Clinical Study Protocol	
Study Intervention	No Drug
Study Code	<<D5161R00037>>
Version	2.0
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Gene Profile in *EGFR*m Locally Advanced or Metastatic NSCLC patients post Osimertinib 1L treatment failure: A real-world, multi-center Study (GPS)

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Regulatory Agency Identifier Number(s) Not Applicable

This CSP has been subject to a peer review according to AstraZeneca Standard procedures. The CSP is publicly registered and the results are disclosed and/or published according to the AstraZeneca Global Policy on Bioethics and in compliance with prevailing laws and regulations.

Protocol Number: D5161R00037

Amendment Number: 1
Study Intervention: No Drug

Study Phase: Phase 4

Short Title: Gene profile post Osimertinib 1L failure

Acronym: GPS

Medical Monitor Name and Contact Information will be provided separately

Principal investigator: PPD

PROTOCOL AMENDMENT SUMMARY OF CHANGES TABLE

DOCUMENT HISTORY	
Document	**Date of Issue**
Original Protocol	[25-Feb-2021]
Protocol 2.0	[20-Jan-2023]

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1 PROTOCOL SUMMARY

1.1 Synopsis

Protocol Title: Gene profile in *EGFR*m locally advanced or metastatic NSCLC patients post Osimertinib 1L treatment failure: A real-world, multi-center study (GPS)

Short Title: Gene profile post Osimertinib 1L failure

Rationale:

Osimertinib is a potent, oral, irreversible tyrosine kinase inhibitor (TKI) of *EGFR* mutation (*EGFR*m)-positive and T790M mutation-positive forms of *EGFR* with demonstrated clinical activity on central nervous system (CNS) metastases. Osimertinib is designed to have limited activity against wild-type (WT) *EGFR*. Osimertinib is approved globally, and marketed as Tagrisso®, for the first-line treatment of patients with locally advanced or metastatic NSCLC whose tumours have *EGFR* exon 19 deletions (ex19del) or exon 21 (L858R) substitution mutations and the treatment of patients with locally advanced or metastatic *EGFR* T790M mutation-positive NSCLC whose disease has progressed on or after *EGFR* TKI therapy. It has been approved as first-line treatment for advanced *EGFR* mutated NSCLC in China, and has also been widely used in clinical practice no matter in the first-line or in the later-line.

While similar to 1/2 G *EGFR*-TKI, acquired resistance to Osimertinib invariably often develops with a median PFS of 19 months[1]. The characterization of the landscape of resistance enables the development of subsequent therapies, but mechanisms of resistance to Osimertinib is primarily derived from patients who received Osimertinib after other *EGFR*-TKIs, such as *EGFR* C797, *EGFR* G724, *EGFR* L792, *MET* amplification, and so on[2-5]. Prior reports of acquired resistance mechanisms to Osimertinib primarily focus on patients who received Osimertinib as the second-line treatment. However, the mechanisms of resistance to Osimertinib in the first-line treatment are far from being clearly explored. As Osimertinib in the first-line treatment is applied more and more widely in advanced *EGFR*m+ NSCLC patients, we urgently need to know the resistance mechanisms to guide subsequent treatment strategy. Until now, the largest study to explore acquired resistance mechanism to 1L Osimertinib was FLAURA study, which detected 91 paired plasma samples by next generation sequencing (NGS) . The most common acquired resistance mechanisms were *MET* amplification (15%) and *EGFR* C797X mutation (7%)[6]. The result was quite different from the study conducted by Adam J. Schoenfeld et al using tissue (N=27), with squamous transformation accounting for 15% and *MET* amplification 7%[7]. The reason for different results maybe because these two studies used different samples to analyse resistance mechanisms, and the above two detection methods may have different sensitivity and specificity. In addition, Schoenfeld et al study was limited to patients at one site, Memorial Sloan Kettering Cancer Center in NY, USA.

Although some small sample studies have reported the possible resistance mechanisms of Osimertinib in the first-line treatment, it is still an urgent need to explore the whole gene profile

in *EGFR*m advanced NSCLC patients post Osimertinib 1L treatment by paired tissue and plasma to guide subsequent treatment strategy[6, 7]. Thus, the gene profile post Osimertinib 1L treatment in tissue and plasma may help to guide the following treatment.

MET overexpression is key resistance mechanism after 1L Osimertinib failure[8]. SAVANNAH observed improved outcomes in post 1L Osimertinib resistant EGFRm NSCLC patients with MET IHC90+(≥90% tumour cells MET IHC3+) and/or FISH10+(MET gene copy number ≥10); the estimated prevalence of IHC90+ is 29% in SAVANNAH study published on 2022 WCLC [9]. MET amplification is also a key resistance mechanism after 1L Osimertinib failure[8].FISH is golden standard testing method for MET amplification. The prevalence of FISH 5+(MET gene copy number ≥5 and/or MET/CEP7≥2) in INSIGHT 2 study [10] is 33% and the prevalence of FISH10+ in SAVANNAH study is 20% [9]. The prevalence data of MET amplification by FISH testing and MET overexpression by IHC testing in Chinese advanced EGFRm NSCLC patients who had disease progression following 1L osimertinib are very limited and still need further explore.

Tissue was unavailable for some advanced EGFRm NSCLC patients who had disease progression following 1L Osimertinib and blood samples show high potential as an alternative sample [11]. ddPCR is as an alternative technique with high sensitivity and specificity for MET amplification testing and it has been explored in previous study[12]. Besides, ddPCR blood assay for MET amplification testing still need be further validated through concordance of MET amplification between FISH testing in tissue and ddPCR testing in plasma.

Objectives and Endpoints

Objectives	Endpoints
Primary	
To describe the gene profile in *EGFR*m advanced NSCLC patients post Osimertinib 1L treatment.	● Gene profile in tissue Gene profile defined as gene alteration detected by NGS and frequency of every gene alteration. All gene alterations include gene mutation, copy number variation(CNV), fusion, etc. The frequency of all gene alteration detected by NGS(%) = (number of patients with every gene alteration detected by NGS)/(total number of patients in the FAS)×100%.
Secondary	

Objectives	Endpoints
To assess the concordance of plasma and tissue	● Gene profile in plasma Gene profile defined as gene alternation detected by NGS and frequency of every gene alternation. ● Concordance of plasma and tissue Using selected representative genes (for example *EGFR*, *MET*, *PIK3CA* and etc.) to measure the concordance Concordance of Gene X in plasma and tissue is defined as sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) between plasma and tissue results., Tissue sample is the reference standard. - sensitivity=(number of patients with positive result in both plasma and tissue)/(total number of patients with positive result in tissue samples)×100%; - specificity=(number of patients with negative result in both plasma and tissue)/(total number of patients with negative result in tissue samples)×100%. - PPV (%)=(number of patients with positive result in both plasma and tissue)/(total number of patients with positive result in plasma samples)×100%; - NPV (%)=(number of patients with negative result in both plasma and tissue)/(total number of patients with negative result in plasma samples)×100%; ● The percentage of pathology transformation Pathology transformation is defined as those transformation from non-small-cell lung cancer to small-cell lung cancer or from adenocarcinoma to squamous carcinoma, can be observed by IHC
CCI	

Objectives	Endpoints
CCI	CCI

Overall Design

Disclosure Statement: This is a real-world, multi-centre prospective study elucidating the gene profile in *EGFR*m locally advanced or metastatic NSCLC patients post Osimertinib 1L treatment.

Participant Population:

The target population of interest in this study is participants with locally advanced or metastatic

NSCLC, harboring common *EGFR*m of ex19del/L858R and having evidence of disease progression following 1L Osimertinib.

Number of Participants:

Approximately 200 participants in *EGFR*m locally advanced or metastatic NSCLC with the evidence of disease progression following 1L Osimertinib will be enrolled and 15 sites will be involved to elucidate the gene profile in paired tissue and plasma so that approximately 180 participants whom would be get gene profile, assuming a 10% drop-out.

Note: "Enrolled" means a participant's, or their legally acceptable representative's, agreement to participate in a clinical study following completion of the informed consent process. Potential participants who are screened for the purpose of determining eligibility for the study, but are not randomly assigned/assigned in the study, are considered "screen failures", unless otherwise specified by the protocol.

Human Biological Sample Analysis:

Participants will be required to provide paired tissue and whole blood after disease progression following 1L Osimertinib. 200 tissue samples and 200 whole blood samples will be used to detect gene alteration by NGS, respectively. 200 tissue samples will be used to detect pathological transformation by IHC. Approximately 80-100 tissue samples will be used to test MET overexpression by MET IHC and MET amplification by FISH respectively. Approximately 80-100 whole blood samples will be used to test MET amplification by ddPCR.

Statistical methods

General Aspects

A Statistical Analysis Plan (SAP) will be prepared and finalised prior to database lock. All statistical analyses will be completed using SAS version 9.4 or later. Descriptive statistics will be provided for all variables, as appropriate. Continuous variables will be summarized by the number of observations, mean, standard deviation, median, interquartile range (Q1, Q3), minimum, and maximum. Categorical variables will be summarized by frequency counts and percentages for each category. The 95% confidence interval (CI) will be calculated as appropriate.

The primary, secondary CCI will be based on Full analysis set (FAS), which will include all patients who meet the required inclusion/exclusion criteria and have valid gene data from both plasma and tissue. FAS will be used for the analyses of all data collected.

The Data Cut-off (DCO) date of the primary endpoint, gene profile in tissue, will be when all participants have completed their gene testing in tissue and whole blood and got their testing

results. The Data Cut-off (DCO) date is excepted to be approximately 12 weeks after LSI. The primary objective is to describe the gene profile in *EGFR*m advanced NSCLC patients after progression on 1L Osimertinib treatment. Gene profile in tissue will be analyzed by NGS method. All gene alterations in tissue(e.g., gene mutation, amplification, deletion, fusion, etc.) detected by NGS will be observed. Proportion of Gene X alteration(%) = (number of patients with Gene X, which is an example of a certain gene)/(total number of patients in the FAS)×100%. Some cases which patients have more than one gene alteration need to be considered.

The secondary objective is to assess gene profile in plasma, concordance of plasma and tissue and the percentage of pathology transformation will be analysed by NGS method and IHC respectively. All gene alterations in plasma(e.g., gene mutation, amplification, deletion, fusion, etc.) detected will be observed. Proportion of Gene X alteration(%) = (number of patients with Gene X, which is an example of a certain gene)/(total number of patients in the FAS)□100%. Some cases which patients have more than one gene alteration need to be considered. Using selected representative genes (for example *EGFR*, MET, PIK3CA and etc.) to measure the concordance. Concordance of Gene X in plasma and tissue is defined as sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) between plasma and tissue results. Tissue sample is the reference standard. Sensitivity=(number of patients with positive result in both plasma and tissue)/(total number of patients with positive result in tissue samples)□100%; Specificity=(number of patients with negative result in both plasma and tissue)/(total number of patients with negative result in tissue samples)□100%. PPV (%)=(number of patients with positive result in both plasma and tissue)/(total number of patients with positive result in plasma samples)□100%; NPV (%)=(number of patients with negative result in both plasma and tissue)/(total number of patients with negative result in plasma samples)□100%.

Pathology transformation defines as pathological type of lung cancer change, which will be confirmed by pathologists. Number and percentage of patients with pathology transformation will be summarized. Proportion of pathology transformation(%) = (number of patients with pathology transformation)/(total number of patients in the FAS)□100%.

Two analyses are planned. The first interim analysis data cut-off (DCO) point will happen when 100 patients have completed their gene testing in tissue and whole blood and got their testing results(50% maturity). The final analysis is planned for when the last patient has completed their gene testing in tissue and whole blood and got their testing results.

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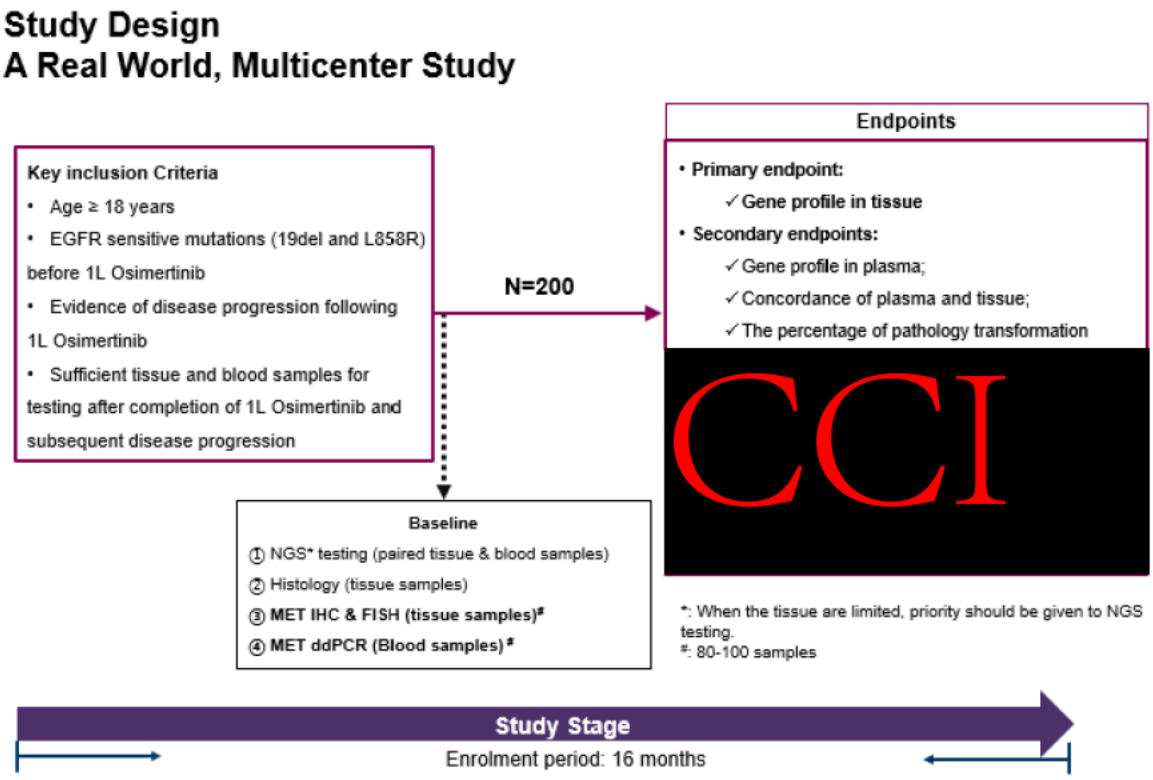
1.2 Schema

Figure 1 Study Design



RECIST 1.1=Response Evaluation Criteria in Solid Tumors, Version 1.1.
NSCLC, non-small cell lung cancer; *EGFR*, epidermal growth factor receptor.

