PHASE II STUDY OF ORAL PHA-848125AC IN PATIENTS WITH THYMIC CARCINOMA PREVIOUSLY TREATED WITH CHEMOTHERAPY

Sponsor:	Tiziana Life Sciences, PLC
Therapeutic Area:	Oncology
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Name of Finished Product: PHA-848125AC (Milciclib maleate)

Title of Study: PHASE II STUDY OF ORAL PHA-848125AC IN PATIENTS WITH THYMIC CARCINOMA PREVIOUSLY TREATED WITH CHEMOTHERAPY

Protocol Number: CDKO-125a-006

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Background Information

Thymic carcinomas are a category of heterogeneous thymic tumors, with distinct molecular characteristics and with the most aggressive behavior and poorest prognosis among thymomas [1]. Thymomas are rare tumors but nevertheless the most common neoplasms of the anterior mediastinal compartment, with an overall incidence in US (1973-1998) of 0.15 per 100000 person/years [2]. Patients with locally advanced or disseminated thymic malignancies are usually symptomatic, presenting with chest pain, shortness of breath, paralysis of the phrenic nerve, pleural effusion and superior vena cava syndrome. Immune disorders have also been associated with thymoma, the most common being myasthenia gravis [3]. There is no known cause for thymomas, however, in recent years, a few studies on molecular changes in thymomas and thymic carcinomas have been published. In particular, the expression of KIT and epidermal growth factor receptor (EGFR), targets, respectively, of imatinib and EGFR inhibitors (ie, erlotinib, gefitinib), have been tested by immunohistochemistry in a limited number of thymic malignancies: KIT was positive in 73% of thymic carcinomas and 5% of thymomas, in contrast, EGFR expression was more often present in thymomas (83%) than in thymic carcinomas (50%) [4]. Another study investigated HER2/neu amplification and expression of the apoptosis-related markers p53, BCL-2, BAX, BCL-XL, and p21 [5]; p53 and Bcl-2 were definitely more expressed in thymic carcinomas than thymomas and correlated with more advanced stages and unresectability, whereas no HER2/neu amplification was observed. Other markers, such as expression of thymidylate synthase and dihydropyrimidine dehydrogenase, which predict sensitivity to 5-fluorouracil-based chemotherapy, were not correlated with clinicopathological characteristics in a series of thymic malignancies [6].
The loss of control of cell cycle checkpoints seems a common occurrence in thymomas, supporting the idea that functional cooperation between different cell cycle inhibitor proteins regulates cell growth control and tumor suppression. Univariate analyses on the expression of p21 and p27 in specimens from 25 encapsulated thymomas (using immunohistochemistry) suggest that negative expressions of p21 and p27 (natural inhibitors of CDKs) significantly correlates with poor prognosis for disease-free survival [7]. In addition, some members of the neurotrophin receptors family seem to play a significant role in this disease: in a study investigating the expression of neurotrophin receptors in thymic epithelial tumors on a quite large series of patients (N=99) [8], the pattern of TRKA expression was analyzed according to WHO classification for the histologic subtypes (A, AB, B1, B2, B3, C). All patients, except one, had immunostaining for TRKA expression in the tumors gradually increasing from type A to C. Conversely, none of the tumors showed TRKB or TRKC immunoreactivity. In addition, from type A to type C, there was a decreasing percentage of tumors with p75 immunostaining. These observations suggest a further elucidation on the specific role of TRKA and p75 in this disease.
Besides the above mentioned WHO histological classification of thymoma, the Masaoka staging system is commonly employed to evaluate invasiveness and to determine the therapy, since the optimal treatment for this disease depends on its clinical stage [1]. Locally advanced or metastatic thymomas are often treated with combined treatment modalities, including surgery, radiation and chemotherapy. There are no drugs officially approved for the treatment of thymoma and thymic carcinoma. However, several antineoplastic agents are used; cisplatin/doxorubicin-based combination chemotherapy [PAC regimen (cisplatin, doxorubicin, cyclophosphamide) or ADOC regimen (doxorubicin, cisplatin, vincristine, cyclophosphamide)] seem to

