 2009;45:228-247.

245 Health UDo, Services H. Common terminology criteria for adverse events (CTCAE) version 4.0. National Institutes of Health, National Cancer Institute 2009;4

246 Benlloch S, Botero ML, Beltran-Alamillo J, Mayo C, Gimenez-Capitan A, de Aguirre I, Queralt C, Ramirez JL, Ramon y Cajal S, Klughammer B, Schlegel M, Bordogna W, Chen D, Zhang G, Kovach B, Shieh F, Palma JF, Wu L, Lawrence HJ, Taron M. Clinical validation of a PCR assay for the detection of EGFR mutations in non-small-cell lung cancer: Retrospective testing of specimens from the EURTAC trial. PloS One 2014;9:e89518.

247 Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MAR, Bender D, Maller J, Sklar P, de Bakker PIW, MJ D, PC S. PLINK: A toolset for whole-genome association and population-based linkage analysis. American Journal of Human Genetics 2007;81

248 Team RC. R: A language and environment for statistical computing. [internet]. 2013 oct. Available from: http://www.r-project.org/.

249 Tzvetkov MV BG, O'Brien VP, Hohloch K, Brockmöller J, Benöhr P. Pharmacogenetic analyses of cisplatin-induced nephrotoxicity indicate a renoprotective effect of ERCC1 polymorphisms. Pharmacogenomics 2011;12:1417-1427.

250 Li Y, Zhao W, Zhao Z, Wu J, Chen L, Ma Y, Li Q, Lu D, Jin L, Wang J. IL1B gene polymorphisms, age and the risk of non-small cell lung cancer in a Chinese population. Lung Cancer 2015;89:232-237.

ANNEXES

9 ANNEXES

9.1 Annexe 1

Servicio Andaluz de Salud
CONSEJERÍA DE SALUD

D. Miguel Ángel Calleja Hernández Secretario del Comité de Ética de la Investigación de Centro de Granada (CEI-GRANADA)

CERTIFICA

Que este Comité ha analizado la propuesta de Dª. Cristina Perez Ramirez para que se realice el proyecto de investigacion titulado: "Detección de marcadores genéticos implicados en la respuesta y toxicidad al tratamiento en pacientes de cáncer de pulmón no microcítico" y considera que:

Se cumplen los requisitos necesarios de idoneidad del proyecto en relación con los objetivos del estudio.

La capacidad del investigador y los medios disponibles son apropiados para llevar a cabo el estudio.

Entendiendo que dicho estudio se ajusta a las normas éticas esenciales y criterios deontológicos que rigen en este centro.

Y que este Comité acepta que dicho estudio sea realizado por Dª Cristina Pérez Ramírez investigador principal en el mismo y colaboradores.

Lo que firmo en Granada a veinticinco de julio de dos mil doce.

Dr. Miguel Ángel Calleja Hernández

CONSEJERÍA DE SALUD
Comité de Ética de la Investigación
Servicio Andaluz de Salud

C.S. 771

Hospital Universitario Virgen de las Nieves
Avda. Fuerzas Armadas, 2. 18014 - Granada
Tel.: 958 02 00 00. Fax: 958 02 01 32

9.2 Annexe 2

HOSPITAL UNIVERSITARIO VIRGEN DE LAS NIEVES
Unidad de Farmacogenética
Servicio de Farmacia. U.G.C. Farmacia
HMI 4ª pta.
Avda. Fuerzas Armadas 2
18014 Granada

CONSENTIMIENTO INFORMADO DEL PACIENTE PARA ANÁLISIS FARMACOGENÉTICOS

Nombre del paciente:
Nº Historia Clínica:
NUHSA:

1. Yo..... declaro bajo mi responsabilidad que he leído y comprendo la Hoja de Información del proyecto **"Detección de marcadores genéticos implicados en la respuesta y toxicidad al tratamiento en pacientes de cáncer de pulmón no microcítico"** y acepto participar.
2. Se me ha entregado una copia de la Hoja de Información al paciente y una copia de este consentimiento informado, fechado y firmado.
3. Comprendo las características y el objetivo del estudio de Seguimiento Farmacoterapéutico complementado con análisis farmacogenético.
4. He recibido suficiente información sobre la donación de muestras biológicas de ADN.
5. Se me ha dado tiempo y oportunidad para realizar preguntas.
6. Comprendo que mi participación es voluntaria.
7. Comprendo que soy libre de retirarme del análisis en cualquier momento. Tras ello se procederá a la destrucción de la muestra codificada. Si se hubiera retirado previamente el vínculo de identificación de la muestra, no se podrá relacionar conmigo, de forma que no se podrá destruir.
8. Entiendo que los resultados del mismo se comunicarán sólo en caso de que dichos hallazgos tengan una implicación significativa para la salud de los participantes y que exista una posibilidad de mejorar su condición de salud.

Punto 1.- Yo DOY / No DOY mi consentimiento voluntariamente para que se pueda realizar el análisis farmacogenético a mi muestra de ADN y tratamiento farmacológico en la Unidad de Farmacogenética del Hospital Universitario Virgen de las Nieves.

Punto 2.- Yo DOY / No DOY mi consentimiento voluntariamente para ceder el remanente de mi muestra de ADN al Biobanco de Células, Tejidos y Tumores del Hospital Universitario Virgen de las Nieves.

Deseo que dichas muestras y los datos clínicos asociados sean tratados de forma (marcar una opción):

☐ **CODIFICADA**: Identificadas con un código que protege mi identidad, siendo posible volver a ligarlas conmigo.

☐ **ANONIMIZADA**: Con desvinculación irreversible de la identidad. No se podrán asociar las muestras conmigo.

Deseo **establecer restricciones** respecto al uso de la muestra, para que no sea utilizada en .....

Autorizo para que se pueda **contactar conmigo posteriormente**
En caso afirmativo, por favor, indique el medio de hacerlo: .....

Fecha: ...../...../..... **Firma del paciente**:.....

Representante legal en caso de incapacidad del paciente, con indicación del carácter con el que interviene (padre, madre, tutor, etc.).
Nombre del Representante Legal .....

DNI ..... **Firma** .....

Persona que proporciona la información y el consentimiento:

Nombre .....

DNI ..... **Firma** .....

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9.3 Annexe 3

HOSPITAL UNIVERSITARIO VIRGEN DE LAS NIEVES
Unidad de Farmacogenética
Servicio de Farmacia. U.G.C. Farmacia
HMI 4ª pta.
Avda. Fuerzas Armadas 2
18014 Granada

HOJA DE INFORMACIÓN AL PACIENTE
PROYECTOS DE INVESTIGACIÓN

Titulo del proyecto: Detección de marcadores genéticos implicados en la respuesta y toxicidad al tratamiento en pacientes de cáncer de pulmón no microcítico.

PROMOTOR DEL PROYECTO: Unidad de Farmacogenética – Servicio de Farmacia Hospitalaria
INVESTIGADOR RESPONSABLE DEL PROYECTO: Cristina Pérez Ramírez

Objetivos: El objetivo global del proyecto es evaluar la presencia de los marcadores genéticos implicados en la respuesta y toxicidad al tratamiento de los pacientes diagnosticados con cáncer de pulmón no microcítico (NSCLC). Se realizará además un seguimiento farmacoterapéutico de estos pacientes, con el objetivo de medir la frecuencia de problemas relacionados con los medicamentos y de resultados negativos de la medicación en pacientes con NSCLC tratados con inhibidores de EGFR o platinos.

Procedimientos: Deseo participar en este estudio y conozco que:

- Tendré citas cada 3 meses con el farmacéutico investigador durante 1 año de seguimiento, coincidiendo con mis citas de seguimiento clínico.
- Se me realizará una extracción de 3 mL de sangre en la primera visita, para analizar los marcadores farmacogenéticos y su relación con mi respuesta al tratamiento prescrito. Esta muestra sólo se utilizará para los fines exclusivos de esta investigación.
- Los marcadores genéticos que se determinan en tejido, se realizarán utilizando parte de la muestra obtenida en la punción de aguja fina (PAAF) y/o cirugía, obtenidas con fines diagnósticos. La determinación de estos marcadores no necesitará de una intervención adicional sobre el paciente.
- En caso de no autorizar la cesión de las muestras al Biobanco de Células, Tejidos y Tumores del Hospital Universitario Virgen de las Nieves (Respuesta Negativa al Punto 2 del Consentimiento Informado), esta muestra sólo se utilizará para los fines exclusivos de esta investigación.
- En caso de autorizar que los remanentes de las muestras pasen a formar parte del Biobanco de Células, Tejidos y Tumores del Hospital Universitario Virgen de las Nieves (Respuesta Positiva al Punto 2 del Consentimiento Informado), la muestra sólo podrá ser utilizada en proyectos de investigación científicamente avalados, que cumplan las exigencias legales y los principios éticos que rigen la investigación en salud (Ley 14/2007, de 3 de julio, de Investigación Biomédica) y que sean autorizados por los órganos competentes, de conformidad con lo establecido en la normativa vigente. En este caso, se renuncia a cualquier derecho de naturaleza económica, patrimonial o potestativa sobre los resultados o potenciales beneficios que puedan derivarse de manera directa o indirecta de las investigaciones que se lleven a cabo con la muestra que cede para investigación.

Beneficios: Pueden no obtenerse beneficios directos con la participación en este proyecto, aunque también puede mejorar el estado de salud del paciente por disponer de la contribución de otro profesional de la salud, el farmacéutico, que es el especialista en medicamentos. Además, tendrá información sobre todos los medicamentos de su tratamiento, así como la posibilidad de aclarar las dudas que tenga sobre ellos. En caso de autorizar la cesión de remanentes de muestras al Biobanco, es posible que la información obtenida de las investigaciones en las que se utilicen sus muestras no le genere un beneficio directo, pero habrá contribuido al avance de la medicina y del conocimiento de diversas enfermedades, lo que repercutirá en un beneficio para la sociedad.