1.3 Schedule of Activities

The procedures for this study are presented in the SoA (Table 1).

Table 1 Schedule of Activities

Procedure	Screening and Data Collection		Notes	Details in CSP Section or Appendix
Visit	**1**			
Informed consent [a]	X			Sections 5.1
Inclusion and exclusion criteria	X			Section 5.1 and 5.2
Diagnostic clinical procedures				
Histology	X			
Clinical stage with lymph node and distant metastasis at diagnosis and post 1L Osimertinib	X			
Record documented common *EGFR* mutation results by an accredited laboratory through ARMS, Super-ARMS, and NGS testing [b]	X			
Routine clinical procedures				
Demography and smoking history [c]	X			Section 7.2.1
Physical examination and weight(Kgs)	X			Section 7.2.1
Height(CMs)	X			Section 7.2.1
Medical history and comorbid conditions [d]	X			Sections 5.1 and 5.2
WHO/ECOG performance status post Osimertinib 1L treatment	X			Section 7.2.2
Osimertinib Efficacy measurements				
Duration of 1L Osimertinib treatment [e]	X			
Best response of 1L Osimertinib	X			
Progression pattern post 1L Osimertinib	X			
Genetic analysis				

Procedure	Screening and Data Collection		Notes	Details in CSP Section or Appendix
Mandatory screening tumour tissue sample (archived or newly acquired biopsy) for Central NGS testing and central MET FISH testing; local IHC testing (included MET overexpression by MET IHC and SCLC pathology transformation by IHC)	X			Section 7.3.1
Mandatory screening blood sample (newly acquired; 2*10ml) for Central NGS testing and central ddPCR MET amplification testing	X			Section 7.3.1and Appendix C

[a] Written informed consent and any locally required privacy act document authorisation must be obtained prior to performing any protocol-specific procedures, including screening/baseline evaluations.

[b] Participants are eligible to be considered for inclusion on the basis of local pre-existing *EGFR* tumour tissue or plasma testing results by molecular testing.

[c] Include date of birth or age, gender, race, and ethnicity for all participants.

[d] Include any treatment history before enrolling the study, complication and related history(chronic diseases which require treatment currently)

[e] Include the start and end dates of 1L Osimertinib treatment; starting dose,

Note: Data collection following study analysis until the end of the study is described in Section 7.
Note: The screening visit will confirm that the patients will be receiving a biopsy (scheduled for after enrolment), but they will not actually receive a biopsy at the screening visit.
CSP=Clinical Study Protocol; ECOG=Eastern Cooperative Oncology Group; ICF=informed consent form; WHO=World Health Organisation

2 INTRODUCTION

According to the GLOBOCAN estimates of cancer incidence and mortality, lung cancer has become the most commonly diagnosed cancer (11.6% of the total cases) and the leading cause of cancer death (18.4% of the total cancer deaths) worldwide[13]. Non-small cell lung cancer (NSCLC) represents approximately 80%-85% of all lung cancers. At the time of diagnosis, about 70% of NSCLC patients have locally advanced or metastatic disease who are not amenable to surgical resection. The 5-year overall survival rate for NSCLC patients with stage IV remains poor[14].

Fortunately, owing to the discovery of Epidermal Growth Factor Receptor (*EGFR*) pathway and the development of *EGFR*-targeted tyrosine kinase (TKI), the prognosis of advanced *EGFR* sensitive mutated (*EGFR*m+) NSCLC patients has improved significantly. The oncogenic *EGFR* mutation is present in 40-55% and 5-15% of lung adenocarcinoma cases in East Asia and in USA/European respectively[15]. The benefit of these *EGFR*-TKIs in *EGFR*m+ patients was initially demonstrated in the second-line and maintenance settings and subsequently confirmed in the first-line setting. As a result, the first- and second-generation *EGFR*-TKIs have been widely used in *EGFR*m+ NSCLC patients and brought improved ORR and PFS, but the benefit of overall survival is limited[16-18]. Based on the statistically significant improved PFS (18.9 months vs. 10.2 months; hazard ratio for disease progression or death, 0.46; 95% confidence interval [CI], 0.37 to 0.57; P<0.001) and OS (38.6 months vs. 31.8 months; hazard ratio for death, 0.80; 95.05% CI, 0.64 to 1.00; P = 0.046) from FLAURA study, the third-generation *EGFR*-TKI Osimertinib has been become the new standard of care for advanced *EGFR* mutated NSCLC patients and has recommended as the preferred first-line treatment by NCCN guideline[1]. In China, Osimertinib has been approved as a first-line treatment for *EGFR* mutated advanced NSCLC in Aug 2019.

2.1 Study Rationale

The third-generation *EGFR*-TKI Osimertinib has been approved as first-line treatment for advanced *EGFR* mutated NSCLC in China, and has also been widely used in clinical practice no matter in the first-line or in the later-line. Prior reports of acquired resistance mechanisms to Osimertinib primarily focus on patients who received Osimertinib as the second-line treatment. Although some small sample studies have reported the possible resistance mechanisms of Osimertinib in the first-line treatment, it is still an urgent need to explore the whole gene profile in *EGFR*m advanced NSCLC patients post Osimertinib 1L treatment failure by paired tissue and plasma after disease progression following 1L Osimertinib to guide subsequent treatment strategy[6, 7].

MET overexpression is key resistance mechanism after 1L Osimertinib failure[8]. SAVANNAH observed improved outcomes in post 1L Osimertinib resistant EGFRm NSCLC

patients with MET IHC90+(≥90% tumour cells MET IHC3+) and/or FISH10+(MET copy number ≥10); the estimated prevalence of IHC90+ is 29% in SAVANNAH study published on 2022 WCLC [9]. MET amplification is also a key resistance mechanism after 1L Osimertinib failure[8].FISH is golden standard testing method for MET amplification. The prevalence of FISH 5+(MET copy number ≥5 and/or MET/CEP7≥2) in INSIGHT 2 study [10] is 33% and the prevalence of FISH10+ in SAVANNAH study is 20% [9]. The prevalence data of MET amplification by FISH testing and MET overexpression by IHC testing in Chinese advanced EGFRm NSCLC patients who had disease progression following 1L osimertinib are very limited and still need further explore.

Tissue were unavailable for some patients post 1L Osimertinib and blood samples show high potential as an alternative sample [11]. ddPCR is as an alternative technique with high sensitivity and specificity for MET amplification testing and it has been explored in previous study[12]. Besides, ddPCR blood assay for MET amplification testing still need be further validated through concordance of MET amplification between FISH testing in tissue and ddPCR testing in plasma.

This real-world, multi-center study will elucidate the gene profile post Osimertinib 1L treatment in tissue and plasma and will also help to guide the following treatment.

2.2 Background

While similar to 1/2 G *EGFR*-TKI, acquired resistance to Osimertinib invariably often develops with a median PFS of 19 months[1]. The characterization of the landscape of resistance enables the development of subsequent therapies, but mechanisms of resistance to Osimertinib is primarily derived from patients who received Osimertinib after other *EGFR*-TKIs, such as *EGFR* C797, *EGFR* G724, *EGFR* L792, *MET* amplification, and so on[2-5]. Besides, MET overexpression is also a key resistance mechanism after 1L Osimertinib failure[8]and the prevalence data of MET amplification by FISH testing (gold standard testing method for MET amplification) and MET overexpression by MET IHC testing in Chinese advanced EGFRm NSCLC patients who had disease progression following 1L osimertinib are very limited. The mechanisms of resistance to Osimertinib in the first-line treatment are far from being clearly explored. As Osimertinib in the first-line treatment is applied more and more widely in advanced *EGFR*m+ NSCLC patients, we urgently need to know the resistance mechanisms to guide subsequent treatment strategy. Until now, the largest study to explore acquired resistance mechanism to 1L Osimertinib was FLAURA study, which detected 91 paired plasma samples by next generation sequencing (NGS) . The most common acquired resistance mechanisms were *MET* amplification (15%) and *EGFR* C797X mutation (7%)[6]. The result was quite different from the study conducted by Adam J. Schoenfeld et al using tissue (N=27), with squamous transformation accounting for 15% and *MET* amplification 7%[7]. The reason for different

results maybe because these two studies used different samples to analyse resistance mechanisms, and the above two detection methods may have different sensitivity and specificity. In addition, Schoenfeld et al study was limited to patients at one site, Memorial Sloan Kettering Cancer Center in NY, USA.

Currently, analysis of circulating tumor DNA(ctDNA) has been the predominant method to investigate acquired resistance mechanisms, but non-genetic mechanisms of resistance, including histologic transformation and protein expression change cannot be captured by plasma NGS analysis. Compared to tissue analysis, sensitivity for copy number changes and chromosomal rearrangements using plasma samples is instable[3, 4, 19-23]. ddPCR is as technique with high sensitivity and specificity, which is widely used for the absolute quantification of nucleic acid through Poisson distribution [24] and also as an alternative technique for MET amplification testing in plasma samples has been explored in some previous studies. Kitazono et al. have attempted to use ddPCR to detect MET amplification in three blood samples of NSCLC patients with EGFR-TKI resistance and proved its feasibility at the first time [25]. The recent study further explored the performance of MET amplification testing by ddPCR in tissues and paired peripheral blood samples from advanced NSCLC patients who had progressed on EGFR-TKI[26]. A total of tissues and the paired peripheral blood samples from 103 advanced NSCLC patients who had progressed on EGFR-TKI were collected for MET amplification testing by using ddPCR. In parallel, MET amplification was verified by FISH in tissue samples. The concordance rate of MET amplification in tissue samples between ddPCR and FISH was 100% (102/102), and it was 94.17% (97/103) by ddPCR testing between tissue and paired and peripheral blood samples with performance of sensitivity 66.67% and specificity 98.86%. Therefore, ddPCR is an potentially alternative method for MET amplification testing in both tissues and peripheral blood samples, which is worthy of exploring and validating in more studies (appendix H).

Thus, this study plans to use paired tumor tissue and whole blood to detect molecular and histologic mechanisms of resistance to 1L Osimertinib and to assess the concordance of plasma and tissue. And these paired tissue and blood samples are also used to assess the concordance of MET amplification between FISH and ddPCR testing as exploratory endpoints. Additional, the prevalence of MET amplification by FISH testing and MET overexpression by IHC testing, and the overlap ratio of prevalence between MET amplification and MET overexpression are also as exploratory endpoints in this study.

2.3 Benefit/Risk Assessment

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2.3.1 Risk Assessment

This is a real-world study, which will not affect or interfere with patients' routine medical

diagnosis and treatment, so it will not increase other related risks.

2.3.2 Benefit Assessment

The results of the research may not be directly used for patients' diagnosis and treatment, but the analysis of gene profile after Osimertinib 1L treatment will eventually help oncologists to make more effective treatment strategies in the future.

2.3.3 Overall Benefit: Risk Conclusion

Based on a review of the potential benefits and risks, it is considered to be reasonable and appropriate to investigate gene profile in *EGFR*m advanced NSCLC patients post Osimertinib 1L treatment failure

3 OBJECTIVES AND ENDPOINTS

Table 2 Objectives and Endpoints

Objectives	Endpoints
Primary	
To describe the gene profile in *EGFR*m advanced NSCLC patients post Osimertinib 1L treatment.	● Gene profile in tissue Gene profile defined as gene alternation detected by NGS and frequency of every gene alternation. All gene alterations include gene mutation, CNV, fusion, etc. The frequency of all gene alteration detected by NGS(%) = (number of patients with every gene alteration detected by NGS)/(total number of patients in the FAS)×100%.
Secondary	
To assess the concordance of plasma and tissue	● Gene profile in plasma Gene profile defined as gene alternation detected by NGS and frequency of every gene alternation. ● Concordance of plasma and tissue Using selected representative genes (for example *EGFR*, *MET*, *PIK3CA* and etc.) to measure the concordance. Concordance of Gene X in plasma and tissue is defined as sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) between plasma and tissue results., Tissue sample is the reference standard. - sensitivity=(number of patients with positive result in both plasma and tissue)/(total number of patients with positive result in tissue samples)×100%; - specificity=(number of patients with negative result in both plasma and tissue)/(total number of patients with negative result in tissue samples)×100%. - PPV (%)=(number of patients with positive result in both plasma and tissue)/(total number of patients with positive result in plasma samples)×100%; - NPV (%)=(number of patients with negative result in both plasma and tissue)/(total number of patients with negative result in plasma samples)×100% ● The percentage of pathology transformation Pathology transformation is defined as those transformation from non-small-cell lung cancer to small-cell lung cancer or from adenocarcinoma to squamous carcinoma, can be observed by IHC
CCI	

CCI	CCI
[redacted]	[redacted]

4 STUDY DESIGN

4.1 Overall Design

This is a real-world, multi-center prospective study elucidating the gene profile in common *EGFR*m (ex19del and L858R) locally advanced or metastatic NSCLC patients post Osimertinib 1L treatment. The main assumption is that gene profile in tissue and plasma could further guide the following treatment strategy.

Approximately 200 participants in *EGFR*m locally advanced or metastatic NSCLC with the evidence of disease progression following 1L Osimertinib will be enrolled.

Participants will be required to provide paired tissue and whole blood after disease progression following 1L Osimertinib. 200 tissue samples and 200 whole blood samples will be used to detect gene alteration by NGS, respectively. 200 tissue samples will be used to detect pathological transformation by IHC. The sample size of MET amplification testing by FISH and MET overexpression by MET IHC in tissue samples is approximately 80-100 respectively. The sample size for MET amplification testing by ddPCR in blood samples is the same.

Baseline information consists of demography, smoking history, physical examination, medical history, ECOG/WHO performance status, *EGFR* mutation type, histology, clinical stage with lymph node and distant metastasis at diagnosis and post 1L Osimertinib, duration of 1L Osimertinib treatment, best response of 1L Osimertinib and progression pattern post 1L Osimertinib.

4.2 Scientific Rationale for Study Design

4.2.1 Rationale for primary, secondary and exploratory endpoint

The primary objective is to describe the gene profile in tissue in *EGFR*m locally advanced or metastatic NSCLC patients post on 1L Osimertinib failure, which could help clinicians provide the following treatment strategy.

The secondary objective is to describe the gene profile in plasma, to assess the concordance of plasma and tissue, and the percentage of pathology transformation. Gene profile in plasma and the percentage of pathology transformation will be detected and analysed by NGS and IHC, respectively. Concordance of plasma and tissue is to explore whether plasma can be substitute for tissue when patients cannot accept tissue biopsy, which is defined as sensitivity, specificity, PPV and NPV of representative genes. According to the assay performance, sensitivity 80% is acceptable. If the point estimate for sensitivity is $\geqslant$ 80%, plasma samples are supposed to be consistent with tissue sample for NGS testing.