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produce the best overall response rate and survival. Other combined and /or single agent chemotherapy with cisplatin, etoposide, ifosfamide, epirubicin, maytansine, octreotide and steroids are used as well [9-11]. Examining treatments for patients with advanced disease who had received one or more prior lines of therapy, published data generally indicates a limited activity. Single agent studies with gefitinib, imatinib or a combination study erlotinib+bevacizumab did not produce satisfactory results [12,13,14], although only two cases of possible clinical benefit were observed with imatinib and sorafenib, due to the rare incidence of KIT mutations in those patients' tumors [15,16]. An optimal treatment strategy has yet to be determined and other drugs are warranted to improve the outcome of patients with advanced invasive tumors [10]. The unmet medical need for new agents for the treatment of thymic malignancies is therefore high, as demonstrated by the number of studies with experimental agents currently in progress.
Given the rarity of thymic carcinoma subtype, prospective phase II studies of chemotherapy specifically targeting thymic carcinoma patients are rare. Combined therapies such as ADOC or VIP (cisplatin, ifosfamide, etoposide) achieved, as a first line treatment for the advanced disease, a median survival time around 19-20 months [17,18]. In previously untreated, unresectable thymic carcinoma, first-line combination of carboplatin and paclitaxel achieved an overall response rate of 36% with a median survival time of 22.7 months and a median progression-free survival of 7.9 months [19].
In a single agent study with pemetrexed, on 23 previously treated evaluable patients with unresectable stage IV A and B disease, there were 4 objective responses (2 CRs and 2 PRs) (17% RR). Responses were obtained only in stage IVA thymoma patients, with a median duration of time to progression of 45.4 weeks; in thymic carcinoma patients the median duration of time to progression was much shorter (5.1 weeks) [20]. In other two recent series of previously treated patients with thymic malignancies, objective responses were reported in B2 and B2/3 tumors (no type C thymic carcinoma patients were enrolled in the first study) with a gemcitabine/ capecitabine combination [21] and with the histone deacetylase inhibitor (HDAC) belinostat [22]. Also in this second study, responses were obtained only in thymoma patients and no responses were seen in the 8 evaluable patients with thymic carcinoma. Thymic carcinoma is therefore confirmed as a type of cancer where prognosis is particularly poor and the course of the disease more aggressive compared to the other thymomas.

Given the current clinical scenario, in thymic carcinoma patients previously treated with chemotherapy, a single agent therapy is able to induce a median PFS of approximately 5.1 weeks [20], roughly corresponding to an estimated PFS rate at 3 months of about 17%. Therefore a 33% target PFS-rate at three months could be regarded as clinically interesting for a new investigational agent.

PHA-848125, N,1,4,4-tetramethyl-8-{[4-(4-methylpiperazin-1-yl)phenyl]amino}-4,5-dihydro-1H-pyrazolo[4,3-h]quinazoline-3-carboxamide, is a potent inhibitor of the kinase activity of the CDK2/Cyclin A complex, some closely related CDKs (i.e. CDK1, CDK4, and CDK5) and TRKA; the block in G1 phase of the cell cycle observed in tumor cells exposed to PHA-848125 supports the postulated mechanism of action as determined by this biochemical activity: in fact, PHA-848125 is able to modulate both the phosphorylation of the Retinoblastoma protein (pRB), a substrate of CDK/Cyclin complex, as well as to reduce the phosphorylation status of TRKA and of the proteins of the TRKA signaling pathway [23].
PHA-848125, formulated for oral administration as maleate salt (PHA-125848AC), is being tested, with different schedules of administration, in two ongoing phase I trials in advanced/metastatic solid tumor patients, in a phase I/II study in patients with recurrent malignant glioma, in a phase I combination study with gemcitabine in advanced/metastatic solid tumor patients and in a phase II study in patients with malignant pleural mesothelioma.
In the first-time in man study CDKO-125a-001, in which PHA-848125AC was administered for 7 consecutive days every 2 weeks to solid tumor patients, the flat dose identified as recommended phase II dose (RP2D) was 150 mg/day; this dose was associated with mild/moderate reversible adverse events, represented by maximum grade 2 nausea and/or vomiting and/or diarrhea and maximum grade 2 reversible tremors.

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As for efficacy, notably, one out of three patients with thymic carcinomas, treated in the study CDKO-125a-001 at the dose 150 mg/day, showed a partial response reached after 11 treatment cycles, and another patient showed a stable disease with progressively shrinking tumor lesions after 2, 4 and 6 treatment cycles; the third patient was taken off study after two cycles for disease progression. It is of note that lesion regression was reported in patients with the most unfavorable histology (thymic carcinoma).