Riesgos: Con respecto al Seguimiento Farmacoterapéutico, no tendrá riesgos, pues sólo consiste en responder una serie de preguntas. Para el estudio Farmacogenético, es necesario una muestra de saliva o una extracción de sangre, por lo que el riesgo en este último caso es el asociado a la extracción de sangre. La extracción de la sangre puede provocar una sensación de ardor en el punto en que se introduce la aguja en la piel y puede ocasionar un pequeño hematoma o una leve infección, que desaparecen en pocos días. Algunas personas pueden experimentar un leve mareo en el momento de la extracción.

Lugar de realización del análisis y destino de la muestra al término de la investigación: Los análisis farmacogenéticos de este estudio se llevarán a cabo en la Unidad de Farmacogenética del Hospital Universitario Virgen de las Nieves. Al término de la investigación, se cederán al Biobanco de Células, Tejidos y Tumores del Hospital Universitario Virgen de las Nieves las muestras de aquellos pacientes que así lo hayan autorizado con una Respuesta Positiva al Punto 2 del Consentimiento Informado. Las muestras de aquellos pacientes que sólo acepten

participar en este estudio, pero no autoricen la donación de su muestra al Biobanco (Respuesta Negativa al Punto 2 del Consentimiento Informado) se considerarán destinadas exclusivamente a fines de investigación, y por tanto se conservarán únicamente en tanto sean necesarias para los fines que justificaron su recogida. Por tanto, serán destruidas a la finalización del mismo. Los datos genéticos de carácter personal de estos pacientes se conservarán durante un período mínimo de cinco años desde la fecha en que fueron obtenidos, transcurrido el cual el interesado podrá solicitar su cancelación, de acuerdo con la Ley 14/2007, de 3 de julio, de Investigación Biomédica.

Posibilidad de ponerse nuevamente en contacto: Puede que sea necesario ponerse en contacto nuevamente con usted, con el fin de recabar datos o muestras adicionales, o proporcionarle la información relevante para su salud, salvo que haya solicitado que las muestras sean anonimizadas.

Confidencialidad: Toda la información obtenida en este estudio es confidencial y será estrictamente utilizada para fines de investigación. Los datos personales serán protegidos e incluidos en un fichero que deberá estar sometido a la Ley Orgánica 15/1999 de 13 de Diciembre. Asimismo, se podrá solicitar en todo momento la información y resultados obtenidos de esta investigación relacionados con su persona, siempre que la muestra no se encuentre anonimizada. Podrá ejercer los derechos de acceso, rectificación, oposición y cancelación de los datos personales, reconocidos en la citada Ley Orgánica 15/1999, con las limitaciones establecidas en dicha Ley. Para ello, deberá dirigirse a la Dirección General de Asistencia Sanitaria del Servicio Andaluz de Salud, Avenida de la Constitución, nº18, Sevilla.

Información sobre resultados del Estudio: Los resultados de la investigación, conforme normativa vigente, se harán públicos mediante difusión y posterior publicación en prensa científica, sin que se facilite ningún dato que identifique al paciente.

En el caso de cesión al Biobanco, éste tendrá a disposición del donante toda la información sobre los proyectos de investigación en los que se utilice su muestra. El comité de ética externo del biobanco o el Comité de Ética de la Investigación que evaluó el proyecto de investigación decidirán en qué casos será imprescindible que se envíe la información de manera individualizada.

Derecho de recusa o desistencia: La participación en el estudio es totalmente voluntaria, siendo libre para retirarse de la investigación en cualquier momento sin que afecte o ponga en riesgo su asistencia médica.

El consentimiento prestado para cesión de muestras al Biobanco podrá ser retirado o revocado en cualquier momento, excepto si las muestras se encuentran anonimizadas. Para ello, deberá dirigirse al Biobanco, pudiendo solicitar la eliminación o la anonimización de las muestras. Los efectos de esta revocación no se extenderán a los resultados de las investigaciones llevadas a cabo con anterioridad.

El farmacéutico **Cristina Pérez Ramírez** le ha comentado toda esa información, poniéndose a disposición del paciente para contestar a cualquier duda que tenga, ya sea por teléfono (958-020-108) o en el Servicio de Farmacia del Hospital Universitario Virgen de las Nieves.

Para consultas relacionadas con el Biobanco, podrá dirigirse al Biobanco de Células, Tejidos y Tumores del Hospital Universitario Virgen de las Nieves, o a su Representante Legal, el Dr. Ángel Concha López, en la dirección de correo electrónico angel.concha.sspa@juntadeandalucia.es. En caso de producirse un eventual cierre del Biobanco o revocación de la autorización para su constitución y funcionamiento, la información sobre el destino de las muestras estará a su disposición en el Registro Nacional de Biobancos para Investigación Biomédica, con el fin de que pueda manifestar su conformidad o disconformidad con el destino previsto para las muestras.

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PUBLICATIONS

10 PUBLICATIONS

The results of this thesis has been partially published as an original article in the journal *Pharmacological Research* (Impact factor: 4.816; Category: Pharmacology and Pharmacy, 27/255; Quartile 1).

Pérez-Ramírez C, Cañadas-Garre M, Alnatsha A, Villar E, Delgado JR, Faus-Dáder MJ, Calleja-Hernández MA. **Pharmacogenetic predictors of toxicity to platinum based chemotherapy in non-small cell lung cancer patients.** Pharmacol Res. 2016 Sep; 111:877-84. doi: 10.1016/j.phrs.2016.08.002. Epub 2016 Aug 3.

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Pharmacogenetic predictors of toxicity to platinum based chemotherapy in non-small cell lung cancer patients

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ABSTRACT

Platinum-based chemotherapy is the standard treatment for NSCLC patients with EGFR wild-type, and as alternative to failure to EGFR inhibitors. However, this treatment is aggressive and most patients experience grade 3–4 toxicities. ERCC1, ERCC2, ERCC5, XRCC1, MDM2, ABCB1, MTHFR, MTR, SLC19A1, IL6 and IL16 gene polymorphisms may contribute to individual variation in toxicity to chemotherapy. The aim of this study was to evaluate the effect of these polymorphisms on platinum-based chemotherapy in NSCLC patients. A prospective cohorts study was conducted, including 141 NSCLC patients. Polymorphisms were analyzed by PCR Real-Time with Taqman® probes and sequencing. Patients with ERCC1 C118T-T allele ($p = 0.00345$; RR = 26.05; $CI_{95\%} = 4.33, 515.77$) and ERCC2 rs50872-CC genotype ($p = 0.00291$; RR = 4.06; $CI_{95\%} = 1.66, 10.65$) had higher risk of general toxicity for platinum-based chemotherapy. ERCC2 Asp312Asn G-alelle, ABCB1 C1236T-TT and the IL1B rs12621220-CT/TT genotypes conferred a higher risk to present multiple adverse events. The subtype toxicity analysis also revealed that ERCC2 rs50872-CC genotype ($p = 0.01562$; OR = 3.23; $CI_{95\%} = 1.29, 8.82$) and IL16 rs7170924-T allele ($p = 0.01007$; OR = 3.19; $CI_{95\%} = 1.35, 7.97$) were associated with grade 3–4 hematological toxicity. We did not found the influence of ERCC1 C8092A, ERCC2 Lys751Gln, ERCC2 Asp312Asn, ERCC5 Asp1104His, XRCC1 Arg194Trp, MDM2 rs1690924, ABCB1 C3435T, ABCB1 Ala893Ser/Thr, MTHFR A1298C, MTHFR C677T, IL1B rs1143623, IL1B rs16944, and IL1B rs1143627 on platinum-based chemotherapy toxicity. In conclusion, ERCC1 C118T, ERCC2 rs50872, ERCC2 Asp312Asn, ABCB1 C1236T, IL1B rs12621220 and IL16 rs7170924 polymorphisms may substantially act as prognostic factors in NSCLC patients treated with platinum-based chemotherapy.

1.

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The most important types of lung cancer are small cell lung cancer and non-small cell lung cancer (NSCLC). NSCLC accounts for approximately 80–85% of all lung cancer cases and is divided into different subtypes: squamous cell carcinoma, adenocarcinoma and large cell carcinoma. At the time of diagnosis, most patients with NSCLC have advanced stage (IIIB–IV), according to American Joint Committee on Cancer (AJCC) [2–4]. Therefore, five-year survival is low with rates of 5% for IIIB and 1% for IV stages [2–4].

Platinum-based doublet-chemotherapy is the standard treatment for NSCLC for EGFR wild-type patients, and as second line in mutated EGFR patients [5]. It is frequently given together with other agents, such as anti-microtubule agents (taxanes and vinca alkaloids), antifolate agents (pemetrexed), or pyrimidine antagonists (gemcitabine). Platinum-based chemotherapy has showed benefits in terms of survival (10.7 months vs 3.9 months, respectively; $p < 0.001$) and symptom control compared with best supportive care [6,7]. However, it is a very aggressive treatment, which presents high percentages of severe adverse events, such as asthenia (44.0%), gastrointestinal toxicity (33.3%), hematological toxicity (67.1%), neurotoxicity (69.9%) and nephrotoxicity (20–30%) [8–10]. This toxicity profile varies from person to person. Various studies have reported that this inter-individual differences may be due to genetic factors, such as single nucleotide polymorphisms (SNPs), which are involved in platinum pharmacodynamics, metabolism and mechanism of action [11–26].