In the case of sufficient tissue samples, the exploratory endpoints is further to assess prevalence of MET amplification by FISH in tissue samples, the concordance of MET amplification

between FISH testing in tissue samples and ddPCR testing in blood samples, the prevalence of MET overexpression by MET IHC testing and the overlap ratio of prevalence between MET amplification by FISH and MET overexpression by MET IHC. The prevalence data of MET overexpression by MET IHC and MET amplification by FISH in post 1L Osimertinib resistance patients in China are very limited.

Tissue were unavailable for some patients post 1L Osimertinib and ddPCR testing will offer alternative method for MET amplification testing in blood samples. Concordance of MET amplification testing between ddPCR in plasma samples and FISH in tissue samples is to explore whether plasma can be substitute for tissue when patients cannot accept tissue biopsy, which is defined as sensitivity, specificity, PPV and NPV of representative genes.

4.2.2 Testing for common *EGFR* mutations

Only participants with NSCLC with common *EGFR* mutations of ex19del and L858R will be eligible for inclusion in the study. A patient can be considered eligible for the study on the basis of documented *EGFR*m through any of ARMS, Super-ARMS, NGS analysis from accredited laboratories.

For patients eligible for inclusion in the study based on the pre-existing common *EGFR*m plasma and/or tissue sample results, the local *EGFR* tissue testing laboratory methods will be recorded in the respective electronic case report forms (eCRF).

4.3 End of Study Definition

The end of the study is defined as the date of the scheduled visit of the last participant in the study shown in the SoA in the study.

The study is expected to start participant enrolment at in Q1 2022 and to end by Q1 2024.

The study may be terminated at individual centres if the study procedures are not being performed according to GCP, or if recruitment is slow. Depend on the determination of the study team. If the site is no patients enrol lasts for 3 months from the site initiation, it will be defined as slow recruitment.

A participant is considered to have completed the study if he/she has completed all phases of the study including the scheduled procedure shown in the SoA.

The study may be stopped if, in the judgment of AstraZeneca, study participants are placed at undue risk because of clinically significant findings.

5 STUDY POPULATION

The target population of interest in this study is participants with locally advanced or metastatic

NSCLC, harbouring common *EGFR*m of ex19del/L858R and having evidence of disease progression following 1L Osimertinib.

Prospective approval of protocol deviations to recruitment and enrolment criteria, also known as protocol waivers or exemptions, is not permitted.

Each subject should must meet all of the inclusion criteria and none of the exclusion criteria for this study. Under no circumstances can there be exceptions to this rule. Participants who do not meet the eligibility criteria requirements are screen failures; refer to Section 5.4.

5.1 Inclusion Criteria

Participants are eligible to be included in the study only if all of the following criteria apply:

Informed Consent

1 Capable of giving signed informed consent, which includes compliance with the requirements and restrictions listed in the informed consent form (ICF) and in this protocol.

2 Provision of signed and dated, written informed consent form prior to any mandatory study-specific procedures, sampling, and analyses.

The ICF process is described in Appendix A3.,

Sex and **Age**

3 Male or female, age at least 18 years.

Type of Participant and Disease Characteristics

4 Pathologically confirmed non-small cell lung cancer (NSCLC) with documented *EGFR* sensitive mutation (*EGFR* 19del and L858R) positive before Osimertinib 1L.

5 Locally advanced (clinical stage IIIB, IIIC) or metastatic NSCLC(clinical stage IVA or IVB) or recurrent NSCLC (per Version 8 of the International Association for the Study of Lung Cancer [IASLC] Staging Manual in Thoracic Oncology), not amenable to curative or radiotherapy (e.g., this may occur as systemic recurrence after prior surgery for early stage disease or patients may be newly diagnosed with stage IIIB/IV disease, which is at the start of Osimertinib therapy).

6 Patients must have been treated with Osimertinib as first line therapy until disease progression. Evidence of disease progression following 1L Osimertinib can be confirmed by investigators with criteria in Response Evaluation Criteria in Solid Tumours (RECIST) Version 1.1.

7 Agree to provide adequate tissue and whole blood for testing after disease progression following 1L Osimertinib.

Reproduction

8 Female participants who are not abstinent (in line with the preferred and usual lifestyle choice of the participant) and intend to be sexually active with a male partner must be using highly effective contraceptive measures, must not be breast feeding, and must have a negative pregnancy test prior to the enrolment or must have evidence of non-child-bearing potential by fulfilling 1 of the following criteria at screening:

- Post-menopausal, defined as more than 50 years of age and amenorrhoeic for at least 12 months following cessation of all exogenous hormonal treatments
- Women under 50 years old would be considered as postmenopausal if they have been amenorrhoeic for 12 months or more following cessation of exogenous hormonal treatments and have luteinizing hormone (LH) and follicle-stimulating hormone (FSH) levels in the post-menopausal range for the institution
- Documentation of irreversible surgical sterilization by hysterectomy, bilateral oophorectomy or bilateral salpingectomy but not tubal ligation.

Further information is available in Appendix E (Definition of Women of Childbearing Potential and Acceptable Contraceptive Methods).

9 Male participants must be willing to use barrier contraception.

5.2 Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply:

Medical Conditions

1 Any concurrent and/or other active malignancy that may affect tissue or whole blood testing results.

2 As judged by the investigator, any evidence of severe or uncontrolled systemic diseases, which in the investigator's opinion, makes it undesirable for the participant to participate in the study or that would jeopardise compliance with the protocol,. Screening for chronic conditions is not required.

Prior/Concomitant Therapy

3 Any concurrent anticancer treatment except local radiotherapy and radiotherapy for CNS metastasis. Concurrent use of hormonal therapy for non-cancer-related conditions (eg, hormone replacement therapy) is allowed.

4

Prior/Concurrent Clinical Study Experience

5 Judgment by the investigator that the patient should not participate in the study if the patient is unlikely to comply with study procedures, restrictions and requirements.

Other Exclusions

6 Involvement in the planning and/or conduct of the study (applies to both AstraZeneca staff and/or staff at the study site).

7 In addition, the following are considered criteria for exclusion from the exploratory genetic research:

- Prior allogeneic bone marrow transplant
- Non-leukocyte depleted whole blood transfusion within 120 days of genetic sample collection.

5.3 Lifestyle Considerations

1 Female participants of child-bearing potential who are not abstinent (in line with the preferred and usual lifestyle choice of the participant) and intend to be sexually active with a male partner must use acceptable methods of contraception from screening until at least 6 weeks after discontinuing study treatment.

2 Male participants must use barrier contraceptives (condoms) during sex with a female partner of child-bearing potential (including a pregnant partner) from the time of screening throughout the completion of the study.

Participants must follow the contraception requirements outlined in Appendix E.

5.4 Screen Failures

Screen failures are defined as participants who consent to participate in the clinical study but are not subsequently entered in the study. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants to meet the Consolidated Standards of Reporting Trials publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria.

5.5 Procedures for handling incorrectly enrolled patients

Patients who fail to meet the eligibility criteria should not, under any circumstances, be enrolled. There can be no exceptions to this rule. Where a patient does not meet all the eligibility criteria but is enrolled in error, the Investigator should inform the AstraZeneca Study Physician immediately, and a discussion should occur between the AstraZeneca Study Physician and the Investigator regarding whether to continue or discontinue the patient from the study. The Study Physician should ensure all decisions are appropriately documented. The Investigator should make documentation in the medical record as appropriate.

6 PARTICIPANT DISCONTINUATION/WITHDRAWAL

6.1 Participant Withdrawal from the Study

- A participant may withdraw from the study at any time at his/her own request, or may be withdrawn at any time at the discretion of the investigator for behavioural, compliance, or administrative reasons. This is expected to be uncommon.
- At the time of withdrawal from the study, if possible, an Early Study Intervention Discontinuation visit should be conducted, as shown in the SoA. See SoA for data to be collected at the time of study withdrawal and for any further evaluations that need to be completed.
- If the participant withdraws consent for disclosure of future information, the sponsor may retain and continue to use any data collected before such a withdrawal of consent.
- If a participant withdraws from the study, it should be confirmed if he/she is still agrees for existing samples to be used in line with the original consent. If he/she requests withdrawal of consent for use of samples, destruction of any samples taken and not tested should be carried in line with what was stated in the informed consent and local regulation. The investigator must document the decision on use of existing samples in the site study records and inform the Global Study Team.

6.2 Lost to Follow-up

A participant will be considered lost to follow-up if he or she repeatedly fails to return for scheduled visits and no contact has been established by the time the study is completed (see Section 4.3), such that there is insufficient information to determine the participant's status at that time.

Participants who decline to continue participation in the study should be documented as "withdrawal of consent" rather than "lost to follow-up." Investigators should document attempts to re-establish contact with missing participants throughout the study period. If contact with a missing participant is re-established, the participant should not be considered lost to follow-up and evaluations should resume according to the protocol.

The following actions must be taken if a participant fails to return to the clinic for a required study visit:

- The site must attempt to contact the participant and reschedule the missed visit as soon as possible and counsel the participant on the importance of maintaining the assigned visit schedule and ascertain whether or not the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the investigator or designee must make every effort to regain contact with the participant (where possible, 3 telephone calls and, if

necessary, a certified letter to the participant's last known mailing address or local equivalent methods). These contact attempts should be documented in the participant's medical record.

Discontinuation of specific sites or of the study as a whole are handled as part of Appendix A.

7 STUDY ASSESSMENTS AND PROCEDURES

Study procedures and their timing are summarized in the SoA. Data collection following study analysis until the end of the study is described below.

- Protocol waivers or exemptions are not allowed.
- Immediate safety concerns should be discussed with the sponsor immediately upon occurrence or awareness to determine if the participant should continue or discontinue study intervention.
- Adherence to the study design requirements, including those specified in the SoA, is essential and required for study conduct.
- All screening evaluations must be completed and reviewed to confirm that potential participants meet all eligibility criteria. The investigator will maintain a screening log to record details of all participants screened and to confirm eligibility or record reasons for screening failure, as applicable.
- Procedures conducted as part of the participant's routine clinical management (eg, blood count) and obtained before signing of the ICF may be utilised for screening or baseline purposes provided the procedures met the protocol-specified criteria and were performed within the time frame defined in the SoA.

7.1 Enrolment/screening period

At screening, consenting patients are assessed to ensure that they meet eligibility criteria. Patients who do not meet these criteria must not be enrolled into the study.

The following will be performed at screening:

Written informed consent

Each potential patient will provide written informed consent prior to starting any study specific procedures.

All patients will be required to consent for collection of a tumor sample and a plasma sample after disease progressed during 1L Osimertinib. This consent is included in the main patient informed consent form (ICF).

Assignment of patient screening number

As per standard, enrolment number(E-code) is assigned to the patient and Principle Investigator or delegate should perform enrolment/screening call.

Collect clinical information

Demographic data and other characteristics will be recorded and will include date of birth or age, gender, race and/or ethnicity, and smoking history, *EGFR* mutation type, histology, clinical stage with lymph node and distant metastasis at diagnosis and post 1L Osimertinib, ECOG/WHO performance status through electronic medical record or information follow-up. A standard medical and surgical history will be obtained. Duration of 1L Osimertinib treatment, best response of 1L Osimertinib and progression pattern post 1L Osimertinib, will be collected for analysing the correlation between gene profile post 1L Osimertinib and effect of Osimertinib 1L treatment.

7.2 General Assessments

7.2.1 Physical Examinations

Physical examination will be performed and include assessments of the following; general appearance, skin, head and neck (including ears, eyes, nose and throat), respiratory, cardiovascular, abdomen, lymph nodes, thyroid, musculoskeletal (including spine and extremities) and neurological systems.

Physical examination, weight, height will be measured during screening only.

7.2.2 WHO/ECOG Performance Status

WHO/ECOG performance status will be assessed at the times specified in the SoA based on the following:

0 Fully active; able to carry out all usual activities without restrictions.

1 Restricted in strenuous activity, but ambulatory and able to carry out light work or work of a sedentary nature (eg, light housework or office work).

2 Ambulatory and capable of self-care, but unable to carry out any work activities; up and about more than 50% of waking hours.

3 Capable of only limited self-care; confined to bed or chair more than 50% of waking hours.

4 Completely disabled; unable to carry out any self-care and totally confined to bed or chair.

5 Dead.

7.3 Biomarker research

Instructions for the collection, handling, storage and shipping of biological samples will be provided in the study-specific laboratory manual, Appendix B and Appendix C. Samples should be stored in a secure storage space with adequate measures to protect confidentiality.

Samples will be destroyed at end of the study (as defined in the protocol) in line with consent and local requirements.

- Samples collected in China will be stored and disposed of according to local laws and regulations during the study.
- Remaining sample aliquots will be destroyed following completion of the study.

For further details on Handling of Human Biological Samples, see Appendix B.

7.3.1 Collection of Samples for Gene Profile Analysis

The subject's consent to the use of donated biological samples for research is mandatory at disease progression following 1L Osimertinib in the SoA Samples will be collected from all participants and be used for detecting gene alteration and pathological transformation to explore the whole gene profile post Osimertinib 1L treatment for guiding subsequent treatment strategy.

A unstained, archival tumor tissue sample from a fresh biopsy tissuein a quantity sufficient to allow for gene profile at the time of screening will be collected. Tissue samples will be used for the extraction and analysis of deoxyribonucleic acid (DNA). For the requirement of tissue sample and plasma sample, refer to appendix F.

Whole blood samples (approx. 2*10 mL) will be taken at disease progression post Osimertinib 1L treatment in the SoA (Table 1). Blood samples to generate plasma samples will be collected from all participants. These samples will be used for the extraction and analysis of circulating tumour deoxyribonucleic acid (ctDNA).

Genetic profiles in tissue and plasma will be analyzed by the central lab NGS testingand then used to analysis positive percentage agreement, sensitivity/specificity of plasma to evaluate concordance between plasma and tissue for exploring whether plasma can be substitute for tissue when patients cannot accept tissue biopsy. The analytical method will be used is described in Section 8.3.2.

7.3.2 Other Study Related Biomarker Assessments

Already collected tissue samples may be analysed on different biomarkers thought to play a role in protocol-specific rationale including, but not limited to, tissue biomarkers of SCLC by IHC to evaluate pathology transformation.Biomarkers of SCLC include, but not limited to, CD56, Syn, CgA, TTF-1, CK, Ki-67 and CAM5.2, which is the routine testing in the clinical practice. The analytical method will be used is described in Section 8.3.2.

In the case of sufficient tissue samples, these collected tissue samples may be analyzed on the prevalence of MET amplification and MET overexpression; the concordance of MET amplification between FSIH in tissue samples and ddPCR in blood samples; the overlap ratio

of prevalence between MET amplification by FISH and MET overexpression by MET IHC.