Study rationale

Chemotherapy was shown to have significant antitumor activity against unresectable, recurrent or metastatic thymomas and thymic carcinomas, but an optimal treatment strategy has not been determined and other drugs are needed to improve the outcome of patients with advanced invasive tumors [10].
In recent years, a few studies on molecular changes in thymomas and thymic carcinomas point to alterations that may be of use in selecting patients for targeted therapies. Unfortunately for some of them, such as KIT and EGFR inhibitors, the reported results seem to be discouraging [12-14]. In addition, despite a lack of correlation of thymidilate synthase expression with thymoma clinicopathological characteristics, pemetrexed obtained interesting responses in stage IVA thymomas, but not in thymic carcinomas [20].
The pyrazoloquinazoline PHA-848125 is a compound counteracting the effects of some of the other so far identified, but not yet exploited, molecular alterations found in thymoma and thymic carcinoma, e.g. underexpressed p21 and p27 and overexpressed TRKA [7,8]. These targets have not yet been investigated in therapeutic intent clinical trials.
In fact, p21 and p27 encode natural potent cyclin-dependent kinase inhibitors and their ablation in the cancer cell results in an activation of cyclin E/CDK2 or cyclin D/CDK4 complexes and thus uncontrolled cell cycle progression and cell proliferation [24-32], while TRKA activation promotes tumor growth [33], probably thorough MAPK pathway activation. PHA-848125 potently inhibits both the CDK/Cyclin complexes and TRKA, proposing in this way to be a “selective dual inhibitor” and appearing particularly attractive in this disease.
The preliminary results in early clinical trials on thymic carcinomas patients (2 out of 3 patients with clinical benefit) support the hypothesis that PHA-848125 could be of therapeutic value for patients with this tumor type.
An exploratory analysis on the relationship of molecular features of p53 (natural regulator of p21), p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action with treatment efficacy variables will be possibly performed in tumor biopsies of consenting patients obtained before study entry.

Trial Objectives

Primary objective:
Assessment of the antitumor activity of PHA-848125AC as second-line treatment in patients with recurrent or metastatic, unresectable thymic carcinoma previously treated with chemotherapy. Antitumor activity will be evaluated on the basis of the progression-free survival status at 3 months.

Secondary objectives:
Assessment of additional measures of tumor control to further characterize the efficacy profile of PHA-848125AC in recurrent or metastatic, unresectable thymic carcinoma patients previously treated with chemotherapy.

Evaluation of the safety profile of repeated administrations of PHA-848125AC in patients with recurrent or metastatic, unresectable thymic carcinoma previously treated with chemotherapy.

Exploratory objective:
Relationship of baseline molecular features in tumor biopsies of p53, p21, p27, cyclin D1, p75, TRKA and

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other genes/proteins involved in the PHA-848125AC mechanism of action with treatment efficacy.

Trial endpoints

Primary endpoint:
Progression-free survival rate at 3 months (PFS-3 rate). The PFS-3 rate, will be calculated as the proportion of evaluable patients known to be alive and progression-free at ≥ 3 months since study treatment start out of the total number of evaluable patients.

Secondary endpoints:

- Confirmed Objective Response Rate (CR + PR) according to RECIST guideline (version 1.1).
- Disease Control Rate (Confirmed Objective Response Rate + ≥ 6 weeks SD rate).
- Progression-free survival, calculated as the time from the date of treatment start to the date of first documentation of objective progression or of death due to any cause, whichever comes first
- Duration of Response, measured from the time measurement criteria are first met for CR/PR (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded on study).
- Overall Survival (OS), i.e. the time from the date of treatment start to the date of death from any cause.
- Overall safety profile, evaluated on the basis of laboratory and clinical safety parameters (i.e. hematology and blood chemistry, urinalysis, vital signs, ophthalmologic examinations and adverse events emerging during the trial). The National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE) Version 3.0 will be used for the severity grading of adverse events and hematological and blood chemistry abnormalities.

Exploratory endpoint:
Baseline characterization of selected biomarkers (p53, p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action) in tumor tissue of consenting patients. Assessments will be done on paraffin embedded blocks, obtained from all patients before study entry, by IHC and other possible techniques, such as FISH and PCR.