Cisplatin and carboplatin are the main platinum compounds used on NSCLC therapy. They are heavy metal complexes that interact with DNA, forming platinum-DNA adducts, which result in severe local distortions of the DNA double helix [27,28]. Therefore, this interaction leads to DNA damage, inhibiting DNA replication and transcription and inducing apoptosis. Several pathways are activated in response to this interaction, which include DNA repair and p53 pathways. Deactivation of platinum drugs increase the activity of DNA repair pathways, which involve nucleotide-excision repair (NER), base excision repair (BER), and double-strand break repair (DSB). There are a great variety of proteins involved in detecting and repairing these adducts, such as excision repair cross-complementing group 1 (ERCC1), excision repair cross-complementation group 2 (ERCC2, also known as XPD), excision repair cross-complementation group 5 (ERCC5) and X-ray repair complementing defective repair in Chinese hamster cells 1 (XRCC1) [29,30]. Cell cycle control and apoptosis initiation is mediated by p53 pathway [31]. MDM2 proto-oncogene, E3 ubiquitin protein ligase (MDM2) plays a crucial role in this pathway, because it interacts with p53, leading to its ubiquination and degradation [32]. Genetic alterations, such as SNPs in any of this genes may modulate repair function and apoptosis, promoting individual variation in the toxicity to platinum-based chemotherapy [11–15].

Other mechanisms involved in platinum toxicity are drug transporters, folate metabolism and cytokine signaling [16–24]. Drug transporters are responsible of pumping out the cell platinum compounds [33–35]. The main gene involved in this process is ATP-binding cassette, sub-family B (MDR/TAP), member 1 (ABCB1, also called MDR1). Polymorphisms in this gene may alter its function and expression, leading to an accumulation of platinum drugs outside the cells [36]. Thus, genetic alterations in this gene may affect the inter-individual toxicity profile of platinum-based chemotherapy. Folate metabolism also plays an essential role on platinum cytotoxicity. Polymorphisms in genes involved in this pathway, such as methylenetetrahydrofolate reductase (MTHFR) and methionine synthase (MTR) modify methylation of DNA [37–41]. In fact, genetic alterations in these genes have been associated with lower enzyme activity and its results have been correlated with DNA hypomethylation, which alter sensitivity of tumor cells to platinum compounds [23,25,26,42]. Other gene involved in folate metabolism is the solute carrier family 19 (folate transporter), members 1 (SLC19A1). This protein transports folate drugs into the cell, such as pemetrexed, a drug that is frequently given in combination with platinum compounds [20,43]. Polymorphisms in this gene may alter the cellular entry of this drug and subsequently affects the cytotoxicity of platinum-pemetrexed based chemotherapy [18–22]. Citokine signaling has also showed an association with tumor progression [44,45]. Several studies have reported a connection between chronic inflammation and early stage of neoplastic development [46]. Innate immune cells are activated to battle infection as a physiological process. However, if this damaged becomes chronic, it could lead to a continuous cellular proliferation and subsequently initiates metaplasia and dysplasia [44,45]. A family of cytokines, which are named interleukins (ILs), induce growth, differentiation and activation of immune cells [47,48]. Moreover, they inhibit apoptosis of malignant cells at the site of inflammation [48]. In NSCLC, IL1B, IL6 and IL16 have recently showed a relevant impact on clinical outcomes for patients treated with platinum-based chemotherapy [16,17].

Based on above, we conducted this study to evaluate the effects of ERCC1, ERCC2, ERCC5, XRCC1, MDM2, ABCB1, MTHFR, SLC19A1, IL1B, IL6 and IL16 gene polymorphisms in toxicity to platinum-based chemotherapy in NSCLC patients.

2. Material and methods

A prospective cohorts study was conducted.

2.1. Study population

This study was performed at Complejo Hospitalario Universitario de Granada (CHUG), Granada, Spain. Between December 2012 and January 2016, 141 NSCLC patients ≥18 years diagnosed histologically or cytologically as NSCLC (stages I–IV) were enrolled in the study. The eligible patients were those with normal results of hematological function (hemoglobin > 9 g/dl, neutrophil count > 1500/mm^3, and platelet count > 100000/mm^3), liver function (bilirubin <1.5 times the normal upper limit, aspartate aminotransferase and alanine aminotransferase <2.5 times the normal upper limit), renal function (creatine clearance rate >50 ml/s) and measurable disease by chest computed tomography (CT) scan.

All patients were treated intravenously with cisplatin or carboplatin in combination with a third-generation drug (gemcitabine, paclitaxel, pemetrexed and vinorelbine) according to the National Comprehensive Cancer Network (NCCN) guidelines [5]. Hematology and biochemistry analyses were done at the end of each cycle. Based on NCCN version 4, 2016 guideline, patients with lymph node metastasis (N2–N3) and distant metastasis (M1) were not surgical candidates, although after neoadjuvant chemotherapy patients with potentially resectable N2 NSCLC were candidates for surgery [5]. Adjuvant chemotherapy was administered in patients with ECOG 0-1. Patients with unresectable stage IIIA NSCLC were candidates for chemoradiotherapy [5].

EGFR status was measured by cobas® EGFR Mutation Test. This study was approved by the CHUG Ethics and Research Committee and was performed conform the declaration of Helsinki. All patients signed an informed consent form for blood sample collection.

2.2. Sociodemographic and clinical variables

Sociodemographic and clinical data were collected by reviewing clinical records. Clinical and histopathological data collected were: gender, family history of cancer, previous non-lung cancer, previous lung disease, smoking status, age, histology, tumor stage, chemotherapy agents, surgery, concomitant or concurrent radiotherapy, EGFR status and response.

The staging system used to classified tumor was based on the guidelines of the AJCC [49]. Platinum-based chemotherapy response was evaluated based on the Response Evaluation Criteria in Solid Tumors (RECIST) guideline (Version 1.1) [50]. Patients classified as complete response plus partial response were catalogued as responders to treatment and stable disease plus progressive disease as non-responders.

Non-patient identifiers were applied to identify samples and they were preserved confidentially.

2.3. Genetic variables

2.3.1. DNA isolation

DNA was extracted using the QIAamp DNA Mini Kit (QiagenGmBH, Hilden, Germany) according to the manufacturer's instructions for DNA purification from blood and stored at −40 °C.

2.3.2. Detection of gene polymorphisms

ERCC1 C118T (rs11615), ERCC1 C8092A (rs3212986), ERCC2 Lys751Gln (rs13181), ERCC2 Asp312Asn (rs1799793), ERCC2 (rs50872) ERCC2 (rs238416), ERCC5 His46His (rs1047768), ERCC5 Asp1104His (rs17655), XRCC1 Arg194Trp (rs1799782), XRCC1 Gln399Arg (rs25487), MDM2 (rs1470383), MDM2 (rs1690924), ABCB1 C3435T (rs1045642), ABCB1 C1236T (rs1128503), MTHFR A1298C (rs1801131), MTHFR C677T (rs1801133), MTR (rs1805087), SLC19A1 Arg27His (rs1051266), IL1B (rs12621220), IL1B (rs1143623), IL1B (rs16944), IL1B (rs1143627), IL6 (rs1800795), IL16 (rs7170924) gene polymorphisms were analyzed by Real-Time PCR using TaqMan® probes. ABCB1 Ala893Ser/Thr (rs2032582) gene polymorphism was evaluated by sequencing. Genotyping methodology was previously described [51].

2.4. Toxicity variables

Toxicity was evaluated according to the Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0 [52]. Based on the severity of adverse events, it was classified into grade 0–2 and 3–4. General toxicity was defined as 3–4 when at least one adverse event was present in grade 3–4, and 0–2 otherwise. The occurrence of more than one and more than two adverse events was also analyzed.

The types of adverse events collected were: asthenia, gastrointestinal toxicity, hematological toxicity, infection, neurotoxicity and nephrotoxicity.

2.5. Statistical analysis

Quantitative data were expressed as the mean (±standard deviation) for normally-distributed variables or medians and percentiles (25 and 75) for non-normal distributed variables. The Shapiro-Wilks test was used to assess normality.

The bivariate association between toxicity and polymorphisms was assessed using the Pearson's chi-square or Fisher's exact test, and evaluated by relative risk (RR) and their corresponding 95% confidence intervals (CI).

Logistic regression model (backward stepwise method) was used to determine the influence of polymorphisms on toxicity.

All tests were two-sided with a significant level of $p < 0.05$. Data analysis was performed using R 3.0.1 [53].

Hardy Weinberg equilibrium and pairwise haplotype frequencies were estimated using the free, open-source whole genome association analysis toolset PLINK [54].

3. Results

3.1. Patients characteristics

The study comprised a total of 141 NSCLC Caucasian patients. The baseline characteristics are summarized in Table 1. Mean age was 61 [52,67] years, 104 were male (104/141; 73.76%) and 100 stage IIIB–IV (100/141; 70.92%). All patients received platinum-based chemotherapy in combination with gemcitabine (21/141; 14.89%), paclitaxel (33/141; 23.40%), pemetrexed (37/141; 26.24%) or vinorelbine (50/141; 35.46%). During follow-up, grade 3–4 toxicity profile presented was: 29.79% (42/141) general toxicity, 2.13% (3/141) asthenia, 2.13% (3/141) gastrointestinal toxicity, 24.11% (34/141) hematological toxicity, 3.55% (5/141) infection, 2.13% (3/141) nephrotoxicity and 0.71% (1/141) neurotoxicity.