The MET amplification testing by FISH in tissue samples and by ddPCR in blood samples (approx. 10 mL) will be tested in central lab.

MET overexpression by MET IHC testing in tissue samples will be tested in the pathology department of local sites.

The detailed analytical method will be used is described in Section 8.3.2.2.3.

7.3.3 Storage and destruction of samples

The processes adopted for the coding and storage of samples for genetic analysis are important to maintain subject confidentiality. Sample storage and destruction will be performed according to the central labs' policy and China HGR regulations. DNA is a finite resource that may be used up during experiments. The results of any further analyses will be reported either in the Clinical Study Report itself or as an addendum, or separately in a scientific report or publication.

All genetic testing and analysis will be performed after China HGR approval. Mutation testing residual samples collected will be destroyed.

For further details on Handling of Human Biological Samples, including storage, re-use and destruction, refer to Appendix B and the Laboratory Manual.

8 STATISTICAL CONSIDERATIONS

Statistical analyses will be performed by AstraZeneca or its representatives. A comprehensive SAP will be prepared with final amendments completed prior to database lock.

There is a potential selection biases in this study. Selection bias: the patients who are eligible and consent to participate in the current study will be enrolled consecutively and without personal preference.

8.1 Sample Size Determination

Approximately 200 participants will be enrolled to achieve NGS and IHC testing.

Assuming some common gene alteration with an incidence of 20%, then with 200 subjects the 95% CI will be around 14.7% - 26.2% based on an exact method (Clopper-Pearson).

Table below summarizes the 95% CIs for the target incidences ranging from 10% to 50% with sample size of 200.

Assumed incidence of gene mutation	95% CIs
10%	6.2%, 15.0%
20%	14.7%, 26.2%
30%	23.7%, 36.9%
40%	33.2%, 47.1%
50%	42.9%, 57.1%

In addition, 200 subjects are enough to have 86% probability to observe at least one case if the incidence of an gene alteration is 1%.

Note: "Enrolled" means a participant's, or their legally acceptable representative's, agreement to participate in a clinical study following completion of the informed consent process. Potential participants who are screened for the purpose of determining eligibility for the study, but are not enrolled in the study, are considered" screen failures", unless otherwise specified by the protocol.

Assuming a potential dropout rate for loss to follow-up of 10%, the number to be accrued will be 180.

8.2 Populations for Analyses

The following populations are defined:

Table 3 Populations for Analysis

Population/Analysis Set	Description
Enrolled	All participants who sign the ICF
FAS	All participants who meet the required inclusion/exclusion criteria and have valid Gene data from both plasma and tissue. The FAS will be used for the analyses of all data.

8.3 Statistical Analyses

The SAP will be finalised prior to database lock and it will include a more technical and detailed description of the statistical analyses described in this section. This section is a summary of the planned statistical analyses of the most important endpoints including primary and key

secondary endpoints.

8.3.1 General Considerations

Descriptive statistics will be provided for all variables, as appropriate. Continuous variables will be summarized by the number of observations, mean, standard deviation, median, interquartile range (Q1, Q3), minimum, and maximum. Categorical variables will be summarized by frequency counts and percentages for each category. The 95% confidence interval (CI) will be calculated as appropriate.

8.3.2 Biomarker analysis

8.3.2.1 Primary Endpoint

To evaluate the potential molecular resistance mechanism by tissue analysis at disease progression post 1L Osimertinib treatment, tissue samples will be collected at the disease progression. And then the gene profiling of molecular alterations including but not limited to mutations in, amplifications, deletion, fusion and expression of relevant pathway gene will be explored by NGS analysis.

The primary objective is to describe the gene profile in *EGFR*m advanced NSCLC Patients after progression on 1L osimertinib treatment.

Gene profile in tissue will be analysed by NGS method. All gene alterations(e.g., gene mutation, amplification, deletion, fusion, etc.) and histological transformation detected will be observed. Proportion of Gene X alteration(%) = (number of patients with Gene X, which is an example of a certain gene)/(total number of patients in the FAS)×100%. Some cases which patients have more than one gene alteration need to be considered.

8.3.2.2 Secondary Endpoints

The secondary objective is to assess the concordance of plasma and tissue. Gene profile in plasma, concordance of plasma and tissue and the percentage of pathology transformation will be analysed by NGS method and IHC respectively.

8.3.2.2.1 Gene profile in plasma

All gene alterations(e.g., gene mutation, amplification, deletion, fusion, etc.) and histological transformation detected will be observed. Proportion of Gene X alteration(%) = (number of patients with Gene X, which is an example of a certain gene)/(total number of patients in the FAS)×100%. Some cases which patients have more than one gene alteration need to be considered. The 95% confidence intervals using Pearson-Clopper method will be provided.

8.3.2.2.2 Concordance of plasma and tissue

Using selected representative genes (for example *EGFR*, MET, PIK3CA and etc.) to measure the concordance. Concordance of Gene X in plasma and tissue is defined as sensitivity,

specificity, positive predictive value (PPV) and negative predictive value (NPV) between plasma and tissue results. Tissue sample is the reference standard. The 95% confidence intervals using Pearson-Clopper method will be provided.

- sensitivity=(number of patients with positive result in both plasma and tissue)/(total number of patients with positive result in tissue samples)×100%;

- specificity=(number of patients with negative result in both plasma and tissue)/(total number of patients with negative result in tissue samples)×100%.

- PPV (%)=(number of patients with positive result in both plasma and tissue)/(total number of patients with positive result in plasma samples)×100%;

- NPV (%)=(number of patients with negative result in both plasma and tissue)/(total number of patients with negative result in plasma samples)×100%

8.3.2.2.3 The percentage of pathology transformation

Pathology transformation defines as pathological type of lung cancer change, which will be confirmed by pathologists. Number and percentage of patients with pathology transformation will be summarized. Proportion of pathology transformation(%) = (number of patients with pathology transformation)/(total number of patients in the FAS)□100%. The 95% confidence intervals using Pearson-Clopper method will be provided.

CCI [redacted]

8.4 Data Analyses

The SAP will describe the planned interim analyses and final analyses in greater detail.

Two analyses are planned. The first interim analysis data cut-off (DCO) point will happen when 100 patients have completed their gene testing in tissue and whole blood and got their testing results. The final analysis is planned when the last patient has completed their gene testing in tissue and whole blood and got their testing results.

9 DEFINITIONS AND REPORTING OF ADVERSE EVENTS

9.1 Definition of Adverse Event

An AE is the development of any untoward medical occurrence (other than progression of the malignancy under evaluation) in a participant or clinical study participant administered a study intervention and which does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavourable and unintended sign (eg, an abnormal laboratory finding), symptom (for example nausea, chest pain), or disease temporally

associated with the use of a medicinal product, whether or not considered related to the study intervention.

The term AE is used to include both serious and non-serious AEs and can include a deterioration of a pre-existing medical occurrence. An AE may occur at any time, including run-in or washout periods, even if no study intervention has been administered.

9.2 Definition of Serious Adverse Event

An SAE is an AE occurring during any study phase (ie, run-in, treatment, washout, follow-up), that fulfils one or more of the following criteria:

- Results in death.
- Is immediately life-threatening.
- Requires in-participant hospitalisation or prolongation of existing hospitalisation.
- Results in persistent or significant disability or incapacity.
- Is a congenital abnormality or birth defect.
- Is an important medical event that may jeopardise the participant or may require medical treatment to prevent one of the outcomes listed above.

AEs for **malignant tumours** reported during a study should generally be assessed as **Serious** AEs. If no other seriousness criteria apply, the "Important Medical Event" criterion should be used. In certain situations, however, medical judgment on an individual event basis should be applied to clarify that the malignant tumour event should be assessed and reported as a **Non-Serious** AE. For example, if the tumour is included as medical history and progression occurs during the study, but the progression does not change treatment and/or prognosis of the malignant tumour, the AE may not fulfil the attributes for being assessed as serious, although reporting of the progression of the malignant tumour as an AE is valid and should occur. Also, some types of malignant tumours, which do not spread remotely after a routine treatment that does not require hospitalisation, may be assessed as Non-Serious; examples in adults include Stage 1 basal cell carcinoma and Stage 1A1 cervical cancer removed via cone biopsy.

The above instruction applies only when the malignant tumour event in question is a new malignant tumour (ie, it is *not* the tumour for which entry into the study is a criterion and that is being treated by the study intervention under study and is not the development of new or progression of existing metastasis to the tumour under study). Malignant tumours that – as part of normal, if rare, progression – undergo transformation (e.g. Richter's transformation of B cell chronic lymphocytic leukaemia into diffuse large B cell lymphoma) should not be considered a new malignant tumour.

Life threatening

"Life-threatening" means that the participant was at immediate risk of death from the AE as it

occurred or it is suspected that use or continued use of the product would result in the participant's death. "Life-threatening" does not mean that had an AE occurred in a more severe form it might have caused death (e.g. hepatitis that resolved without hepatic failure).

Hospitalisation

Outpatient treatment in an emergency room is not in itself a SAE, although the reasons for it may be (eg, bronchospasm, laryngeal oedema). Hospital admissions and/or surgical operations planned before or during a study are not considered AEs if the illness or disease existed before the participant was enrolled in the study, provided that it did not deteriorate in an unexpected way during the study.

Important medical event or medical treatment

Medical and scientific judgment should be exercised in deciding whether a case is serious in situations where important medical events may not be immediately life threatening or result in death, hospitalisation, disability or incapacity but may jeopardise the participant or may require medical treatment to prevent one or more outcomes listed in the definition of serious. These should usually be considered as serious.

Simply stopping the suspect drug does not mean that it is an important medical event; medical judgment must be used.

- Angioedema not severe enough to require intubation but requiring iv hydrocortisone treatment
- Hepatotoxicity caused by paracetamol (acetaminophen) overdose requiring treatment with N-acetylcysteine
- Intensive treatment in an emergency room or at home for allergic bronchospasm
- Blood dyscrasias (e.g. neutropenia or anaemia requiring blood transfusion, etc.) or convulsions that do not result in hospitalisation
- Development of drug dependency or drug abuse

Intensity rating scale:

The grading scales found in the revised National Cancer Institute CTCAE latest version 5.0 will be utilised for all events with an assigned CTCAE grading. For those events without assigned CTCAE grades, the recommendation in the CTCAE criteria that converts mild, moderate and severe events into CTCAE grades should be used. A copy of the CTCAE can be downloaded from the Cancer Therapy Evaluation Program website (http://ctep.cancer.gov). The applicable version of CTCAE should be described clearly.

It is important to distinguish between serious and severe AEs. Severity is a measure of intensity whereas seriousness is defined by the criteria in Section 9.2. An AE of severe

intensity need not necessarily be considered serious. For example, nausea that persists for several hours may be considered severe nausea, but not a SAE unless it meets the criteria shown in Section 9.2. On the other hand, a stroke that results in only a limited degree of disability may be considered a mild stroke but would be a SAE when it satisfies the criteria shown in Section 9.2.

9.3 Definition of Adverse Drug Reaction (ADR)

An Adverse Drug Reaction (ADR) is an adverse event suspected to be causally related to the medicinal product.

9.4 Reporting of Adverse Events

Since this study does not involve a specific AZ product, there is no requirement of proactive collection of AEs and other safety data for this study. However, we encourage investigators to report any ADRs related to AstraZeneca products to AstraZeneca spontaneously.

Contact information of AstraZeneca PV China Team:

Hotline: 4008208116 (Mobile & Landline Telephone)

8008208116 (Landline Telephone)

(Opening hours: 8:30-17:30 every working day)

Online: https://aereporting.astrazeneca.com

10 SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

Appendix A Regulatory, Ethical, and Study Oversight Considerations

A 1 Regulatory and Ethical Considerations

- This study will be conducted in accordance with the protocol and with the following:
  - Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines
  - Applicable ICH GCP Guidelines
  - Applicable laws and regulations
- The protocol, protocol amendments, ICF, IB, and other relevant documents (eg, advertisements) must be submitted to an IRB/IEC by the investigator and reviewed and approved by the IRB/IEC before the study is initiated.
- Any amendments to the protocol will require IRB/IEC and applicable Regulatory Authority approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.
- AstraZeneca will be responsible for obtaining the required authorisations to conduct the study from the concerned Regulatory Authority. This responsibility may be delegated to a Contract Research Organisation but the accountability remains with AstraZeneca.

A 2 Financial Disclosure

Investigators and sub-investigators will provide the sponsor with sufficient, accurate financial information as requested to allow the sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are responsible for providing information on financial interests during the course of the study and for 1 year after completion of the study.

A 3 Informed Consent Process

The investigator or his/her representative will explain the nature of the study to the participant or his/her legally authorised representative and answer all questions regarding the study.

Participants must be informed that their participation is voluntary and they are free to refuse to participate and may withdraw their consent at any time and for any reason during the study. Participants or their legally authorised representative will be required to sign a statement of informed consent that meets the requirements of 21 Code of Federal Regulations 50, local regulations, ICH guidelines, Health Insurance Portability and Accountability Act requirements, where applicable, and the IRB/IEC or study centre.

The medical record must include a statement that written informed consent was obtained before the participant was enrolled in the study and the date the written consent was obtained. The authorised person obtaining the informed consent must also sign the ICF.

Participants must be re-consented to the most current version of the ICF(s) during their participation in the study.

A copy of the ICF(s) must be provided to the participant or the participant's legally authorised representative.

Participants who are rescreened are required to sign a new ICF.The investigator or authorised designee will explain to each participant the objectives of the analysis to be done on the samples and any potential future use. Participants will be told that they are free to refuse to participate in the future use and may withdraw their consent at any time and for any reason during the retention period.

A 4 Data Protection

- Participants will be assigned a unique identifier by the sponsor. Any participant records or datasets that are transferred to the sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred.
- The participant must be informed that his/her personal study-related data will be used by the sponsor in accordance with local data protection law. The level of disclosure and use of their data must also be explained to the participant in the informed consent
- The participant must be informed that his/her medical records may be examined by Clinical Quality Assurance auditors or other authorised personnel appointed by the sponsor, by appropriate IRB/IEC members, and by inspectors from regulatory authorities.

Unless previously specified, the genetic data will have unknown clinical significance and AstraZeneca will not provide genetic results to participants, their family members, any insurance company, any employer, a clinical study investigator, a general physician, or any other third party, unless required to do so by law.

The participant's samples will not be used for any purpose other than those described in the study protocol.

A 5 Committees Structure

The safety of all AstraZeneca clinical studies is closely monitored on an on-going basis by AstraZeneca representatives in consultation with Patient Safety. Issues identified will be addressed; for instance, this could involve amendments to the CSP and letters to investigators.

A 6 Dissemination of Clinical Study Data

A description of this clinical study will be available on http://astrazenecaclinicaltrials.com and http://www.clinicaltrials.gov as will the summary of the study results when they are available.