Study Design

In consideration of the exploratory nature of the study, the Simon's optimal 2 stage design [23] is adopted for this single-arm, open-label, multicentre phase II clinical trial.

Histological confirmation of diagnosis of thymic carcinoma for all patients will be obtained by an Independent Review Committee.

Sample Size Determination and Statistical Methods:

The total number of evaluable patients for the primary efficacy analysis ranges from 17 (if the trial stops at the end of the 1st stage) to 54 (if the trial proceeds up to the completion of the 2nd stage). Accounting for a 10%-15% proportion of non evaluable patients, up to 20 patients could be required in the 1st stage of the study and up to overall 60 patients could be required for completing the trial (1st and 2nd stage).

The primary endpoint of the study is the progression-free survival status at 3 months and the primary efficacy analysis will be performed on the proportion of successes (i.e. patients alive and in a progression-free status at 3 months since treatment start) out of the total number of evaluable patients (PFS-3 rate).

Considering a 33% PFS-3 rate as clinically interesting, against a clinically uninteresting hypothesis of a PFS-3 rate no higher than 17%, the system of hypotheses to be tested is:

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$$H_0: p \le p_0, \quad p_0 = 0.17 \quad vs. \quad H_1: p \ge p_1, \quad p_1 = 0.33$$

The analysis will be performed at the overall level $\alpha = 0.05$ (1-sided).
The design below outlined will provide 80% power (1-β) not to reject the treatment as insufficiently effective, if the true PFS-3 rate is 33% or higher.

In the first stage, 17 evaluable patients are to be assessed. If ≤ 3 successes are observed, the study treatment will be considered as providing insufficient evidence of efficacy and the trial will be terminated.
If at least 4 successes are observed in the 1st stage, patients' enrollment will proceed up to an overall enrollment of 54 evaluable patients. At the final analysis, ≥ 14/54 successes (PFS-3 rate ≥ 25.9%) will be required to reject the null hypothesis and suggest that the drug might have an interesting level of efficacy.

			1st Stage (*)		2nd Stage		
H_0 *vs.* H_1	**α (1-sided)**	**power (1-β)**	**Pts**	**STOP and reject drug**	**Pts**	**Reject drug**	**Do not reject drug**
$p_0 \le 0.17$ *vs.* $p_1 \ge 0.33$ *(PFS-3 rate)*	0.05	0.80	17 evaluable pts	≤ 3 / 17 successes	54 evaluable pts	≤ 13 / 54 succ. (PFS-3 rate ≤ 24.1%)	≥ 14 / 54 succ. (PFS-3 rate ≥ 25.9%)
(*) Probability of Early Termination: PET=0.675, if the drug is actually ineffective; Probability to continue the trial > 0.863, if the true PFS-3 rate is at least 33%:							
Expected sample size assuming ineffective drug: 29							
No. of enrolled patients: ≤ 20 patients in the 1st stage, ≤ 60 patients overall (if 10%-15% inevaluable patients)							

The progression free survival at 3 months will be evaluated based on the antitumor activity evaluated during the oncologic assessment at 3 months after first drug administration. The oncologic assessment will be performed preferably between 92-98 days from treatment start, but all patients with assessments performed up to 134 days will be considered evaluable for the primary end-point.

Supportive analyses of the primary endpoint will include the estimation of the PFS-3 rate together with its exact, two-tail, 95% confidence interval and the estimation of the PFS curve by the Kaplan-Meier method [24] in both the evaluable and the treated patient population. The other efficacy endpoints including the confirmed objective response rate, the disease control rate, the duration of response, and the overall survival will be descriptively analyzed in both the evaluable and the treated patient populations. Kaplan-Meier estimates will be generated and plotted for the overall survival endpoint.
Patients' baseline characteristics, treatment exposure and safety data will be analyzed in the treated patient population and, if clinically interesting, in other subsets such as the evaluable patient population.
Descriptive statistical analyses and individual data listings will be used to report all collected data including patient disposition, protocol deviations, baseline characteristics, treatment exposure, efficacy, and safety data.

Patient Populations:

- *Screened Patients:* This population will include all subjects who are screened about their eligibility for the trial, regardless of whether or not they will be enrolled in the study. This population will be

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evaluated in the analysis of patients' disposition.