3.2. Influence of clinico-pathologic characteristics on toxicity

General toxicity was associated with previous non-lung cancer ($p = 0.007653$; RR = 7.99; $CI_{95\%} = 1.73–36.8$; Table S1) and chemotherapy agents ($p = 0.0002537$; Table S1). Hematological toxicity was also associated with previous non-lung cancer ($p = 0.03109$; RR = 6.43; $CI_{95\%} = 1.18–34.91$; Table S2) and chemotherapy agents ($p = 0.002183$; Table S2). Nephrotoxicity was associated with histology ($p = 0.0486$; Table S3). Infection was associated with response to platinum-based chemotherapy ($p = 0.02655$; RR = 9.56; $CI_{95\%} = 1.07–85.21$; Table S4).

Asthenia, gastrointestinal toxicity, neurotoxicity and the occurrence of multiple adverse events were not associated with clinical or demographic characteristics (Tables S5–S9).

3.3. Genotypes distribution

All gene polymorphisms distributions were in agreement with those expected according to the Hardy-Weinberg equilibrium model. Linkage disequilibrium values D' and r^2 are shown in Table S10.

3.4. Influence of gene polymorphisms on toxicity

3.4.1. General toxicity

ERCC1 C118T, ERCC2 rs50872 and ABCB1 C1236T were associated with general toxicity. Patients with CT/TT genotypes for ERCC1 C118T ($p = 0.01864$; RR = 6.78; $CI_{95\%} = 1.38, 33.4$; Table S11), CC for ERCC2 rs50872 ($p = 0.02336$; RR = 2.03; $CI_{95\%} = 1.1, 3.74$; Table S11) or TT for ABCB1 C1236T ($p = 0.01483$; RR = 2.08; $CI_{95\%} = 1.15, 3.75$; Table S11) were in higher risk of grade 3–4 toxicity. Logistic regression analysis adjusted by chemotherapy agents and previous non-lung cancer revealed that ERCC1 C118T-T allele and ERCC2 rs50872-CC genotype were the only genetic independent factors associated with higher general toxicity ($p_{likelihoodratiotest} = 1.898 \cdot 10^{-7}$; Table 2).

ERCC2 Asp312Asn and IL1B polymorphisms were associated with multiple adverse events (>1). In particular, ERCC2 Asp312Asn ($p = 0.03778$; Table S12), IL1B rs12621220-CT/TT ($p = 0.01627$; RR = 1.35; $CI_{95\%} = 1.06, 1.72$; Table S12), IL1B rs1143623-CG/GG ($p = 0.01627$; RR = 1.35; $CI_{95\%} = 1.06, 1.72$; Table S12), IL1B rs16944 ($p = 0.02977$; Table S12) or IL1B rs1143627 ($p = 0.04501$; Table S12) were in risk of experiencing more than one adverse event. A logistic regression analysis revealed that the AG genotype for ERCC2 Asp312Asn and CT/TT for IL1B-rs12621220 were independently associated with the occurrence of more than one adverse event ($p_{likelihoodratiotest} = 0.0009103$; Table 3). Similarly, carriers of ABCB1 C1236T-TT genotype ($p = 0.009664$; RR = 2.32; $CI_{95\%} = 1.23, 4.39$; Table S13), ERCC2 Asp312Asn ($p = 0.04637$; Table S13) or XRCC1 Gln399Arg-AG/GG genotype ($p = 0.04589$; RR = 5.79; $CI_{95\%} = 1.03,$

Table 1
Clinico-pathologic characteristics of the 141 NSCLC patients treated with platinum based chemotherapy.

	n	%
Gender		
Female	37	25.24
Male	104	73.76
Family history of cancer		
YES	85	60.28
NO	56	39.72
Previous non-lung cancer		
YES	23	16.31
NO	118	83.69
Previous lung disease		
YES	34	24.11
NO	107	75.89
Smoking status		
Current-Smokers	72	51.06
Former-smokers	52	36.88
Non-smokers	17	12.06
Age at NSCLC diagnosis	61 [52,67]	
≤60	68	48.23
>60	73	51.77
Histology		
Adenocarcinoma	87	61.70
Squamous cell carcinoma	51	36.17
Unknown	3	2.13
Tumor stage		
I, II or IIIA	41	29.5
Neoadjuvant Chemotherapy	20	48.78
Adjuvant Chemotherapy	16	39.02
Chemoradiotherapy	5	12.20
IIIB or IV	98	70.5
Chemoradiotherapy	37	37.76
Palliative	61	62.24
Chemotherapy agents		
Gemcitabine	21	14.89
Paclitaxel	33	23.40
Pemetrexed	37	26.24
Vinorelbine	50	35.46
Surgery		
YES	33	23.4
NO	108	76.6
Radiotherapy		
YES	45	31.91
NO	96	68.09
EGFR status		
Wild-type	72	51.06
Mutated	16	11.35
Unknown	53	37.59
Response		
CR	38	27.34
PR	60	43.17
SD	8	5.76
PD	33	23.74
Response/No response (divided by stage)		
I, II or IIIA		
Neoadjuvant Chemotherapy	18/2	90.0/10.0
Adjuvant Chemotherapy	15/1	93.8/6.2
Chemoradiotherapy	3/2	60.0/40.0
IIIB or IV		
Chemoradiotherapy	30/7	81.1/18.9
Palliative	31/29	51.7/48.3
Toxicity		
Grade 0–2	99	70.21
Grade 3–4	42	29.79

Qualitative variables: number (percentage).
Quantitative variables.
Normal distribution: mean ± standard deviation.
Non-normal distribution: P_{50} [P_{25}, P_{75}].
CR: Complete Response; PR: Partial Response; SD: Stable Disease; PD: Progressive Disease.

Table 2
Multivariate regression analysis for general toxicity according to clinical characteristics and gene polymorphisms.

	General Toxicity	
	OR ($CI_{95\%}$)	p
Chemotherapy agents		
Gemcitabine (Reference)		
Paclitaxel	0.13 (0.02, 0.52)	0.00587
Pemetrexed	0.10 (0.02, 0.38)	0.00147
Vinorelbine	0.27 (0.07, 0.91)	0.04098
Previous non-lung cancer		
No	11.57 (2.09, 217.62)	0.02245
ERCC1 C118T-CT/TT	26.05 (4.33, 515.77)	0.00345
ERCC2 rs50872-CC	4.06 (1.66, 10.65)	0.00291

32.47; Table S13), showed higher risk of experiencing more than two adverse events. Logistic regression analysis revealed that the G-allele of ERCC2 Asp312Asn and the TT genotype of ABCB1 C1236T were the only independent factors associated with higher risk of more than two adverse events ($p_{likelihoodratiotest}$ = 0.0007531; Table 3).

3.4.2. Hematological toxicity

ERCC2 rs50872 and IL16 rs7170924 presented influence on hematological toxicity (Table S14). In fact, patients carrying the CC genotype for ERCC2 rs50872 (p = 0.02228; RR = 2.34; $CI_{95\%}$ = 1.13, 4.85; Table S17) or IL16 rs7170924 T-allele (p = 0.02772; RR = 2.06; $CI_{95\%}$ = 1.08, 3.92; Table S17) presented higher grade of hematological toxicity. A multivariate logistic regression analysis adjusted by chemotherapy agents was used to further investigate the impact of ERCC2 rs50872 and IL16 rs7170924 polymorphisms on hematological toxicity. Both remaining significantly associated to grade 3–4 hematological toxicity ($p_{likelihoodratiotest} = 6.861 \cdot 10^{-5}$; Table 4).

3.4.3. Other toxicity subtypes

The bivariate analysis showed that higher grade of asthenia was associated to IL16 rs7170924 (TT > GT > GG) polymorphism (p = 0.03769; Table S15). No association was found for other genetic or clinical variables. ERCC5 His46His and SLC19A1 Arg27His were associated with gastrointestinal toxicity. Particularly, patients carrying the CC genotype for ERCC5 His46His (p = 0.02699; Table S16) or AA for SLC19A1 Arg27His (p = 0.01431; Table S16) showed greater risk of gastrointestinal toxicity. No association was reached on multivariate logistic regression model. ERCC2 rs238416 was the only polymorphism that showed influence on grade 3–4 infection. Particularly, carriers of AA genotype showed greater risk of grade 3–4 infection to those carrying the G-allele (p = 0.01767; RR = 9.63; $CI_{95\%}$ = 1.56, 59.6; Table S17). No multivariate regression analysis was investigated. The bivariate analysis revealed that IL6 rs1800795-CC, MTR rs1805087-AG/GG and XRCC1 Gln399Arg-GG genotypes were associated with severe nephrotoxicity (p = 0.04286 for IL6 rs1800795, p = 0.0251 for MTR rs1805087 and p = 0.03836 for XRCC1 Gln399Arg) (Table S18). However, multivariate logistic regression analysis did not revealed influence of these polymorphisms on severe nephrotoxicity. The only polymorphism associated with grade 3–4 neurotoxicity was MDM2 rs1470383, particularly the CC genotype (p = 0.04965; Table S19). No multivariate regression analysis was furtherly investigated.

4. Discussion

Platinum-based doublet chemotherapy remains as the standard treatment for NSCLC patients with EGFR wild-type, and as alternative to failure to EGFR inhibitors in mutated EGFR patients [5]. This highly aggressive treatment is generally accompanied by

Table 3
Multivariate regression analysis for multiple adverse events according to gene polymorphisms.