The clinical study and/or summary of study results may also be available on other websites according to the regulations of the countries in which the study is conducted.

A 7 Data Quality Assurance

- All participant data relating to the study will be recorded on the CRF unless transmitted to the sponsor or designee electronically (e.g. laboratory data). The investigator is responsible for verifying that data entries are accurate and correct by physically or electronically signing the eCRF.
- The investigator must maintain accurate documentation (source data) that supports the information entered in the eCRF.
- The investigator must permit study-related monitoring, audits, IRB/IEC review, and regulatory authority inspections and provide direct access to source data documents.
- Monitoring details describing strategy (eg, risk-based initiatives in operations and quality such as Risk Management and Mitigation Strategies and Analytical Risk-Based Monitoring), methods, responsibilities and requirements, including handling of noncompliance issues and monitoring techniques (central, remote, or on-site monitoring) are provided in the Monitoring Plan.
- The sponsor or designee is responsible for the data management of this study including quality checking of the data.
- The sponsor assumes accountability for actions delegated to other individuals (e.g. Contract Research Organisations).
- Study monitors will perform ongoing source data verification to confirm that data entered into the eCRF by authorised site personnel are accurate, complete, and verifiable from source documents; that the safety and rights of participants are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.
- Records and documents, including signed ICFs, pertaining to the conduct of this study must be retained by the investigator for 15 years from the end of the study unless local regulations or institutional policies require a longer retention period. No records may be destroyed during the retention period without the written approval of the sponsor. No records may be transferred to another location or party without written notification to the sponsor.

A 8 Source Documents

- Source documents provide evidence for the existence of the participant and substantiate the integrity of the data collected. Source documents are filed at the investigator's site.
- Data reported on the eCRF or entered in the eCRF that are transcribed from source documents must be consistent with the source documents or the discrepancies must be

explained. The investigator may need to request previous medical records or transfer records, depending on the study. Also, current medical records must be available.

A 9 Study and Site Start and Closure

The study start date is the date on which the clinical study will be open for recruitment of participants.

The first act of recruitment is the first site open.

The sponsor designee reserves the right to close the study site or terminate the study at any time for any reason at the sole discretion of the sponsor. Study sites will be closed upon study completion. A study site is considered closed when all required documents and study supplies have been collected and a study-site closure visit has been performed.

The investigator may initiate study-site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the sponsor or investigator may include but are not limited to:

- Failure of the investigator to comply with the protocol, the requirements of the IRB/IEC or local health authorities, the sponsor's procedures, or GCP guidelines
- Inadequate recruitment of participants by the investigator
- Discontinuation of further study intervention development

If the study is prematurely terminated or suspended, the sponsor shall promptly inform the investigators, the IECs/IRBs, the regulatory authorities, and any Contract Research Organisation(s) used in the study of the reason for termination or suspension, as specified by the applicable regulatory requirements. The investigator shall promptly inform the participant and should assure appropriate participant therapy and/or follow-up.

Participants from terminated sites will have the opportunity to be transferred to another site to continue the study.

A 10 Publication Policy

- The results of this study may be published or presented at scientific meetings. If this is foreseen, the investigator agrees to submit all manuscripts or abstracts to the sponsor before submission. This allows the sponsor to protect proprietary information and to provide comments.
- The sponsor will comply with the requirements for publication of study results. In accordance with standard editorial and ethical practice, the sponsor will generally support

publication of multicentre studies only in their entirety and not as individual site data. In this case, a co-ordinating investigator will be designated by mutual agreement.

- Authorship will be determined by mutual agreement and in line with International Committee of Medical Journal Editors authorship requirements. Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work in Medical Journals, which states:
  - Authorship credit should be based on (1) substantial contributions to conception and design, acquisition of data, or analysis and interpretation of data; (2) drafting the article or revising it critically for important intellectual content; (3) final approval of the version to be published and (4) agreement to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. Authors should meet conditions 1, 2, and 3 and 4.
  - When a large, multicenter group has conducted the work, the group should identify the individuals who accept direct responsibility for the manuscript. These individuals should fully meet the criteria for authorship defined above.
  - Acquisition of funding, collection of data, or general supervision of the research group alone does not justify authorship.
  - All persons designated as authors should qualify for authorship, and all those who qualify should be listed.
  - Each author should have participated sufficiently in the work to take public responsibility for appropriate portions of the content.
- All publications (e.g., manuscripts, abstracts, oral/slide presentations, book chapters) based on this study must be submitted to AstraZeneca for corporate review. The vendor agreement will detail the procedures for, and timing of, AstraZeneca's review of publications.

Appendix B Handling of Human Biological Samples

B 1 Chain of Custody

A full chain of custody is maintained for all samples throughout their lifecycle.

The investigator at each centre keeps full traceability of collected biological samples from the participants while in storage at the centre until shipment or disposal (where appropriate) and records relevant processing information related to the samples whilst at site.

The sample receiver keeps full traceability of the samples while in storage and during use until used or disposed of or until further shipment and keeps record of receipt of arrival and onward shipment or disposal.

AstraZeneca or delegated representatives will keep oversight of the entire life cycle through internal procedures, monitoring of study sites, auditing or process checks, and contractual requirements of external laboratory providers

Samples retained for further use will be stored in the AstraZeneca-assigned biobanks or other sample archive facilities and will be tracked by the appropriate AstraZeneca Team during for the remainder of the sample life cycle. If required, AstraZeneca will ensure that remaining biological samples are returned to the site according to local regulations or at the end of the retention period, whichever is the sooner.

Withdrawal of Informed Consent for Donated Biological Samples AstraZeneca ensures that biological samples are returned to the source or destroyed at the end of the study as described in the informed consent.

If a participant withdraws consent specifically to the subsequent use of donated biological samples, the samples will be disposed of/destroyed/repatriated, and the action documented. If samples are already analysed, AstraZeneca is not obliged to destroy the results of this research. The participant will be presented with the option to opt out of the subsequent use of the donated samples during the withdrawal process. If the participant decides to opt out, then the donated samples will be disposed of. If the participant withdraws consent without opting out for the subsequent use of the donated samples, then the samples will be used as per protocol.

Following withdrawal of consent for biological samples, further study participation should be considered in relation to the withdrawal processes outlined in the informed consent.

The investigator:

- Ensures participant's withdrawal of informed consent to the use of donated samples is highlighted immediately to AstraZeneca or delegate.

- Ensures that relevant human biological samples from that participant, if stored at the study site, are immediately identified, disposed of as appropriate, and the action documented.
- Ensures that the participant and AstraZeneca are informed about the sample disposal.

AstraZeneca ensures the organisation(s) holding the samples is/are informed about the withdrawn consent immediately and that samples are disposed of or repatriated as appropriate, and the action documented and study site notified.

B 2 International Airline Transportation Association (IATA) 6.2 Guidance Document

LABELLING AND SHIPMENT OF BIOHAZARD SAMPLES

IATA (**https://www.iata.org/whatwedo/cargo/dgr/Pages/download.aspx**) classifies infectious substances into 3 categories: Category A, Category B or Exempt

Category A Infectious Substances are infectious substances in a form that, when exposure to it occurs, is capable of causing permanent disability, life-threatening or fatal disease in otherwise healthy humans or animals.
Category A pathogens are, for example, Ebola, Lassa fever virus. Infectious substances meeting these criteria which cause disease in humans or both in humans and animals must be assigned to UN 2814. Infectious substances which cause disease only in animals must be assigned to UN 2900.

Category B Infectious Substances are infectious Substances that do not meet the criteria for inclusion in Category A. Category B pathogens are, for example, Hepatitis A, C, D, and E viruses. They are assigned the following UN number and proper shipping name:

- UN 3373 – Biological Substance, Category B
- are to be packed in accordance with UN3373 and IATA 650

Exempt - Substances which do not contain infectious substances or substances which are unlikely to cause disease in humans or animals are not subject to these Regulations unless they meet the criteria for inclusion in another class.

- Clinical study samples will fall into Category B or exempt under IATA regulations.
- Clinical study samples will routinely be packed and transported at ambient temperature in IATA 650 compliant packaging. (https://www.iata.org/whatwedo/cargo/dgr/Documents/DGR-60-EN-PI650.pdf).
- Biological samples transported in dry-ice require additional dangerous goods specification for the dry-ice content.

Appendix C Genetic Sample

C 1 Use/Analysis of DNA

- The investigator at each centre intends to collect and store DNA for genetic research to explore how genetic variations may affect clinical parameters, risk and prognosis of diseases, and the response to medications. This genetic research may lead to better understanding of diseases, better diagnosis of diseases or other improvements in health care and to the discovery of new diagnostics, treatments or medications. Therefore, where local regulations and IRB/IEC allow, tissue and blood samples will be collected for DNA analysis from consenting participants.
- This genetic research may consist of the analysis of the structure of the participant's DNA, ie, the entire genome.
- The results of genetic analyses may be reported in a separate study summary.
- Each centre will store the DNA samples in a secure storage space with adequate measures to protect confidentiality.
- Samples will be destroyed following completion of the study.

C 2 Genetic Research Plan and Procedures

Selection of genetic research population

- All participants will be asked to participate in this genetic research. Participation is voluntary and if a participant declines to participate there will be no penalty or loss of benefit.

Inclusion criteria

For inclusion in this genetic research, participants must fulfil all of the inclusion criteria described in the main body of the CSP and: Provide informed consent for the Genomics Initiative sampling and analyses.

Exclusion criteria

- Exclusion from this genetic research may be for any of the exclusion criteria specified in the main study or any of the following:
  - Previous allogeneic bone marrow transplant.
  - Transfusion of non-leukocyte depleted blood or blood component within 120 days of genetic sample collection.
  - Healthy volunteers and paediatric participant samples will not be collected for the Genomics Initiative.

Withdrawal of consent for genetic research

Participants may withdraw from this genetic research at any time, independent of any decision concerning participation in the study. Procedures for withdrawal are outlined in Section 6.1 of the CSP.

Collection of samples for genetic research

The tissue and blood sample for this genetic research will be obtained from the participants at baseline(disease progression after 1L Osimertinib). Paired tissue and whole blood sample should be collected per participant for genetics during the study.

Coding and storage of DNA samples

- The processes adopted for the coding and storage of samples for genetic analysis are important to maintain participant confidentiality. Samples will be destroyed following completion of the study. DNA is a finite resource that is used up during analyses. Samples will be stored and used until no further analyses are possible.
- An additional second code will be assigned to the sample either before or at the time of DNA extraction replacing the information on the sample tube. Thereafter, the sample will be identifiable only by the second, unique number. This number is used to identify the sample and corresponding data at the AstraZeneca genetics laboratories, or at the designated organisation. No personal details identifying the individual will be available to any person (AstraZeneca employee or designated organisations working with the DNA).
- The link between the participant enrolment/randomisation code and the second number will be maintained and stored in a secure environment, with restricted access at AstraZeneca or designated organisations. The link will be used to identify the relevant DNA samples for analysis, facilitate correlation of genotypic results with clinical data, allow regulatory audit, and permit tracing of samples for destruction in the case of withdrawal of consent.

Ethical and regulatory requirements

The principles for ethical and regulatory requirements for the study, including this genetics research component, are outlined in Appendix A.

Informed consent

To participate in the genetic component of the study the participant must sign and date the consent form. Copies of signed and dated consent forms must be given to the participant and the original filed at the study centre. The principal investigator(s) is responsible for ensuring that consent is given freely and that the participant understands that they may freely withdrawal from the study at any time.

Participant data protection

- AstraZeneca will not provide individual genotype results to participants, any insurance company, any employer, their family members, general physician unless required to do so by law.

- Extra precautions are taken to preserve confidentiality and prevent genetic data being linked to the identity of the participant. In exceptional circumstances, however, certain individuals might see both the genetic data and the personal identifiers of a participant. For example, in the case of a medical emergency, an AstraZeneca physician or an investigator might know a participant's identity and also have access to his or her genetic data. Regulatory authorities may require access to the relevant files, though the participant's medical information and the genetic files would remain physically separate.

Data management

- Any genetic data generated in this study will be stored at a secure system at AstraZeneca and/or designated organisations to analyse the samples.
- AstraZeneca and its designated organisations may share summary results from this genetic research with other researchers, such as hospitals, academic organisations or health insurance companies. This can be done by placing the results in scientific databases, where they can be combined with the results of similar studies to learn even more about health and disease. The researchers can only use this information for health-related research purposes. Researchers may see summary results but they will not be able to see individual participant data or any personal identifiers.
- Some or all of the clinical datasets from the study may be merged with the genetic data in a suitable secure environment separate from the clinical database.

Appendix D Guidelines for Evaluation of Objective Tumour Response Using RECIST 1.1 Criteria (Response Evaluation Criteria in Solid Tumors)

Introduction

This appendix details the implementation of RECIST 1.1 guidelines (Eisenhauer et al 2009). Investigator assessments will use the RECIST 1.1 guidelines described in this appendix.

Imaging modalities and acquisition specifications for RECIST 1.1

A summary of the imaging modalities that can be used for tumour assessment of TLs, NTLs and NLs is provided in Table .

Table 4 **Summary of Imaging Modalities for Tumour Assessment**

Target Lesions	Non-Target Lesions	New Lesions
CT MRI	CT MRI Plain X-ray Chest X-ray	CT MRI Plain X-ray Chest X-ray Bone scan (Scintigraphy) ^{18}F-fluoro-deoxyglucose-PET/CT

CT=computed tomography; PET/CT=positron emission tomography/CT; MRI=magnetic resonance imaging.

Computed Tomography and Magnetic Resonance Imaging

CT with IV contrast is the preferred imaging modality (although MRI with IV contrast is acceptable if CT is contraindicated) to generate reproducible anatomical images for tumour assessments (ie, for measurement of TLs, assessment of NTLs, and identification of NLs). It is essential that the same correct imaging modality, image acquisition parameters (e.g. anatomic coverage, imaging sequences, etc), imaging facility, tumour assessor (e.g. radiologist), and method of tumour assessment (e.g. RECIST 1.1) are used consistently for each participant throughout the study. The use of the same scanner for serial scans is recommended, if possible. It is important to follow the image collection/tumour assessment schedule as closely as possible (refer to the SoA), and this on-study imaging schedule MUST be followed regardless of any delays in dosing or missed imaging visits. If an unscheduled assessment is performed (eg, to investigate clinical signs/symptoms of progression) and the participant has not progressed, every attempt should be made to perform the subsequent scan acquisitions at the next scheduled imaging visit.