- *Enrolled Patients:* This population will include all subjects who are enrolled in the trial, regardless of whether subjects receive the study drug or not. This population will be evaluated in the analysis of patients' disposition.
- *Treated Patients:* The treated patient population consists of all enrolled patients who actually receive at least one study drug administration. This population will be evaluated in the analysis of patient disposition, baseline characteristics, treatment efficacy and safety and treatment exposure.
- *Patients Evaluable for Efficacy Analysis:* This is the patient population for the primary efficacy analysis of PFS-3 rate and consists of all treated patients who fulfill the following additional conditions:
    - They have received histological confirmation of thymic carcinoma by an Independent Review Committee
    - They have received at least 80% of drug in the first two cycles overall.
    - They have baseline and ≥1 on-treatment tumor/oncologic assessment(s) or die before tumor re-assessment.

  If deemed of clinical interest, patient disposition, baseline characteristics, treatment efficacy and safety and treatment exposure will be analyzed also in this population.

Exploratory studies

Exploratory analyses on the relationship of molecular features of p53, p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action with treatment efficacy variables will be performed if sufficient data and samples (tumor biopsies obtained before study entry) are collected.

Subject Selection

Subject Inclusion Criteria

Subjects must meet all of the following inclusion criteria to be eligible for enrollment into the study:

1. Signed and dated IRB/IEC-approved Informed Consent.
2. Histologically or cytologically proven diagnosis of unresectable B3 thymoma or thymic carcinoma recurrent or progressing after prior chemotherapy (only one prior systemic therapy allowed).
3. Presence of measurable disease defined as at least one lesion that can be accurately measured by CT scan in at least one dimension, as > 10 mm for non nodal lesions (longest diameter to be recorded) and ≥ 15 mm for lymph nodal lesions (short axis to be recorded). (CT scan is the desirable method for lesion measurement. Other measurement techniques [*eg* MRI] are acceptable [but not for lung lesions] provided that the size of the measurable lesion is twice the slice thickness of the MRI).
   Tumor lesions situated in a previously irradiated area, or in an area subjected to other loco-regional therapy, are usually not considered measurable unless there has been demonstrated progression in the lesion.
4. Age ≥ 18 years.
5. ECOG performance status 0-1.
6. Estimated life expectancy of at least 3 months.
7. Negative pregnancy test (if female in reproductive years).

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8. Agreement upon the use of effective contraceptive methods (hormonal or barrier method of birth control, or abstinence) prior to study entry and for the duration of study participation, if men and women of child-producing potential.
9. Adequate liver function:
   - Total Serum Bilirubin ≤ 1.5 x upper limit of normal (ULN)
   - Transaminases (AST/ALT) ≤ 2.5 ULN (if liver metastases are present, then ≤ 5 ULN is allowed)
   - ALP ≤ 2.5 ULN (if liver and/or bone metastases are present, then ≤ 5 ULN is allowed).
10. Adequate renal function:
    - Serum Creatinine ≤ ULN
    or
    - Creatinine Clearance calculated by Cockcroft and Gault's formula > 60 mL/min..
11. Adequate hematologic status:
    - ANC ≥ 1,500 cells/mm^3
    - Platelet Count ≥ 100,000 cells/mm^3
    - Hemoglobin ≥ 9.0 g/dL.
12. At the time of start of treatment, at least 2 weeks must have elapsed since completion of prior chemotherapy, minor surgery and radiotherapy (provided that no more than 25% of bone marrow reserve has been irradiated).
13. With the exception of alopecia, resolution of all acute toxic effects of any prior chemotherapy, surgery or radiotherapy to NCI CTC (Version 3.0) grade ≤ 1 and to the baseline laboratory values as defined in Inclusion Criteria Number 9, 10, 11.
14. Able and willing to comply with scheduled visits, therapy plans, and laboratory tests required in this protocol.
15. Capability to swallow capsules intact (without chewing, crushing, or opening).

Subject Exclusion Criteria

The presence of any of the following will exclude a subject from study enrollment:

1. Any of the following in the past 6 months: myocardial infarction, uncontrolled cardiac arrhythmia, unstable angina, coronary/peripheral artery bypass graft, symptomatic congestive heart failure, cerebrovascular accident or transient ischemic attack, pulmonary embolism, deep vein thrombosis.
2. Grade >1 retinopathy as determined by an ophthalmologist.
3. Known brain metastases.
4. Major surgery, other than diagnostic surgery, within 4 weeks prior to treatment.
5. Active, uncontrolled bacterial, viral, or fungal infections.
6. Known infection with HIV, active hepatitis B or hepatitis C.
7. Pregnant or breast feeding women.
8. Previous (within the last 5 years) or current malignancies at other sites, except for adequately treated basal cell or squamous cell skin cancer or in situ carcinoma of the cervix uteri.
9. Current enrollment in or participation in another therapeutic clinical trial within 4 weeks preceding

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treatment start.