	Number of adverse events	
	OR ($CI_{95\%}$)	p
	>1 adverse events	
ERCC2 Asp312Asn		
AA	0.23(0.07–0.71)	0.01140
(Reference) AG		
GG	0.31(0.13–0.73)	0.00891
IL1B rs12621220-CT/TT	3.37(1.55–7.69)	0.00280
	>2 adverse events	
ERCC2 Asp312Asn-AG/GG	5.92(1.52–40.18)	0.026040
ABCB1 C1236T-TT	4.35(1.67–11.76)	0.002842

Table 4
Multivariate regression analysis for hematological toxicity according to clinical characteristics and gene polymorphisms.

	Hematological Toxicity OR ($CI_{95\%}$)	p
Chemotherapy agents		
Gemcitabine (Reference)		
Paclitaxel	0.15 (0.03, 0.54)	0.00559
Pemetrexed	0.11 (0.02, 0.42)	0.00218
Vinorelbine	0.36 (0.11, 1.10)	0.07525
ERCC2 rs50872-CC	3.23 (1.29, 8.82)	0.01562
IL16 rs7170924- GT/TT	3.19 (1.36, 7.97)	0.01007

severe adverse events (asthenia, gastrointestinal toxicity, hematological toxicity, neurotoxicity and nephrotoxicity) [8–10], a toxicity profile which great inter-individual differences probably due to genetic factors [11–24]. Several SNPS in different genes involved in platinum pharmacodynamics, metabolism and mechanism of action have been proposed as potential causes of this variability. In this study, 141 NSCLC patients from a single institution treated with cisplatin or carboplatin in combination with gemcitabine, paclitaxel, pemetrexed or vinorelbine were recruited to investigate the potential role of ERCC1, ERCC2, ERCC5, XRCC1, MDM2, ABCB1, MTHFR, MTR, SLC19A1, IL1B, IL6 and IL16 gene polymorphisms in chemotherapy toxicity. In these patients, ERCC1 C118T-T allele and ERCC2 rs50872-CC genotype acted as predictors of occurrence of at least one case of grade 3–4 adverse events, along with gemcitabine as combination agent and non-history of previous non-lung cancer (Table 2). Despite the influence of ERCC1 gene polymorphisms on toxicity has been widely investigated in NSCLC, no association has been previously reported [11,24,56–68]. Only a study of 81 Caucasian patients diagnosed with late-stage solid tumors (55.6% lung cancer) has reported a role of ERCC1 gene polymorphisms as predictors of cisplatin-induced nephrotoxicity [69]. In particular, patients carrying the T-allele of ERCC1 rs11615 showed a 12.8% mean decrease in estimated glomerular filtration rates (p = 0.047) [69]. The effect of ERCC2 rs50872 polymorphism on platinum-based chemotherapy toxicity had not been previously investigated in NSCLC, although C-allele was associated to higher median overall survival (OS) and progression-free survival (PFS) (p = 0.009 and p = 0.032, respectively) in 129 unresectable Asian NSCLC patients [70]. In our patients, the occurrence of multiple adverse events was influenced by ERCC2 Asp312Asn G-alelle, ABCB1 C1236T-TT genotype and IL1B rs12621220C-allele (Table 3). Studies with large samples have not reported association between ERCC2 Asp312Asn and toxicity [58–60,64–67,71,72]. However, the G-allele for ERCC2 Asp312Asn was associated with lower frequency of severe hematological toxicity in 55 Polish stage IIIB-IV NSCLC patients (OR = 0.08; $CI_{95\%}$ = 0.01, 0.40; p = 0.0005; AG/GG vs AA) [11]. The effect of ABCB1 C1236T polymorphism on toxicity has not previously described, although the CC genotype has been associated with lower OS and PFS in previous studies [73,74]. The influence of IL1B-rs12621220 on platinum-based chemotherapy toxicity has not either been previously investigated, although the heterozygous variant has recently been associated with decreased risk of NSCLC in patients over age 63 from a study including 889 lung cancer and 1005 controls (OR = 0.71; $CI_{95\%}$ = 0.52-0.98; p = 0.04) [75].

Since platinum-based chemotherapy present a wide toxicity profile, we analyzed different subtypes of toxicity: asthenia, gastrointestinal toxicity, infection, hematological toxicity, nephrotoxicity and neurotoxicity. According to our results, patients carrying IL16 rs7170924 T-allele were in higher risk of hematological toxicity (OR = 3.19; $CI_{95\%}$ = 1.35, 7.97; Table 4). No studies have evaluated the impact of IL16 rs7170924 polymorphism on this toxicity subtype. However, a protective effect of the IL16 rs7170924 T-allele on PFS has been reported in 651 Caucasian stage I–IV NSCLC patients (HR = 0.65; $CI_{95\%}$ = 0.50, 0.83 for GT/TT vs GG) [17]. In our patients, hematological toxicity adjusted by chemotherapy agents was also associated with ERCC2 rs50872 polymorphism. Particularly, the CC genotype was associated with higher risk of hematological toxicity (OR = 3.23; $CI_{95\%}$ = 1.29, 8.82; Table 4). No studies have been published related to the effect of ERCC2 rs50872 polymorphism on hematological toxicity. In our patients, although some gene polymorphisms were associated to particular subtypes of toxicity in the bivariate analysis (ERCC2 rs238416, ERCC5 His46His, XRCC1 Gln399Arg, MDM2 rs1470383, MTR rs1805087, SLC19A1 Arg27His, IL6 rs1800795), the independency of those associations could not be analyzed, due to the insufficient number of patients who presented severe asthenia (3) gastrointestinal toxicity (3), infection (5), nephrotoxicity (3) and neurotoxicity (1). Other studies have attempted to find a role for MDM2 rs1470383, ERCC5 His46His, SLC19A1 Arg27His and XRCC1 Gln399Arg on toxicity to platinum-based chemotherapy, not always successfully. Whereas MDM2 rs1470383 has been proposed as a risk factor for hematological chemotherapy-related toxicity (OR = 4.10; $CI_{95\%}$ = 1.73, 9.71 for CC vs TT) [15], no significant association was found for ERCC5 His46His in 74 Spanish stage IIIA-IV NSCLC patients [64] or for SLC19A1 Arg27His in 94 Caucasian stage IIIB-IV NSCLC patients [20]. The role of XRCC1 Gln399Arg on toxicity to platinum-based chemotherapy in NSCLC patients has showed conflicting results. Although no effect on nephrotoxicity was found in 55 Caucasian stage IIIB-IV NSCLC patients [11,13,14], the GG genotype has been associated with other subtypes of toxicity, specifically, increased hematological toxicity (OR = 0.323; $CI_{95\%}$ = 0.121, 0.862 for AG/AA vs GG and OR = 0.22; $CI_{95\%}$ = 0.06, 0.82 for AG/AA vs GG) and gastrointestinal toxicity (OR = 0.298; $CI_{95\%}$ = 0.108, 0.825 for AG/AA vs GG) [11,13,14]. On the contrary, other study in 487 Chinese stage IIIA–IV NSCLC patients has shown a protective effect of GG genotype for hematological toxicity (OR = 2.135; $CI_{95\%}$ = 1.207, 3.777 for AG/GG vs GG) [14]. The influence of ERCC2 rs238416, MTR rs1805087 and IL6rs1800795 gene polymorphism on toxicity to chemotherapy in NSCLC patients has not been previously evaluated.

In our study, ERCC2 Lys751Gln polymorphism showed no influence on toxicity, in consonance with other studies developed in larger samples (up to 493 patients) [57–61,64–67,71,72,76,77]. However, small studies developed have reported an association with hematological toxicity and nephrotoxicity [11,24]. The A-allele for ERCC2 Lys751Gln polymorphism was associated with increased grade 2–3 neutropenia ($p = 0.04$) in 62 Spanish stage IIIB-IV NSCLC patients [24] and severe nephrotoxicity in 55 Polish late-stage NSCLC patients (OR = 0.07; $CI_{95\%}$ = 0.02, 0.31 for AC/CC vs AA) [11]. ERCC5 Asp1104His was not either associated with toxicity in our patients, despite a previous study with 388 Chinese NSCLC patients had proposed ERCC5 rs17655 as a risk factor for infection ($p = 0.017$) [12]. Similarly, no association was found between ABCB1 C3435T, ABCB1 Ala893Ser/Thr and XRCC1 Arg194Trp in our patients, which is in consonance with previous studies [11,56,64,66,73,78–83]. MDM2 rs1690924-GG genotype has been related to lower gastrointestinal toxicity in 663 Asian stage IIIA-IV NSCLC patients (OR = 2.32; $CI_{95\%}$ = 1.30, 4.14 for AG vs AA) [15]. However, MDM2 rs1690924 did not influence toxicity in our study. A similar outcome was observed for polymorphisms in MTHFR, which did not demonstrate an association with toxicity in our patients, despite a previous study in 1004 Chinese stage III–IV NSCLC patients reported that MTHFR C677T-CC and MTHFR A1298C-AA genotype were predictors of higher hematological toxicity (OR = 0.40; $CI_{95\%}$ = 0.19, 0.85 for CT vs CC and OR = 0.40; $CI_{95\%}$ = 0.22, 2.23 for AC vs AA, respectively) [23]. The effect of IL1B rs1143623, rs16944 and rs1143627 on platinum-based chemotherapy toxicity, which showed no significant association in the present study, has not been previously evaluated.

The patients enrolled for this study were recruited from one hospital, which ensures the homogeneity of the sample, regarding individuals, treatment administered and measure of the toxicity variables. The main limitation of this study is the sample size, which may have been responsible for the lack of association of some SNPs, particularly when investigating those toxicity subtypes which occurred in a very low number of patients. Despite of this, the prognostic value of polymorphisms in ERCC1, ERCC2, ABCB1, IL1B and IL16 genes to predict severe toxicity was evident.