Due to its inherent rapid acquisition (seconds), CT is the imaging modality of choice. Body scans should be performed with breath-hold scanning techniques, if possible. Therefore, CT of the chest is recommended over MRI due to significant motion artefacts (e.g. heart, major blood vessels, breathing) associated with MRI. MRI has excellent contrast and spatial and temporal

resolutions; however, there are many image acquisition variables involved in MRI, which greatly impact image quality, lesion conspicuity, and measurement. Furthermore, the availability of MRI is variable globally. The modality used at follow-up should be the same as was used at baseline, and the lesions should be measured/assessed on the same pulse sequence. In general, local oncology diagnostic imaging parameters are applied for scan acquisition. It is beyond the scope of this appendix to prescribe specific MRI pulse sequence parameters for all scanners, body parts, and diseases.

The most critical CT and MRI image acquisition parameters for optimal tumour evaluation are anatomic coverage, contrast administration, slice thickness, and reconstruction interval.

a. Anatomic coverage: Optimal anatomic coverage for most solid tumours is the chest-abdomen (-pelvis). Coverage should encompass all areas of known predilection for metastases in the disease under evaluation and should additionally investigate areas that may be involved based on signs and symptoms of individual participants. Because a lesion later identified in a body part not scanned at baseline would be considered as a NL representing PD, careful consideration should be given to the extent of imaging coverage at baseline and at subsequent follow-up time points. This will enable better consistency not only of tumour measurements but also identification of new disease.

Required anatomical regions to be imaged for assessment of tumour burden (TLs and/or NTLs) at baseline and follow-up visits vary according to the study, and these time points are specified in the SoA. Examples include the following:

- IV contrast-enhanced CT of chest-abdomen (including the entire liver and both adrenal glands) (-pelvis).
- Non-contrast CT of chest and IV contrast-enhanced abdomen (including the entire liver and both adrenal glands) (-pelvis).
- IV contrast-enhanced CT or MRI of the head and neck.
- IV contrast-enhanced MRI (preferred) or CT of the brain.

For chest-abdomen (-pelvis) imaging, the following are scanning options in decreasing order of preference, with additional options (2 to 4) for consideration when participants have sensitivity to IV contrast or have compromised renal function:

1 Chest-abdomen (-pelvis) CT with IV CT contrast (most preferred).
2 Chest CT without IV-contrast + abdomen (-pelvis) MRI with IV MRI contrast, if CT IV contrast (iodine based) is medically contraindicated at any time during the study.
3 Chest-abdomen (-pelvis) CT without IV contrast, if both IV CT and MRI contrast are medically contraindicated or the participant has compromised renal function.

4 Chest-abdomen (-pelvis) MRI with IV MRI contrast, if CT cannot be performed at any time during the study.

b. IV contrast administration: Optimal visualisation and measurement of metastases in solid tumours require consistent administration (dose and rate) of IV contrast as well as timing of scanning. An adequate volume of a suitable contrast agent should be given so that the tumour lesions are demonstrated to best effect and a consistent method is used on subsequent examinations for any given participant. Oral contrast is recommended to help visualise and differentiate structures in the abdomen and pelvis.

c. Slice thickness and reconstruction interval: It is recommended that CT or MRI scans be acquired/reconstructed as contiguous (no gap) slices with ≤5 mm thickness throughout the entire anatomic region of interest for optimal lesion measurements. Exceptionally, particular institutions may perform medically acceptable scans at slice thicknesses >5 mm. If this occurs, the minimum size of measurable lesions at baseline should be twice the slice thickness of the baseline scans.

For CT scans, all window settings should be included in the assessment, particularly in the thorax where lung and soft tissue windows should be considered. When measuring lesions, the TL should be measured on the same window setting for repeated examinations throughout the study.

Chest X-ray

Chest X-ray assessment will not be used for the assessment of TLs. Chest X-ray can, however, be used to assess NTLs and to identify the presence of NLs. However, there is preference that a higher resolution modality, such as CT, be used to confirm the presence of NLs.

Plain X-ray

Plain X-ray may be used as a method of assessment for bone NTLs and to identify the presence of new bone lesions.

Isotopic bone scan

Bone lesions identified on an isotopic bone scan at baseline and confirmed by CT, MRI, or X-ray at baseline should be recorded as NTLs and followed by the same method per baseline assessment (CT, MRI, or X-ray).

Isotopic bone scans may be used as a method of assessment to identify the presence of new bone lesions at follow-up visits. NLs may be recorded in case positive hot-spots appear on a bone scan that were not present on a previous bone scan; however, a newly observed equivocal hot-spot on a bone scan that cannot be verified with correlative imaging (CT, MRI, or X-ray) of the same anatomical region shall not be the only trigger for a PD assessment at that time point.

^{18}F-Fluoro-deoxyglucose-PET/CT

^{18}F-fluoro-deoxyglucose positron emission tomography(PET)/CT scans may be used as a method for identifying new extrahepatic lesions (but not intrahepatic lesions) for RECIST 1.1 assessments according to the following algorithm: NLs will be recorded where there is positive ^{18}F-Fluoro-deoxyglucose uptake[1] not present on baseline or prior ^{18}F-fluoro-deoxyglucose-PET scan or in a location corresponding to a NL on a companion CT/MRI collected close in time to the ^{18}F-fluoro-deoxyglucose-PET scan. The PET portion of the PET/CT introduces additional data that may bias an investigator if it is not routinely or serially performed. Therefore, if there is no baseline or prior ^{18}F-fluoro-deoxyglucose-PET scan available for comparison, and no evidence of NLs on companion CT/MRI scans, then follow-up CT/MRI assessments should continue as per the regular imaging schedule to verify the unequivocal presence of NLs.

At present, low-dose or attenuation correction CT portions of a combined ^{18}F-fluoro-deoxyglucose-PET/CT scan are of limited use in anatomically based efficacy assessments, and it is therefore suggested that they should not substitute for dedicated diagnostic contrast-enhanced CT scans for tumour measurements by RECIST 1.1. In exceptional situations, if a site can document that the CT performed, as part of a PET/CT examination, is of identical diagnostic quality (with IV contrast) to a dedicated diagnostic CT scan, then the CT portion of the PET/CT can be used for RECIST 1.1 tumour assessments. Caution that this is not recommended because the PET portion of the CT introduces additional (PET) data that may bias an investigator if it is not routinely or serially performed.

Ultrasound

Ultrasound examination will not be used for RECIST 1.1 assessment of tumours as it is not a reproducible acquisition method (operator dependent), is subjective in interpretation, and may not provide an accurate assessment of the true tumour size. Tumours identified by ultrasound will need to be assessed by correlative CT or MRI anatomical scan.

Other tumour assessments

Clinical examination

Clinical examination of skin/surface lesions (by visual inspection or manual palpation) will not be used for RECIST 1.1 assessments. Tumours identified by clinical examination will need to be assessed by correlative CT or MRI anatomical scans.

Endoscopy and laparoscopy

Endoscopy and laparoscopy will not be used for tumour assessments as they are not validated in the context of tumour assessment.

1 A positive ^{18}F-fluoro-deoxyglucose-PET scan lesion should be reported only when an uptake (eg, standard uptake value) greater than twice that of the surrounding tissue or liver is observed.

Histology and cytology

Histology or tumour markers on tumour biopsy samples will not be used as part of the tumour response assessment as per RECIST 1.1.

Results of cytological examination for the neoplastic origin of any effusion (eg, ascites, pericardial effusion, and pleural effusion) that appears or worsens during the study will not be used as part of the tumour response assessment as per RECIST 1.1.

Furthermore, an overall assessment of CR (all other disease disappears/reverts to normal) would be changed to PR if an effusion remains present radiologically.

Measurability of tumour lesions at baseline

RECIST 1.1 measurable lesions at baseline

A tumour lesion that can be accurately measured at baseline as ≥10 mm in the longest diameter for non-nodal lesions or ≥15 mm in short axis[2] diameter for lymph node lesions with IV contrast-enhanced CT or MRI and that is suitable for accurate repeated measurements. Please see additional RECIST 1.1 guidance below on measurability of intrahepatic hepatocellular carcinoma lesions and porta hepatis lymph nodes.

Non-measurable lesions at baseline

- Truly non-measurable lesions include the following:
  - Bone lesions (see exception below for soft tissue component).
  - Leptomeningeal disease.
  - Ascites, pleural effusion, or pericardial effusion.
  - Inflammatory breast disease.
  - Lymphangitic involvement of skin or lung.
- All other lesions, including small lesions (longest diameter <10 mm or pathological lymph nodes with ≥10 mm to <15 mm short axis diameter at baseline).[3]
- Previously irradiated lesions.[4]
- Brain metastasis.

Special considerations regarding lesion measurability at baseline

- Bone lesions:

[2] The short axis is defined as the longest in-plane axis perpendicular to the long axis.
[3] Lymph nodes with <10 mm short axis diameter are considered non-pathological and should not be recorded or followed as NTLs.
[4] Lymph nodes with <10 mm short axis diameter are considered non-pathological and should not be recorded or followed as NTLs.

  - Bone scan, PET scan, or plain X-ray are not considered adequate imaging techniques to measure bone lesions; however, these techniques can be used to confirm the presence or disappearance of bone lesions.
  - Lytic bone lesions or mixed lytic-blastic lesions, with identifiable soft tissue components, can be considered measurable if the soft tissue component meets the definition of measurability.
  - Blastic lesions are considered non-measurable.
- Cystic lesions thought to represent cystic metastases can be considered measurable lesions if they meet the criteria for measurability from a radiological point of view, but if non-cystic lesions are present in the same participant, these should be selected over cystic lesions as TLs.

RECIST 1.1 TL selection at baseline

A maximum of 5 measurable lesions, with a maximum of 2 lesions per organ (including lymph nodes collectively considered as a single organ), representative of all lesions involved should be identified as TLs at baseline. TLs should be selected on the basis of their size (longest diameter for non-nodal lesions or short axis diameter for nodal lesions), but in addition should be those that lend themselves to reproducible repeated measurements. It may be the case that, on occasion, the largest lesion does not lend itself to reproducible measurement, in which circumstance the next largest lesion that can be measured reproducibly should be selected.

Lymph nodes, in any location (local/regional and distant), are collectively considered as a single organ, with a maximum of 2 lymph nodes as TLs. A bilateral organ (e.g. adrenal glands), a segmented organ (e.g. liver), or a multilobed organ (e.g. lung) is each considered as a single organ.

The site and location of each TL should be documented, as well as the longest axis diameter for non-nodal lesions (or short axis diameter for lymph nodes). All measurements should be recorded in millimetres. At baseline, the sum of the diameters for all TLs will be calculated and reported as the baseline sum of diameters. At follow-up visits, the sum of diameters for all TLs will be calculated and reported as the follow-up sum of diameters.

Special cases for TL assessment at baseline

- For TLs measurable in 2 or 3 dimensions, always report the longest diameter. For pathological lymph nodes measurable in 2 or 3 dimensions, always report the short axis diameter.
- When lymph nodes are coalesced and no longer separable in a conglomerate mass, the vector of the longest diameter should be used to determine the perpendicular vector for the maximal short axis diameter of the coalesced mass. Non-nodal lesions that coalesce should similarly be assessed by the longest axis diameter.

- Tumour lesions selected for newly acquired screening biopsy should not be selected as TLs, unless imaging occurred at least approximately 2 weeks after biopsy, allowing time for healing.
- If the CT/MRI slice thickness used is >5 mm, the minimum size of measurable disease at baseline should be twice the slice thickness of the baseline scan.
- If a lesion has completely disappeared, the diameter should be recorded as 0 mm. If a lesion appears in the same location on a subsequent scan, it will be recorded as a NL.

RECIST 1.1 NTL selection at baseline

All other lesions, including non-measurable lesions and surplus measurable lesions, not recorded as TLs should be identified as NTLs at baseline. Measurements of these lesions are not required, but the presence or absence of each should be noted throughout follow-up.

Evaluation of tumour response and progression

RECIST 1.1 TL assessment at follow-up

This section defines the criteria used to determine objective tumour visit response for RECIST 1.1-defined TLs. The imaging modality, location, and scan date of each TL identified previously at baseline should be documented at follow-up visits with the long axis diameter for non-nodal lesions or short axis diameter for lymph node lesions. All measurements should be recorded in millimetres. The sum of the diameters for all TLs at each follow-up visit will be compared with the baseline sum of diameters (for response or SD) or to the smallest prior (nadir) sum of diameters (for progression).

Special cases for TL assessment at follow-up:

- If a lesion has completely disappeared, the diameter should be recorded as 0 mm. If a lesion appears in the same location on a subsequent scan, it will be recorded as an NL.
- If a TL splits into 2 or more parts, the sum of the diameters of those parts should be recorded.
- If 2 or more TLs merge, then the sum of the diameters of the combined lesion should be recorded for 1 of the lesions and 0 mm recorded for the other lesion(s). If the merged TLs are non-nodal lesions, record the long axis diameter of the merged lesion. If pathologic lymph nodes coalesce and are no longer individually separable within a conglomerate mass, the vector of the longest diameter of the coalesced mass should be used to determine the perpendicular vector for the maximal short axis diameter.
- If a TL is believed to be present and is faintly seen but too small to measure, a default value of 5 mm should be assigned. If an accurate measure can be given, this should be recorded, even if it is below 5 mm.

- If a TL cannot be measured accurately due to it being too large, provide an estimate of the size of the lesion. The choice of "Too large to measure" in the CRF will trigger an overall visit response of PD.
- When a TL has had any intervention (e.g. definitive radiotherapy, embolization, surgery, transarterial chemoembolization, etc) during the study, the size of the TL should still be provided where possible and the intervention recorded in the RECIST 1.1 CRF for the current imaging visit and all subsequent visits. If a TL has been completely removed (surgery) or disappears, the longest diameter should be recorded as 0 mm.

Table 5 **RECIST 1.1 Evaluation of Target Lesions**

CR	Disappearance of all TLs since baseline. Any pathological lymph nodes selected as TLs must have a reduction in short axis diameter to <10 mm.
PR	At least a 30% decrease in the sum of the diameters of TL, taking as reference the baseline sum of diameters.
SD	Neither sufficient decrease in the sum of diameters to qualify for PR nor sufficient increase to qualify for PD.
PD	At least a 20% increase in the sum of diameters of TLs, taking as reference the smallest previous sum of diameters (nadir). This includes the baseline sum if that is the smallest on study. In addition to the relative increase of 20%, the sum must demonstrate an absolute increase of at least 5 mm from nadir.
NE	Only relevant if any of the TLs at follow-up were not assessed or NE (eg, missing anatomy) or had a lesion intervention at this visit. Note: If the sum of diameters meets the PD criteria, PD overrides NE as a TL response.
Not applicable	Only relevant if no TLs present at baseline.

CR=complete response; NE=not evaluable; PD=progression of disease; PR=partial response; SD=stable disease; TL=target lesion.