10. Diabetes mellitus uncontrolled.
11. Gastrointestinal disease (e.g. Crohn's disease, ulcerative colitis, or short gut syndrome) that would impact on drug absorption.
12. Patients under treatment with anticoagulants or with coagulation disorders or with signs of hemorrhage at baseline.
13. Patients with previous history or current presence of neurological disorders, including epilepsy (although controlled by anticonvulsant therapy), Parkinson's disease and extra-pyramidal syndromes.
14. Other severe acute or chronic medical or psychiatric condition or laboratory abnormality that may increase the risk associated with study participation or may interfere with the interpretation of study results and, in the judgment of the Investigator, would make the patient inappropriate for entry into this study or could compromise protocol objectives in the opinion of the Investigator and/or the Sponsor.

Treatment and Test Articles

On the basis of the results of the first single agent Phase I study in solid tumor patients (CDKO-125a-001 study), PHA-848125AC will be administered at the flat dose of 150 mg/day once daily for 7 consecutive days in each treatment cycle. A treatment cycle will comprise 7 days of PHA-848125AC administration (Days 1 to 7) followed by 7 days of rest (Days 8 to 14) for a total of a 14 days period (2-week cycle).

PHA-848125AC is formulated as 10 mg, 50 mg and 100 mg hard gelatin capsules to be swallowed intact (without chewing, crushing or opening). The drug product is to be stored under refrigerated conditions (5 ± 3°C; 36-46 °F) and in the original packaging, out of the reach of children.
PHA-848125AC is administered as home-based treatment. After an overnight fasting, with free access to water, patients will take the study drug with a large glass of plain water without ice. A light breakfast can be served 1.5-2 hours after study drug intake.

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Schedule of Events

The following table summarizes information on the timing of study assessments.

SCHEDULE OF EVENTS

Protocol Activities and Forms to Be Completed	Pre Treatment		Treatment			Post Treatment	
			Cycles		Last Cycle		
	≤ 21 Days	≤ 7 Days	Week 1 Day 1 (*)	Week 2 between Days 11-14		AE FU (§)	Follow up
Informed Consent **1**	X						
Medical/ Oncologic History & Physical Examination **2**	X			X	X		
Vital Signs **3**		X	X				
ECG **4**	X		As medically indicated		X		
Chest X-Ray **5**	X				X		
Ophthalmologic exam. **6**	X			X (even cycles)	X		
Laboratory Assessments							
Serum/ Urine Pregnancy Test **7**		X					
Urinalysis **8**		X	X **(*)**	X (even cycles)	X		
Hematology **9**		X	X **(*)**	X			
Blood Chemistry **10**		X	X **(*)**	X			
Coagulation **11**		X	X **(*)**	X (even cycles)	X		
Tumor Assessments							
Tumor Imaging: chest/abdomen/pelvic CT scan or MRI **12**	X			q6wks	X		q6wks
Survival **13**							q6wks (+ q6mos)
Other Clinical Assessments							
Adverse Event Assessment (including neurological status evaluation) **14**		X	X			X	
Concomitant Medications **15**		X	X				
Special Studies							
Biomarker Status Assessment **16**	X						
Pharmacokinetic Blood Sampling **17**			As needed				
Histological confirmation **18**	X						
Study Treatment							
Enrollment **19**		X					
PHA-848125AC Administration **20**			Once daily for 7 cons. days (D1 to D7)				