These results suggested that ERCC1 C118T, ERCC2 Asp312Asn, ERCC2 rs50872, ABCB1 C1236T, IL1B rs12621220 and IL16 rs7170924 polymorphisms may substantially act as prognostic factors in NSCLC patients treated with platinum-based chemotherapy. These associations should be validated in a new, independent cohort of patients. Furthermore, the influence of ERCC2 rs238416, ERCC5 His46His, XRCC1 Gln399Arg, MDM2 rs1470383, MTR rs1805087, SLC19A1 Arg27His, IL6 rs1800795 gene polymorphisms on toxicity to chemotherapy in NSCLC patients should be elucidated in further studies with larger samples.

5. Conclusions

Our results suggest that NSCLC patients with ERCC1 C118T-T allele and ERCC2 rs50872-CC genotype have higher risk of general toxicity for platinum-based chemotherapy. The G-alelle of ERCC2 Asp312Asn, ABCB1 C1236T-TT and the IL1B rs12621220-CT/TT genotypes conferred a higher risk to present multiple adverse events. The subtype toxicity analysis also revealed ERCC2 rs50872-CC genotype and IL16 rs7170924-T allele were associated with grade 3–4 hematological toxicity. No influence of ERCC1 C8092A, ERCC2 Lys751Gln, ERCC5 Asp1104His, XRCC1 Arg194Trp, MDM2 rs1690924, ABCB1 C3435T, ABCB1 Ala893Ser/Thr, MTHFR A1298C, MTHFR C677T, IL1B rs1143623, IL1B rs16944, and IL1B rs1143627on platinum-based chemotherapy toxicity was found in our patients.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.phrs.2016.08.002.

References

[1] R.L. Siegel, K.D. Miller, A. Jemal, Cancer statistics, 2016, Cancer J. Clin. 66 (2016) 7—30.
[2] S. Edge, D.R. Byrd, C.C. Compton, A.G. Fritz, F.L. Greene, A. Trotti, AJCC Cancer Staging Manual, 7th ed., 2010.
[3] R.S. Herbst, J.V. Heymach, S.M. Lippman, Lung cancer, N. Engl. J. Med. 359 (2008) 1367–1380.
[4] J.R. Molina, P. Yang, S.D. Cassivi, S.E. Schild, A.A. Adjei, Non-small cell lung cancer: epidemiology, risk factors, treatment, and survivorship, Mayo Clin. Proc. 83 (2008) 584–594.
[5] D.S. Ettinger, D.E. Wood, W. Akerley, L.A. Bazhenova, H. Borghaei, D.R. Camidge, R.T. Cheney, L.R. Chirieac, T.A. D'Amico, T.J. Dilling, M.C. Dobelbower, R. Govindan, M. Hennon, L. Horn, T.M. Jahan, R. Komaki, R.P. Lackner, M. Lanuti, R. Lilenbaum, J. Lin, B.W. Loo Jr., R. Martins, G.A. Otterson, J.D. Patel, K.M. Pisters, K. Reckamp, G.J. Riely, S.E. Schild, T.A. Shapiro, N. Sharma, J. Stevenson, S.J. Swanson, K. Tauer, S.C. Yang, K. Gregory, M. Hughes, Nccn guidelines insights: non-small cell lung cancer, version 4.2016, J. Nat. Compr. Cancer Netw. 14 (2016) 255–264.
[6] S.Y. Brule, K. Al-Baimani, H. Jonker, T. Zhang, G. Nicholas, G. Goss, S.A. Laurie, P. Wheatley-Price, Palliative systemic therapy for advanced non-small cell lung cancer: investigating disparities between patients who are treated versus those who are not, Lung Cancer 97 (2016) 15–21.
[7] C. Zhong, H. Liu, L. Jiang, W. Zhang, F. Yao, Chemotherapy plus best supportive care versus best supportive care in patients with non-small cell lung cancer: a meta-analysis of randomized controlled trials, PLoS One 8 (2013) e58466.
[8] T.S. Mok, Y.-L. Wu, S. Thongprasert, C.H. Yang, D.T. Chu, N. Saijo, P. Sunpaweravong, B. Han, B. Margono, Y. Ichinose, Y. Nishiwaki, Y. Ohe, J.J. Yang, B. Chewaskulyong, H. Jiang, E.L. Duffield, C.L. Watkins, A.A. Armour, M. Fukuoka, Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma, N. Engl. J. Med. 361 (2009) 947–957.
[9] N.E. Madias, J.T. Harrington, Platinum nephrotoxicity, Am. J. Med. 65 (1978) 307–314.
[10] R.S. Goldstein, G.H. Mayor, Minireview: the nephrotoxicity of cisplatin, Life Sci. 32 (1983) 685–690.
[11] T. Powrozek, R. Mlak, P. Krawczyk, I. Homa, M. Ciesielka, P. Koziol, M. Prendecka, J. Milanowski, T. Malecka-Massalska, The relationship between polymorphisms of genes regulating DNA repair or cell division and the toxicity of platinum and vinorelbine chemotherapy in advanced nsclc patients, Clin. Transl. Oncol. 18 (2016) 125–131.
[12] L. Zhang, G. Gao, X. Li, S. Ren, A. Li, J. Xu, J. Zhang, C. Zhou, Association between single nucleotide polymorphisms (snps) and toxicity of advanced non-small-cell lung cancer patients treated with chemotherapy, PLoS One 7 (2012) e48350.
[13] J.H. Deng, J. Deng, D.H. Shi, X.N. Ouyang, P.G. Niu, Clinical outcome of cisplatin-based chemotherapy is associated with the polymorphisms of gstp1 and xrcc1 in advanced non-small cell lung cancer patients, Clin. Transl. Oncol. 17 (2015) 720–726.
[14] Y. Peng, Z. Li, S. Zhang, Y. Xiong, Y. Cun, C. Qian, M. Li, T. Ren, L. Xia, Y. Cheng, D. Wang, Association of DNA base excision repair genes (ogg1, ape1 and xrcc1) polymorphisms with outcome to platinum-based chemotherapy in advanced nonsmall-cell lung cancer patients, Int. J. Cancer 135 (2014) 2687–2696.
[15] J. Qian, H. Liu, S. Gu, Q. Wu, X. Zhao, W. Wu, H. Wang, J. Wang, H. Chen, W. Zhang, Q. Wei, L. Jin, D. Lu, Genetic variants of the mdm2 gene are predictive of treatment-related toxicities and overall survival in patients with advanced nsclc, Clin. Lung Cancer 16 (2015) e37–53.
[16] M. Gomes, A. Coelho, A. Araujo, A. Azevedo, A.L. Teixeira, R. Catarino, R. Medeiros, Il-6 polymorphism in non-small cell lung cancer: a prognostic value? Tumour Biol. 36 (2015) 3679–3684.