RECIST 1.1 NTL assessment at follow-up

All other lesions (or sites of disease) not recorded as TLs should be identified as NTLs at baseline. Measurements are not required for these lesions, but their status should be followed at subsequent visits. At each visit, an overall assessment of the NTL response should be recorded by the investigator.

To achieve "unequivocal progression" on the basis of NTLs, there must be an overall level of substantial worsening in non-target disease such that, even in presence of SD or PR in TLs, the overall tumour burden has increased sufficiently to merit unequivocal progression by NTLs. A modest "increase" in the size of 1 or more NTLs is usually not sufficient to qualify for unequivocal progression status. The designation of overall progression solely on the basis of change in non-target disease in the face of SD or PD of target disease will therefore be extremely rare.

Table 6 **RECIST 1.1 Evaluation of Non-Target Lesions**

CR	Disappearance of all NTLs since baseline. All lymph nodes must be non-pathological in size (<10 mm short axis).
Non CR/non PD	Persistence of 1 or more NTLs.
PD	Unequivocal progression of existing NTLs. Unequivocal progression may be due to an important progression in 1 lesion only or in several lesions. In all cases, the progression MUST be clinically significant for the physician to consider changing (or stopping) therapy.
NE	Only relevant when 1 or some of the NTLs were not assessed and, in the investigator's opinion, they are not able to provide an evaluable overall NTL assessment at this visit. Note: For participants without TLs at baseline, this is relevant if any of the NTLs were not assessed at this visit and the progression criteria have not been met.
Not applicable	Only relevant if no NTLs present at baseline.

CR=complete response; NE=not evaluable; NTL=non-target lesion; PD=progression of disease; TL=target lesion.

RECIST 1.1 NL identification at follow-up

Details, including the imaging modality, the date of scan, and the location of any NLs will also be recorded in the CRF. The presence of 1 or more NLs is assessed as progression. The finding of a NL should be unequivocal, ie, not attributable to differences in scanning technique, change in imaging modality, or findings thought to represent something other than tumour. If a NL is equivocal, for example because of its small size, the treatment and tumour assessments should be continued until the previously (pre-existing) NL has been assessed as unequivocal at a follow-up visit, and then the progression date should be declared using the date of the initial scan when the NL first appeared.

A lesion identified at a follow-up assessment in an anatomical location that was not scanned at baseline is considered a NL and will indicate PD.

RECIST 1.1 evaluation of overall visit response at follow-up

Derivation of overall visit response as a result of the combined assessment of TLs, NTLs, and NLs uses the algorithm shown in Table .

Table 7 **RECIST 1.1 Overall Visit Response**

Target Lesions	Non-Target Lesions	New Lesions	Overall Visit Response
CR	CR	No	CR
CR	NA	No	CR
NA	CR	No	CR
CR	Non CR/Non PD	No	PR
CR	NE	No	PR

Target Lesions	Non-Target Lesions	New Lesions	Overall Visit Response
PR	Non PD or NE or NA	No	PR
SD	Non PD or NE or NA	No	SD
NA	Non-CR/Non-PD	No	SD (non-CR/non-PD)
NE	Non PD or NE	No	NE
NA	NE	No	NE
NA	NA	No	NED
PD	Any	Yes or No	PD
Any	PD	Yes or No	PD
Any	Any	Yes	PD

Non-CR/Non-PD for overall response if only NTL (no TLs) are present at baseline.

Note: An overall assessment of CR (all other disease disappears/reverts to normal) would be changed to PR if ascites remains present radiologically.

CR=complete response; NA=not applicable (only relevant if there were no TLs at baseline or NTLs at baseline), NE=not evaluable; NED=no evidence of diseases (only relevant if there were neither target lesions nor non-target lesions at baseline); NTL=non-target lesion; PD=progression of disease; PR=partial response; SD=stable disease; TL=target lesion.

The following overall visit responses are possible depending on the extent of tumour disease at baseline:

- For participants with TLs (at baseline): CR, PR, SD, PD, or NE.
- For participants with NTLs only (at baseline): CR, Non-CR/Non-PD, PD, or NE.
- For participants with no disease at baseline: no evidence of disease (available as an option in the eCRF), PD, or NE.

References

Eisenhauer et al 2009

Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). Eur J Cancer 2009;45(2):228-47.

Appendix E Contraception Requirements

Contraception requirements for this study are as follows.

E 1 Female Participants

Women of Childbearing Potential (WoCBP) are defined as women between menarche and menopause who have not been permanently or surgically sterilized and are capable of procreation.

Women not of childbearing potential are defined as those who are surgically sterile (ie, bilateral salpingectomy, bilateral oophorectomy, or complete hysterectomy) or who are post-menopausal.

Women will be considered post-menopausal if they have been amenorrhoeic for 12 months without an alternative medical cause. The following age-specific requirements apply:

- Women <50 years of age would be considered post-menopausal if they have been amenorrhoeic for 12 months or more following cessation of all hormonal replacement therapy and if they have luteinizing hormone and follicle-stimulating hormone levels in the post-menopausal range for the institution.
- Women ≥50 years of age would be considered post-menopausal if they have been amenorrhoeic for 12 months or more following cessation of all hormonal replacement therapy, or had radiation-induced menopause with last menses >1 year ago, or had chemotherapy-induced menopause with last menses >1 year ago.

Women of childbearing potential who are not totally sexually abstinent (ie, refraining from heterosexual intercourse during the entire period of risk associated with study interventions) and intend to be sexually active with a nonsterilised male partner must use at least 1 highly effective method of contraception (Table). They should have been stable on their chosen method of birth control for a minimum of 3 months before entering the study and continue to use it throughout the study.

Non-sterilised male partners of a woman of childbearing potential must use a male condom plus spermicide (condom alone in countries where spermicides are not approved) throughout this period. Periodic abstinence, the rhythm method, and the withdrawal method are not acceptable methods of contraception. Total sexual abstinence is an acceptable method provided it is the usual lifestyle of the participant. Female participants should refrain from breastfeeding throughout this period.

E 2 Male Participants with a Female Partner of Childbearing Potential

Non-sterilised male participants (including males sterilised by a method other than bilateral orchidectomy, e.g. vasectomy) who intend to be sexually active with a female partner of

childbearing potential must be using an acceptable method of contraception such as male condom plus spermicide (condom alone in countries where spermicides are not approved) from the time of screening throughout the completion of the study to prevent pregnancy in a partner.

Periodic abstinence, the rhythm method, and the withdrawal method are not acceptable methods of contraception. Male participants should refrain from sperm donation or banking throughout this period.

Vasectomised (ie, sterile) males are considered fertile and should still use a male condom plus spermicide as indicated above during the clinical study.

Even if the female partner is pregnant, male participants should still use a condom plus spermicide, as indicated above during the clinical study, if there is a concern about damaging the developing foetus from drug in ejaculate.

Female partners (of childbearing potential) of male participants must also use a highly effective method of contraception throughout this period (Table).

E 3 Highly Effective Methods of Contraception

Highly effective methods of contraception, defined as one that results in a low failure rate (ie, less than 1% per year) when used consistently and correctly, are described in Table . Note that some contraception methods are not considered highly effective (eg, male or female condom with or without spermicide; female cap, diaphragm, or sponge with or without spermicide; non copper containing intrauterine device; progestogen-only oral hormonal contraceptive pills where inhibition of ovulation is not the primary mode of action [excluding Cerazette/desogestrel which is considered highly effective]; and triphasic combined oral contraceptive pills).

Table 8 Highly Effective Methods of Contraception (<1% Failure Rate)

Non-Hormonal Methods	Hormonal Methods
• Total sexual abstinence (evaluate in relation to the duration of the clinical study and the preferred and usual lifestyle choice of the participant) • Vasectomised sexual partner (with participant assurance that partner received post-vasectomy confirmation of azoospermia) • Tubal occlusion • Intrauterine device (provided coils are copper-banded)	• Injection: Medroxyprogesterone injection (e.g. Depo-Provera®)[a] • Levonorgestrel-releasing intrauterine system (e.g. Mirena®)[a] • Implants: Etonogestrel-releasing implants (e.g. Implanon® or Norplant®) • Intravaginal devices: Ethinylestradiol/etonogestrel-releasing intravaginal devices (e.g. NuvaRing®) • Combined pill: Normal and low dose combined oral contraceptive pill • Patch: Norelgestromin/ethinylestradiol-releasing transdermal system (e.g. Ortho Evra®) • Mini pill: Progesterone based oral contraceptive pill using desogestrel: Cerazette® is currently the only highly effective progesterone-based pill

[a] Hormonal methods not prone to drug-drug interactions.

Appendix F NGS technology, FISH technology, ddPCR technology and sample requirements

NGS Company: Geneplus-Beijing Clinical Laboratory Co.,Ltd

Tissue Sample requirements

FFPE: 10 slides, 5mm length * 5mm width * 5um thickness;

Tumor content: ≥20%

De-duplicate sequencing depth: ≥500x

Table 9 Validation information of tumor tissue sample detection

Sample type	Tissue		
De-duplicate sequencing depth	≥500x		
Accuracy	PPA>95%; PPV >95%； NPA>95%		
Sensitivity	Mutation type	Lod of Hot spot （Limit of Detection）	Lod of Non hot spot （Limit of Detection）
	SNV	≥2.0%	≥5.0%
	InDel	≥2.0%	≥5.0%
	SV(Fusion)	≥2.0%	≥5.0%
	CNV	≥3.8 copies	
Specificity	>95%		
Repeatability	100%		

Plasma Sample requirements

Whole blood: 10ml

De-duplicate sequencing depth: ≥1000x

Table 10 Validation information of plasma sample detection

Sample type	Plasma		
De-duplicate sequencing depth	≥1000x		
Accuracy	PPA>95%; PPV >95%； NPA>95%		
Sensitivity	Mutation type	Lod of Hot spot (Limit of Detection)	Lod of Non hot spot (Limit of Detection)
	SNV	≥0.5%	≥1.0%
	InDel	≥0.5%	≥1.0%
	SV(Fusion)	≥0.5%	≥1.0%
	CNV	≥3.2 copies	
Specificity	>95%		
Repeatability	100%		

Table 11 Panel list of NGS

ABL1	FANCC	NFKBIA	TP53	CBR1	EIF4A2	HIST1H2AC	MCC	PLCG2	SMC1B	XPC
ACVR1B	FANCD2	NKX2-1	TSC1	CBR3	EIF4G3	HIST1H2AG	MCM3	PLK1	SNCAIP	XRCC1
AKT1	FANCE	NOTCH1	TSC2	CCDC168	ELAC2	HIST1H2AL	MDC1	PLXNA1	SNTG1	XRCC3
AKT2	FANCF	NOTCH2	TSHR	CCNA1	ELF1	HIST1H2AM	MECOM	PLXNB2	SNX29	YAP1

AKT3	FANCG	NOTCH3	U2AF1	CCNB3	ELF3	HIST1H2BC	MEF2C	PNRC1	SOD2	YY1AP1
ALK	FANCL	NPM1	VEGFA	CCT3	ELMO1	HIST1H2BD	MGA	POLQ	SOS1	ZBTB16
APC	FANCM	NRAS	VHL	CCT5	ELN	HIST1H2BJ	MIB1	POM121	SOX10	ZC3H11A
AR	FAS	NSD1	WRN	CCT6B	EME2	HIST1H2BK	MIOS	POM121L12	SOX17	ZFHX3
ARAF	FAT1	NTHL1	WT1	CD22	EMID2	HIST1H2BO	MKL1	POU2AF1	SPEN	ZFP36L1
ARID1A	FAT2	NTRK1	XPO1	CD33	EML4	HIST1H3B	MLL4	PPM1D	SPRR3	ZFP36L2
ARID1B	FBXW7	NTRK2	XRCC2	CD5L	EPC1	HIST1H3C	MLLT3	PPP1R17	SPSB4	ZFPM2
ARID2	FGF19	NTRK3	ZMAT3	CD74	EPHA1	HIST1H3D	MMP11	PPP6C	SPTA1	ZIC3
ASXL1	FGF3	PALB2	ABCA13	CDA	EPHA4	HIST1H3F	MMP2	PRDM16	SRD5A2	ZNF217
ATM	FGF4	PARK2	ABCB1	CDH11	EPHA7	HIST1H3G	MN1	PREX2	SRGAP1	ZNF384
ATR	FGFR1	PARP1	ABCC1	CDH18	EPHB2	HIST1H3H	MNDA	PRF1	SRGAP3	ZNF521
ATRX	FGFR2	PAX5	ABCC11	CDH23	EPHB4	HIST1H3I	MNX1	PRKAA1	SRSF2	ZNF638
AURKA	FGFR3	PBRM1	ABCC2	CDK13	EPOR	HIST1H4I	MSH4	PRKCB	SRSF7	ZNF750
AURKB	FGFR4	PCK1	ABCG2	CHD1	EPPK1	HIST3H3	MSN	PRKCI	STAG1	ZNF804B
AXIN1	FH	PDCD1	ABL2	CHD1L	EPS15	HLF	MSR1	PRKDC	STAT1	HLA-A
AXIN2	FLCN	PDCD1LG2	ACACA	CHD4	ERBB2IP	HMCN1	MTHFR	PRRX1	SUCLG1	HLA-B
AXL	FLT1	PDGFRA	ACIN1	CHD6	ERCC2	HNF1B	MTRR	PRX	SUCLG2	HLA-C
B2M	FLT3	PDGFRB	ACTB	CHD8	ESR2	HNRPDL	MUC5B	PSG2	SULT1A1	
BAP1	FLT4	PDK1	ACTG1	CHD9	ETS1	HOXA11	MYH11	PSIP1	SUZ12	
BARD1	FOXA1	PIK3CA	ACTG2	CHFR	ETV1	HOXA13	MYH14	PSMB1	SVEP1	
BCL2	FOXL2	PIK3CB	ACVR2A	CHI3L1	ETV5	HOXA3	MYH9	PSMB5	SYNCRIP	
BCL2L1	FOXP1	PIK3CG	ACVRL1	CHN1	ETV6	HOXA9	MYO3A	PTGS1	SYNE1	
BCOR	FUBP1	PIK3R1	ADAM29	CIITA	EWSR1	HOXC13	MYOD1	PTGS2	TAF1	
BLM	GALNT12	PIK3R2	ADAMTS5	CLDN18	EZR	HOXD11	NAP1L1	PTPN13	TAF15	
BMPR1A	GATA3	PMS1	ADCY1	CLP1	F8	HOXD13	NAV3	PTPN2	TAF1L	
BRAF	GNA11	PMS2	AFF1	CLSPN	FAM131B	HSD3B1	NCAM2	PTPRB	TAL1	