[17] N.T. Woods, A.N. Monteiro, Z.J. Thompson, E.K. Amankwah, N. Naas, E.B. Haura, A.A. Beg, M.B. Schabath, Interleukin polymorphisms associated with overall survival, disease-free survival, and recurrence in non-small cell lung cancer patients, Mol. Carcinog. 54 (2015) e172–e184.
[18] W.J. Li, H. Jiang, X.J. Fang, H.L. Ye, M.H. Liu, Y.W. Liu, Q. Chen, L. Zhang, J.Y. Zhang, C.L. Yuan, Q.Y. Zhang, Polymorphisms in thymidylate synthase and reduced folate carrier (SLC19A1) genes predict survival outcome in advanced non-small cell lung cancer patients treated with pemetrexed-based chemotherapy, Oncol. Lett. 5 (2013) 1165–1170.
[19] M. Tiseo, E. Giovannetti, C. Tibaldi, A. Camerini, F. Di Costanzo, F. Barbieri, J.A. Burgers, A. Vincent, G.J. Peters, E.F. Smit, A. Ardizzoni, Pharmacogenetic study of patients with advanced non-small cell lung cancer (nsclc) treated with second-line pemetrexed or pemetrexed-carboplatin, Lung Cancer 78 (2012) 92–99.
[20] A. Corrigan, J.L. Walker, S. Wickramasinghe, M.A. Hernandez, S.J. Newhouse, A.A. Folarin, C.M. Lewis, J.D. Sanderson, J. Spicer, A.M. Marinaki, Pharmacogenetics of pemetrexed combination therapy in lung cancer: pathway analysis reveals novel toxicity associations, Pharmacogen. J. 14 (2014) 411–417.
[21] A.A. Adjei, O.E. Salavaggione, S.J. Mandrekar, G.K. Dy, K.L. Ziegler, C. Endo, J.R. Molina, S.E. Schild, A.A. Adjei, Correlation between polymorphisms of the reduced folate carrier gene (slc19a1) and survival after pemetrexed-based therapy in non-small cell lung cancer: a north central cancer treatment group-based exploratory study, J. Thorac. Oncol. 5 (2010) 1346–1353.
[22] E.F. Smit, S.A. Burgers, B. Biesma, H.J. Smit, P. Eppinga, A.M. Dingemans, M. Joerger, J.H. Schellens, A. Vincent, N. van Zandwijk, H.J. Groen, Randomized phase ii and pharmacogenetic study of pemetrexed compared with pemetrexed plus carboplatin in pretreated patients with advanced non-small-cell lung cancer, J. Clin. Oncol. 27 (2009) 2038–2045.
[23] X. Li, M. Shao, S. Wang, X. Zhao, H. Chen, J. Qian, X. Song, J. Wang, L. Jin, J. Wu, Q. Li, C. Bai, B. Han, Z. Gao, D. Lu, Heterozygote advantage of methylenetetrahydrofolate reductase polymorphisms on clinical outcomes in advanced non-small cell lung cancer (nsclc) patients treated with platinum-based chemotherapy, Tumour Biol. 35 (2014) 11159–11170.
[24] D. Isla, C. Sarries, R. Rosell, G. Alonso, M. Domine, M. Taron, G. Lopez-Vivanco, C. Camps, M. Botia, L. Nunez, M. Sanchez-Ronco, J.J. Sanchez, M. Lopez-Brea, I. Barneto, A. Paredes, B. Medina, A. Artal, P. Lianes, Single nucleotide polymorphisms and outcome in docetaxel-cisplatin-treated advanced non-small-cell lung cancer, Ann. Oncol. 15 (2004) 1194–1203.
[25] L.H. Cui, Z. Yu, T.T. Zhang, M.H. Shin, H.N. Kim, J.S. Choi, Influence of polymorphisms in mthfr 677 c→t, tyms 3r→2r and mtr 2756 a→g on nsclc risk and response to platinum-based chemotherapy in advanced nsclc, Pharmacogenomics 12 (2011) 797–808.
[26] A. Matakidou, R. El Galta, M.F. Rudd, E.L. Webb, H. Bridle, T. Eisen, R.S. Houlston, Prognostic significance of folate metabolism polymorphisms for lung cancer, Br. J. Cancer 97 (2007) 247–252.
[27] M. Kartalou, J.M. Essigmann, Recognition of cisplatin adducts by cellular proteins, Mutat. Res. 478 (2001) 1–21.
[28] P. Jordan, M. Carmo-Fonseca, Molecular mechanisms involved in cisplatin cytotoxicity, Cell Mol. Life Sci. 57 (2000) 1229–1235.
[29] A. Sancar, DNA repair in humans, Annu. Rev. Genet. 29 (1995) 69–105.
[30] R. Garcia-Campelo, G. Alonso-Curbera, L.M. Anton Aparicio, R. Rosell, Pharmacogenomics in lung cancer: an analysis of DNA repair gene expression in patients treated with platinum-based chemotherapy, Expert Opin. Pharmacother. 6 (2005) 2015–2026.
[31] S. Jin, A.J. Levine, The p53 functional circuit, J. Cell Sci. 114 (2001) 4139–4140.
[32] D. Michael, M. Oren, The p53–mdm2 module and the ubiquitin system, Semin. Cancer Biol. 13 (2003) 49–58.
[33] T. Sakaeda, T. Nakamura, K. Okumura, Mdr1 genotype-related pharmacokinetics and pharmacodynamics, Biol. Pharm. Bull. 25 (2002) 1391–1400.
[34] F. Thiebaut, T. Tsuruo, H. Hamada, M.M. Gottesman, I. Pastan, M.C. Willingham, Cellular localization of the multidrug-resistance gene product p-glycoprotein in normal human tissues, Proc. Natl. Acad. Sci. U. S. A. 84 (1987) 7735–7738.
[35] K. Takara, T. Sakaeda, K. Okumura, An update on overcoming mdr1-mediated multidrug resistance in cancer chemotherapy, Curr. Pharm. Des. 12 (2006) 273–286.
[36] S. Hoffmeyer, O. Burk, O. von Richter, H.P. Arnold, J. Brockmoller, A. Johne, I. Cascorbi, T. Gerloff, I. Roots, M. Eichelbaum, U. Brinkmann, Functional polymorphisms of the human multidrug-resistance gene: multiple sequence variations and correlation of one allele with p-glycoprotein expression and activity in vivo, Proc. Natl. Acad. Sci. U. S. A. 97 (2000) 3473–3478.
[37] P. Frosst, H.J. Blom, R. Milos, P. Goyette, C.A. Sheppard, R.G. Matthews, G.J. Boers, M. den Heijer, L.A. Kluijtmans, L.P. van den Heuvel, et al., A candidate genetic risk factor for vascular disease: a common mutation in methylenetetrahydrofolate reductase, Nat. Genet. 10 (1995) 111–113.
[38] P.F. Jacques, A.G. Bostom, R.R. Williams, R.C. Ellison, J.H. Eckfeldt, I.H. Rosenberg, J. Selhub, R. Rozen, Relation between folate status, a common mutation in methylenetetrahydrofolate reductase, and plasma homocysteine concentrations, Circulation 93 (1996) 7–9.
[39] I. Weisberg, P. Tran, B. Christensen, S. Sibani, R. Rozen, A second genetic polymorphism in methylenetetrahydrofolate reductase (mthfr) associated with decreased enzyme activity, Mol. Genet. Metab. 64 (1998) 169–172.
[40] D. Leclerc, E. Campeau, P. Goyette, C.E. Adjalla, B. Christensen, M. Ross, P. Eydoux, D.S. Rosenblatt, R. Rozen, R.A. Gravel, Human methionine synthase: cdna cloning and identification of mutations in patients of the cblg complementation group of folate/cobalamin disorders, Hum. Mol. Genet. 5 (1996) 1867–1874.
[41] L. Sharp, J. Little, Polymorphisms in genes involved in folate metabolism and colorectal neoplasia: a huge review, Am. J. Epidemiol. 159 (2004) 423–443.
[42] K.S. Crider, T.P. Yang, R.J. Berry, L.B. Bailey, Folate and DNA methylation: a review of molecular mechanisms and the evidence for folate's role, Adv. Nutr. (Bethesda, Md.) 3 (2012) 21–38.
[43] C.G. Azzoli, S. Baker, S. Temin, W. Pao, T. Aliff, J. Brahmer, D.H. Johnson, J.L. Laskin, G. Masters, D. Milton, American society of clinical oncology clinical practice guideline update on chemotherapy for stage iv non–small-cell lung cancer, J. Clin. Oncol. 27 (2009) 6251—6266.
[44] L.M. Coussens, Z. Werb, Inflammation and cancer, Nature 420 (2002) 860–867.
[45] W.W. Lin, M. Karin, A cytokine-mediated link between innate immunity, inflammation, and cancer, J. Clin. Invest. 117 (2007) 1175–1183.
[46] B.B. Aggarwal, R.V. Vijayalekshmi, B. Sung, Targeting inflammatory pathways for prevention and therapy of cancer: short-term friend, long-term foe, Clin. Cancer Res. 15 (2009) 425–430.
[47] G. Dranoff, Cytokines in cancer pathogenesis and cancer therapy, Nat. Rev. Cancer 4 (2004) 11–22.
[48] J.W. Pollard, Tumour-educated macrophages promote tumour progression and metastasis, Nat. Rev. Cancer 4 (2004) 71–78.
[49] S.B.B.R. Edge, C.C. Compton, A.G. Fritz, F.L. Green, A. Trotti (Eds.), AJCC Cancer Staging Manual, 7 ed., Springer-Verlag, New York, 2010, XV–648.
[50] E.A. Eisenhauer, P. Therasse, J. Bogaerts, L.H. Schwartz, D. Sargent, R. Ford, J. Dancey, S. Arbuck, S. Gwyther, M. Mooney, L. Rubinstein, L. Shankar, L. Dodd, R. Kaplan, D. Lacombe, J. Verweij, New response evaluation criteria in solid tumours: revised recist guideline (version 1.1), Eur. J. Cancer 45 (2009) 228—247.
[51] E. Jimenez-Varo, M. Canadas-Garre, C.I. Henriques, A.M. Pinheiro, M.J. Gutierrez-Pimentel, M.A. Calleja-Hernandez, Pharmacogenetics role in the safety of acenocoumarol therapy, Thromb. Haemost. 112 (2014) 522–536.
[52] Health UDo, Services H. Common Terminology Criteria for Adverse Events (ctcae) Version 4.0, National Institutes of Health, National Cancer Institute, 2009, pp. 4.
[53] **Team rc. R: A language and environment for statistical computing. Available from: http://www.r-project.org/, 2013 [internet Oct. 2013].**
[54] S. Purcell, B. Neale, K. Todd-Brown, L. Thomas, M.A.R. Ferreira, D. Bender, J. Maller, P. Sklar, P.I.W. de Bakker, M.J. Daly, P.C. Sham, Plink: a toolset for whole-genome association and population-based linkage analysis, Am. J. Hum. Genet. (2007) 81.
[56] S. Chen, X. Huo, Y. Lin, H. Ban, Y. Lin, W. Li, B. Zhang, W.W. Au, X. Xu, Association of mdr1 and ercc1 polymorphisms with response and toxicity to cisplatin-based chemotherapy in non-small-cell lung cancer patients, Int. J. Hyg. Environ. Health 213 (2010) 140–145.
[57] D.R. Gandara, T. Kawaguchi, J. Crowley, J. Moon, K. Furuse, M. Kawahara, S. Teramukai, Y. Ohe, K. Kubota, S.K. Williamson, O. Gautschi, H.J. Lenz, H.L. McLeod, P.N. Lara Jr., C.A. Coltman Jr., M. Fukuoka, N. Saijo, M. Fukushima, P.C. Mack, Japanese-us common-arm analysis of paclitaxel plus carboplatin in advanced non-small-cell lung cancer: a model for assessing population-related pharmacogenomics, J. Clin. Oncol. 27 (2009) 3540–3546.
[58] M. Joerger, S.A. Burgers, P. Baas, E.F. Smit, T.J. Haitjema, M.P. Bard, V.D. Doodeman, P.H. Smits, A. Vincent, A.D. Huitema, J.H. Beijnen, J.H. Schellens, Germline polymorphisms in patients with advanced nonsmall cell lung cancer receiving first-line platinum-gemcitabine chemotherapy: a prospective clinical study, Cancer 118 (2012) 2466–2475.
[59] A. Kalikaki, M. Kanaki, H. Vassalou, J. Souglakos, A. Voutsina, V. Georgoulias, D. Mavroudis, DNA repair gene polymorphisms predict favorable clinical outcome in advanced non-small-cell lung cancer, Clin. Lung Cancer 10 (2009) 118–123.
[60] A. Kalikaki, A. Voutsina, A. Koutsopoulos, C. Papadaki, M. Sfakianaki, E. Yachnakis, A. Xyrafas, A. Kotsakis, S. Agelaki, J. Souglakos, D. Mavroudis, V. Georgoulias, Ercc1 snps as potential predictive biomarkers in non-small cell lung cancer patients treated with platinum-based chemotherapy, Cancer Invest. 33 (2015) 107–113.
[61] V. Ludovini, I. Floriani, L. Pistola, V. Minotti, M. Meacci, R. Chiari, D. Garavaglia, F.R. Tofanetti, A. Flacco, A. Siggillino, E. Baldelli, M. Tonato, L. Crino, Association of cytidine deaminase and xeroderma pigmentosum group d polymorphisms with response, toxicity, and survival in cisplatin/gemcitabine-treated advanced non-small cell lung cancer patients, J. Thorac. Oncol. 6 (2011) 2018–2026.
[62] G. Metro, R. Chiari, M. Mare, D. Giannarelli, F.R. Tofanetti, V. Minotti, M. Ferraldeschi, D. Giuffrida, L. Marcomigni, C. Bennati, M.J. Fischer, M. Meacci, R. Bellavita, L. Pistola, V. Ludovini, L. Crino, Carboplatin plus pemetrexed for platinum-pretreated, advanced non-small cell lung cancer: a retrospective study with pharmacogenetic evaluation, Cancer Chemother. Pharmacol. 68 (2011) 1405–1412.
[63] R. Suk, S. Gurubhagavatula, S. Park, W. Zhou, L. Su, T.J. Lynch, J.C. Wain, D. Neuberg, G. Liu, D.C. Christiani, Polymorphisms in ercc1 and grade 3 or 4 toxicity in non-small cell lung cancer patients, Clin. Cancer Res. 11 (2005) 1534–1538.
[64] I. Sullivan, J. Salazar, M. Majem, C. Pallares, E. Del Rio, D. Paez, M. Baiget, A. Barnadas, Pharmacogenetics of the DNA repair pathways in advanced non-small cell lung cancer patients treated with platinum-based chemotherapy, Cancer Lett. 353 (2014) 160–166.

[65] C. Tibaldi, E. Giovannetti, E. Vasile, V. Mey, A.C. Laan, S. Nannizzi, R. Di Marsico, A. Antonuzzo, C. Orlandini, S. Ricciardi, M. Del Tacca, G.J. Peters, A. Falcone, R. Danesi, Correlation of cda, ercc1, and xpd polymorphisms with response and survival in gemcitabine/cisplatin-treated advanced non-small cell lung cancer patients, Clin. Cancer Res. 14 (2008) 1797–1803.
[66] N. Vinolas, M. Provencio, N. Reguart, F. Cardenal, V. Alberola, J.M. Sanchez-Torres, F.J. Baron, M. Cobo, I. Maestu, I. Moreno, C. Mesia, A. Izquierdo, E. Felip, M. Lopez-Brea, A. Marquez, M. Sanchez-Ronco, M. Taron, M.C. Santarpia, R. Rosell, Single nucleotide polymorphisms in mdr1 gen correlates with outcome in advanced non-small-cell lung cancer patients treated with cisplatin plus vinorelbine, Lung Cancer 71 (2011) 191–198.
[67] V. Iranzo, R. Sirera, R.M. Bremnes, A. Blasco, E. Jantus-Lewintre, M. Taron, A. Berrocal, S. Blasco, C. Caballero, N. Del Pozo, R. Rosell, C. Camps, Chemotherapy-induced neutropenia does not correlate with DNA repair gene polymorphisms and treatment efficacy in advanced non-small-cell lung cancer patients, Clin. Lung Cancer 12 (2011) 224–230.
[68] V. KimCurran, C. Zhou, G. Schmid-Bindert, R. Shengxiang, S. Zhou, L. Zhang, J. Zhang, Lack of correlation between ercc1 (c8092a) single nucleotide polymorphism and efficacy/toxicity of platinum based chemotherapy in chinese patients with advanced non-small cell lung cancer, Adv. Med. Sci. 56 (2011) 30–38.
[69] M.V. Tzvetkov, G. Behrens, V.P. O'Brien, K. Hohloch, J. Brockmöller, P. Benöhr, Pharmacogenetic analyses of cisplatin-induced nephrotoxicity indicate a renoprotective effect of ercc1 polymorphisms, Pharmacogenomics 12 (2011) 1417—1427.
[70] S.H. Kim, G.W. Lee, M.J. Lee, Y.J. Cho, Y.Y. Jeong, H.C. Kim, J.D. Lee, Y.S. Hwang, I.S. Kim, S. Lee, S.Y. Oh, Clinical significance of ercc2 haplotype-tagging single nucleotide polymorphisms in patients with unresectable non-small cell lung cancer treated with first-line platinum-based chemotherapy, Lung Cancer 77 (2012) 578–584.
[71] M. Provencio, C. Camps, M. Cobo, R. De las Penas, B. Massuti, R. Blanco, V. Alberola, U. Jimenez, J.R. Delgado, F. Cardenal, M. Taron, J.L. Ramirez, A. Sanchez, R. Rosell, Prospective assessment of xrcc3, xpd and aurora kinase a single-nucleotide polymorphisms in advanced lung cancer, Cancer Chemother. Pharmacol. 70 (2012) 883–890.
[72] W. Wu, H. Li, H. Wang, X. Zhao, Z. Gao, R. Qiao, W. Zhang, J. Qian, J. Wang, H. Chen, Q. Wei, B. Han, D. Lu, Effect of polymorphisms in xpd on clinical outcomes of platinum-based chemotherapy for chinese non-small cell lung cancer patients, PLoS One 7 (2012) e33200.
[73] J.K. Lamba, B.L. Fridley, T.M. Ghosh, Q. Yu, G. Mehta, P. Gupta, Genetic variation in platinating agent and taxane pathway genes as predictors of outcome and toxicity in advanced non-small-cell lung cancer, Pharmacogenomics 15 (2014) 1565–1574.
[74] J.L. Weissfeld, B. Diergaarde, T. Nukui, S. Buch, A. Pennathur, M.A. Socinski, J.M. Siegfried, M. Romkes, Inherited variation in the atp-binding cassette transporter abcb1 and survival after chemotherapy for stage iii–iv lung cancer, J. Thorac. Oncol. 9 (2014) 1264–1271.
[75] Y. Li, W. Zhao, Z. Zhao, J. Wu, L. Chen, Y. Ma, Q. Li, D. Lu, L. Jin, J. Wang, Il1b gene polymorphisms, age and the risk of non-small cell lung cancer in a chinese population, Lung Cancer 89 (2015) 232–237.
[76] H. Cheng, Q. Qin, X. Sun, F. Li, N. Sun, L. Cheng, Z. Lu, B. Chen, Predictive effect of xpa and xpd polymorphisms on survival of advanced nsclc patients treated with platinum-based chemotherapy: a three-dimensional (3-d), polyacrylamide gel-based DNA microarray method, Technol. Cancer Res. Treat. 12 (2013) 473–482.
[77] Y. Li, X.E. Huang, G.F. Jin, H.B. Shen, L. Xu, Lack of any relationship between chemotherapy toxicity in non-small cell lung cancer cases and polymorphisms in xrcc1 codon 399 or xpd codon 751, Asian Pacific J. Cancer Prev. 12 (2011) 739–742.
[78] H.G. Ke, J. Li, Y. Shen, Q.S. You, Y. Yan, H.X. Dong, J.H. Liu, Z.Y. Shen, Prognostic significance of gstp1, xrcc1 and xrcc3 polymorphisms in non-small cell lung cancer patients, Asian Pacific J. Cancer Prev. 13 (2012) 4413–4416.
[79] L. Zhang, W. Ma, Y. Li, J. Wu, G.Y. Shi, Pharmacogenetics of DNA repair gene polymorphisms in non-small-cell lung carcinoma patients on platinum-based chemotherapy, Genet. Mol. Res. 13 (2014) 228–236.
[80] W. Zhao, L. Hu, J. Xu, H. Shen, Z. Hu, H. Ma, Y. Shu, Y. Shao, Y. Yin, Polymorphisms in the base excision repair pathway modulate prognosis of platinum-based chemotherapy in advanced non-small cell lung cancer, Cancer Chemother. Pharmacol. 71 (2013) 1287–1295.
[81] B. Han, Z. Guo, Y. Ma, S. Kang, Y. Wang, Q. Wei, X. Wu, Association of gstp1 and xrcc1 gene polymorphisms with clinical outcome of advanced non-small cell lung cancer patients with cisplatin-based chemotherapy, Int. J. Clin. Exp. Pathol. 8 (2015) 4113–4119.
[82] J.Y. Liu, Q.M. Liu, L.R. Li, Association of gstp1 and xrcc1 gene polymorphisms with clinical outcomes of patients with advanced non-small cell lung cancer, Genet. Mol. Res. 14 (2015) 10331–10337.
[83] D. Liu, J. Wu, G.Y. Shi, H.F. Zhou, Y. Yu, Role of xrcc1 and ercc5 polymorphisms on clinical outcomes in advanced non-small cell lung cancer, Genet. Mol. Res. 13 (2014) 3100–3107.