BRCA1	GNAQ	POLD1	AFF2	CLTC	FAM135B	HSP90AA1	NCF2	PTPRK	TBL1XR1
BRCA2	GNAS	POLE	AFF3	CNOT3	FAM157B	HSP90AB1	NCF4	PTPRO	TBX15
BRD4	GRIN2A	POT1	AHNAK	CNOT4	FAM46C	HSPA8	NCK1	PTPRS	TBX22
BRIP1	GRM3	PPP2R1A	AKAP9	CNTN1	FAM5C	HSPD1	NCOA3	PTPRT	TCEB1
BTK	HDAC1	PRDM1	ALB	CNTN5	FAP	HSPH1	NCOA4	PTPRU	TCF12
CARD11	HGF	PRKAR1A	AMOT	CNTNAP1	FASLG	ICK	NCOR2	RAB35	TCF3
CASP8	HNF1A	PTCH1	ANGPT1	CNTNAP5	FAT3	ICOSLG	NCSTN	RAC2	TCF4
CBFB	HOXB13	PTCH2	ANK3	COL1A1	FAT4	ID3	NDUFA13	RAD21	TCL1A
CBL	HRAS	PTEN	ANKRD11	COL2A1	FCGR1A	IFITM3	NFATC4	RAD54B	TEC
CCND1	IDH1	PTPN11	ANKRD30A	COL5A1	FCGR2A	IGF1	NFE2L3	RANBP2	TENM3
CCND2	IDH2	PTPRD	ANKRD30B	COL5A2	FCGR2B	IGF2	NKX3-1	RASA1	TERT
CCND3	IFNG	RAC1	APEX1	COL5A3	FCGR3A	IGF2R	NLRC3	RASGRP1	TET1
CCNE1	IFNGR1	RAD50	APOBEC3B	COPS2	FCRL4	IGLL5	NOD1	RBL1	TFDP1
CD274	IGF1R	RAD51	ARAP3	CPS1	FGF10	IKZF2	NOS3	REL	TFDP2
CDC73	IKBKE	RAD51B	ARFGEF1	CRIPAK	FGF12	IKZF3	NOTCH4	RELN	TFE3
CDH1	IKZF1	RAD51C	ARFGEF2	CRLF2	FGF14	IL10	NQO1	RFC1	TGFBR1
CDK12	IL7R	RAD51D	ARHGAP29	CRNKL1	FGF23	IL1RAPL1	NR1I2	RGS3	THBS2
CDK4	INPP4B	RAD52	ARHGAP35	CRTC1	FGF6	IL21R	NR2F2	RHEB	TJP1
CDK6	IRF2	RAD54L	ARID4B	CSF1	FLG	IL6	NR4A2	RHOH	TLE1
CDK8	IRS2	RAF1	ARID5B	CSF3R	FLI1	IL6ST	NRG1	RHOT1	TLL2
CDKN1A	JAK1	RARA	ARNT	CSMD1	FLNC	IMPG1	NRP2	RIT1	TLR4
CDKN1B	JAK2	RB1	ASCL4	CSMD3	FMN2	ING1	NRXN1	RNASEL	TLX3
CDKN2A	JAK3	RBM10	ASH1L	CSNK1A1	FN1	INHBA	NTM	ROBO1	TMEM132D
CDKN2B	JUN	RECQL	ASMTL	CSNK1G3	FNDC4	INPP4A	NUMA1	ROBO2	TNFSF11
CDKN2C	KDM5A	RECQL4	ASPM	CTLA4	FOXA2	INPPL1	NUP107	ROBO3	TNN
CEBPA	KDM5C	RET	ASTN1	CTNNA2	FOXO1	INSR	NUP210	ROCK1	TP53BP1

CHEK1	KDM6A	RHOA	ASXL2	CTNND1	FOXO3	IRF4	NUP93	RPGR	TP63
CHEK2	KDR	RICTOR	ATIC	CUX1	FOXQ1	IRF6	NUP98	RPS6KB1	TP73
CIC	KEAP1	RINT1	ATP11B	CXCR4	FRMPD4	IRS1	OBSCN	RPS6KB2	TPM3
CREBBP	KIT	RNF43	ATP12A	CYBA	FUS	ITGB3	OGDH	RSPO2	TPR
CRKL	KRAS	ROS1	ATP1A1	CYP19A1	FXR1	ITK	OMD	RSPO3	TRAF2
CSF1R	LRP1B	RPTOR	ATP2B3	CYP1A1	FYN	ITSN1	OPCML	RUNX1T1	TRAF7
CTCF	MAF	RUNX1	BAZ2B	CYP1B1	FZD1	JARID2	OR11G2	RUNX2	TRIM24
CTNNA1	MAP2K1	SDHA	BBC3	CYP2A13	G3BP1	KALRN	OR2T4	RXRA	TRIM58
CTNNB1	MAP2K2	SDHAF2	BBS9	CYP2C8	G3BP2	KAT6A	OR4A15	RYR1	TRIO
CUL3	MAP2K4	SDHB	BCAS1	CYP2D6	GAB2	KAT6B	OR4C6	RYR2	TRPC5
CYLD	MAP3K1	SDHC	BCL10	CYP3A4	GABRA6	KCNJ5	OR5L2	SBDS	TRRAP
DAXX	MAPK1	SDHD	BCL11A	CYP3A5	GATA1	KCNQ2	OR6F1	SCUBE2	TSHZ2
DDR1	MAX	SERPINB3	BCL11B	DCC	GATA2	KDM2B	P2RY8	SDC4	TSHZ3
DDR2	MCL1	SERPINB4	BCL2A1	DDX3X	GFRAL	KEL	P4HB	SEC31A	TTF1
DICER1	MDM2	SETD2	BCL2L11	DDX5	GIGYF1	KIF5B	PABPC1	SEMA3A	TUBA3C
DNMT3A	MDM4	SF3B1	BCL3	DEK	GKN2	KLF4	PABPC3	SEMA3E	TUBB3
DOT1L	MED12	SLX4	BCL6	DHX35	GLB1L3	KLHL6	PAG1	SEMA6A	TUSC3
EGFR	MEF2B	SMAD2	BCL9	DHX9	GLI1	KLK1	PAK1	SERPINA7	TXNIP
EIF1AX	MEN1	SMAD3	BCORL1	DIAPH1	GLI2	KRTAP5-5	PAK3	SETBP1	TYMS
C11orf30	MET	SMAD4	BCR	DIS3L2	GLI3	L3MBTL1	PASK	SETDB1	TYR
EP300	MITF	SMARCA4	BIRC3	DLC1	GMPS	LAMA2	PAX3	SF1	UBE2D2
EPAS1	MLH1	SMARCB1	BMPR2	DMD	GNA13	LATS1	PAX7	SF3A1	UBR5
EPCAM	MLH3	SMO	BNC2	DNAH6	GNG2	LATS2	PC	SFPQ	UGT1A1
EPHA2	MLL	SOCS1	BPTF	DNAJB1	GPC3	LCP1	PCDH18	SGCZ	UMPS
EPHA3	MLL2	SOX2	BRD2	DNM2	GPR124	LEF1	PCSK6	SGK1	UPF3B
EPHA5	MLL3	SOX9	BRD3	DNMT1	GPS2	LGALS8	PCSK7	SH2B3	USH2A

EPHB1	MPL	SPOP	BRSK1	DNMT3B	GPX1	LIFR	PDCD11	SH2D1A	USP6
EPHB6	MRE11A	SRC	BRWD1	DOCK2	GRB7	LPHN2	PDE4DIP	SH3PXD2A	USP8
ERBB2	MS4A1	STAG2	BTLA	DOCK7	GSK3B	LPP	PDGFB	SHH	VEZF1
ERBB3	MSH2	STAT3	BUB1	DPYD	GSTM5	LRP2	PDILT	SI	VIM
ERBB4	MSH3	STK11	C15orf23	DRGX	GSTP1	LRP4	PER1	SIN3A	VTCN1
ERCC1	MSH6	SUFU	C15orf55	DTX1	GUSB	LRP5	PGR	SLC16A1	WASF3
ERCC3	MST1R	SYK	C1QA	DUSP22	H3F3A	LRP6	PHF1	SLC1A2	WDR90
ERCC4	MTOR	TBX3	C1S	DYSF	H3F3B	LRRC7	PHF6	SLC22A16	WDTC1
ERCC5	MUTYH	TCF7L2	C3orf70	E2F3	H3F3C	LRRK2	PIK3C2A	SLC22A18	WHSC1
ERG	MYC	TERC	C7orf53	EBF1	HCLS1	LYN	PIK3C2B	SLC22A2	WHSC1L1
ERRFI1	MYCL1	TET2	C8orf34	ECT2L	HCN1	LZTS1	PIK3C2G	SLC22A3	WIPF1
ESR1	MYCN	TGFBR2	CACNA1E	EED	HDAC4	MACF1	PIK3C3	SLC34A2	WNK1
EXT1	MYD88	TMEM127	CADM2	EEF1A1	HDAC9	MAD1L1	PIM1	SLCO1B3	WNT5A
EXT2	NBN	TMPRSS2	CALR	EGFL7	HECW1	MAGI2	PKD1L2	SLIT1	WSCD2
EZH2	NCOR1	TNFAIP3	CAMTA1	EGR3	HEY1	MAML2	PKHD1	SLIT2	WWOX
FAM123B	NF1	TNFRSF14	CASP1	EIF2AK3	HIST1H1C	MAML3	PLAG1	SMARCD1	WWP1
FAM175A	NF2	TOP1	CASQ2	EIF2C3	HIST1H1D	MAP3K13	PLCB1	SMARCE1	WWP2
FANCA	NFE2L2	TOP2A	CBLB	EIF3A	HIST1H1E	MAPK3	PLCG1	SMC1A	XIAP

NGS raw data Process:

1. Filter out low quality reads from raw data by NCfilter.

2. The clean reads were aligned to the human genome build hs37d5 using sentieon bwa software. Subsequent BAM files were further processed for deduplication, base quality recalibration, and indel realignment using sentieon software.

3. Data quality control was performed using NCbamInfo from the final BAM files.

4. Somatic single nucleotide variants (SNVs) and somatic small insertions and deletions (Indels) were called using realDcaller and sentieon TNScope, respectively. The candidate somatic SNVs

and indels were annoted, and reviewed by BedAnno and HChot software. A series of designed filter rules were subsequently applied to filter out false positive sites.

5. Somatic structural variants (SV) were identified, annoted and filtered by by NCsv, annosv, and filtersv software, respectively.

6. Gene-level copy number variations (CNVs) were calculated by using CNVKit. Annocnv and filtercnv software were used for subsequent annotation and filterition.

All final candidate variants were manually verified with the integrative genomics viewer browser.

The central lab of FISH testing (tissue samples) and ddPCR testing (plasma samples): Righton Clinical Laboratory Co.,Ltd

Tissue Sample requirements

FFPE: 2 slides, 5mm length * 5mm width * 5um thickness;

Tumor content: ≥20%

Table 12 Validation information of tumor tissue sample detected by FISH

Assay	Vysis MET SpectrumRed FISH Probe kit (Abbott)
Sample type	Tissue
Tumor cells	20-30 cells
Cut off	GCN ⩾ 10

Plasma Sample requirements

Peripheral whole blood sample: 10ml

Table 13 Validation information of plasma sample detected by ddPCR

Sample type	Plasma

Sample requirement	1. non- hemolysis 2. extraction DNA ⩾ 0.5 ng/ul 3. total DNA content ⩾ 25 ng
Assay	MET gene amplification testing assay -ddPCR
Cut off	MET/CEP7 ⩾ 2

Appendix G Abbreviations

Abbreviation or Special Term	Explanation
CD	cluster of differentiation
CI	confidence interval
CR	complete response
CRF	Case report form
CSP	Clinical Study Protocol
CSR	Clinical Study Report
CT	computed tomography
ctDNA	circulating tumour DNA
DCO	data cut-off
DNA	deoxyribonucleic acid
EC	Ethics Committee, synonymous to Institutional Review Board (IRB) and Independent Ethics Committee (IEC)
ECOG	Eastern Cooperative Oncology Group
eCRF	electronic Case Report Form
EGFR	epidermal growth factor receptor
FAS	Full Analysis Set
FDA	Food and Drug Administration
FFPE	formalin fixed and paraffin embedded
FISH	fluorescent in situ hybridization
GCP	Good Clinical Practice
HR	hazard ratio
IATA	International Airline Transportation Association
IB	Investigator's Brochure
ICF	informed consent form
ICH	International Council for Harmonisation
IEC	Independent Ethics Committee
IHC	Immunohistochemistry
IRB	Institutional Review Board
MRI	magnetic resonance imaging
NE	not evaluable
NL	new lesion
NSCLC	non-small cell lung cancer
NTL	non-target lesion
ORR	objective response rate

Abbreviation or Special Term	Explanation
OS	overall survival
PD	progression of disease
PET	positron emission tomography
PFS	progression-free survival
PR	partial response
RECIST 1.1	Response Evaluation Criteria in Solid Tumors, Version 1.1
SAP	Statistical Analysis Plan
SD	stable disease
SoA	Schedule of Activities
TKI	tyrosine kinase inhibitor
TL	target lesion
TOC	Table of Contents
TTD	time to treatment discontinuation or death
VAF	variant allele frequency
WHO	World Health Organisation
ddPCR	droplet digital PCR

Appendix H Performance of droplet digital PCR (ddPCR) for MET amplification testing

Table A: The consistency of ddPCR and FISH in detecting MET amplification in tissue samples

FISH	ddPCR, *n*				Sensitivity	Specificity	Concordance	κ
	MET amp	Polysomy	MET neg	Total	100%	100%	100%	1.00
MET high amp	1	0	0	1				
MET low amp	8	0	0	8				
Polysomy	0	6	0	6				
MET neg	0	0	87	87				
Total	9	6	87	102				

amp amplification, *neg* negative

Table B: The concordance of MET amplification between tissue and matched peripheral blood samples detected by ddPCR

Tissue	Peripheral blood, *n*				Sensitivity	Specificity	Concordance	κ
	MET amp	Polysomy	*MET* neg	Total	66.67%	98.86%	94.17%	0.75
MET amp	4	0	5	9				
Polysomy	0	6	0	6				
MET neg	1	0	87	88				
Total	5	6	92	103				

amp amplification, *neg* negative

Table A: In tissue samples from 102 advanced NSCLC patients who had progressed on EGFR-TKI, the consistency of MET amplification between ddPCR testing and FISH testing. (1 case failed for FISH detection owing to the lack of sufficient tumor cells, remaining 102 cases had both FISH and ddPCR results) [26]

Table B: In tissue and the matched peripheral blood samples from 103 advanced NSCLC patients who had progressed on EGFR-TKI, the concordance of MET amplification detected by ddPCR between tissue and matched peripheral blood samples (In total, all 103 patients were involved in this analysis) [26].

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