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Footnotes for Schedule of Events
1. Informed Consent: Every patient must sign the informed consent to participate in this trial before starting any trial related procedures. After signature, a Patient Screening Number is assigned centrally by CLIOSS on behalf of Tiziana Life Sciences, PLC.
2. Medical/Oncology History & Physical Examination: Includes oncologic history, history of other disease processes (active, controlled or resolved) and concomitant illnesses (all of them at pretreatment only). Physical examination will be done at pretreatment, and, on treatment, at the end of each cycle (see Section 11.4.3 Other safety assessments). For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the physical examination will be done in concomitance with the visit to the site (once every 3 cycles, 1.5 months). Oncologic and non-oncologic medical history, including prior antitumor treatments/procedures, are to be reported in the relevant case report form (CRF). **Following approval of Amendment 4, management of patients should be as per Section 9.5**
3. Vital signs: Blood pressure/pulse (supine): at baseline and on Day 1 of each treatment cycle, before treatment administration. Height (in cm or inches): at baseline only. Weight (in kilograms or pounds): at baseline and on Day 1 of each treatment cycle. ECOG performance status: at baseline and on Day 1 of each treatment cycle. Temperature (in °C or in °F): at baseline and on Day 1 of each treatment cycle. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, vital signs (including blood pressure/pulse, weight, ECOG performance status and temperature) will be done in concomitance with the visit to the site (once every 3 cycles, 1.5 months). **Following approval of Amendment 4, management of patients should be as per Section 9.5**
4. ECG: 12-lead study. To be done at baseline, at the end of the last cycle and to be repeated during treatment as medically indicated.
5. Chest X-Ray: Mandatory at screening visit and at the end of last cycle as safety assessment.
6. Ophthalmologic Examination: to be conducted at baseline, at the end of even cycles (between days 11 and 14) and at the end of last cycle. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, opthalmological examination will be done in concomitance with the visit to the site (once every 3 cycles, 1.5 months). Includes visual acuity test and fundoscopic examination by an ophthalmologist. Additional assessments will be performed if clinically indicated. Ophthalmologic evaluations will be possibly done by the same ophthalmologist for a given patient. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
7**. Pregnancy Test:** For women of reproductive potential. Must be done within 7 days prior to initial study treatment.
8. Urinalysis: pH, dipstick for glucose, protein and blood. To be done at baseline and at the end of last cycle. During treatment, should be performed at the end of even cycles (between days 11 and 14). For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the urinalysis tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
9. Hematology (local laboratory): Hemoglobin, erythrocytes (RBC), white blood cell (WBC) with differential count (neutrophils, lymphocytes, monocytes, eosinophils, basophils, differential other cells), platelets (PLTs). To be done at baseline and between Day 11-14 of each cycle. In case of toxicity preventing re-treatment, hematology should be re-assessed on Day 1 of the next cycle before starting treatment. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the hematological tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
10. Blood Chemistry (local laboratory)**:** sodium, potassium, chloride, magnesium, blood urea nitrogen (BUN) or urea, creatinine, albumin, AST/SGOT, ALT/SGPT, Alkaline Phosphatase (ALP), total bilirubin, creatinine clearance (calculated by Cockroft and Gault's formula), amylase and lipase. To be done at baseline and between Day 11-14 of each cycle. In case of toxicity preventing re-treatment, blood chemistry should be re-assessed on Day 1 of the next cycle before starting treatment. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the biochemical tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**

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11. Coagulation: INR (International Normalized Ratio), activated partial thromboplastin time (APTT in seconds). To be done at baseline, at the end of last cycle and, during treatment, at the end of even cycles (between Days 11 and 14). For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the coagulation tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF.
Following approval of Amendment 4, management of patients should be as per Section 9.5

12. Tumor imaging: chest/abdomen/pelvic CT scan/ MRI : At baseline, to be done within 21 days before Day 1; during the treatment period (irrespectively from cycles duration), the first one to be done 6 weeks (between days 42 and 48) after first drug administration, **the second one to be done 3 months (between days 92 and 98) after first drug administration**, following ones to be repeated every 6 weeks (between days 42 and 48) after the previous one, and at end of last cycle (if not done in the previous 4 weeks). During Follow Up, to be performed every 6 weeks, until PD or until a new antitumor therapy starts. Patients with responding tumors (complete or partial response) must have response confirmed by CT scan at least 4 weeks after the 1st documentation of response. The same method of assessment and the same technique should be used to characterize and follow the same lesion at baseline and during treatment. CT scan is the desirable method for lesion measurement. Other measurement techniques (eg MRI) are acceptable (but not for lung lesions) provided that the size of the measurable lesion is twice the slice thickness of the MRI. **After data cut-off of 31 May 2017, the patients should perfom CT scan according to routine clinical practice and/or at investigator's discretion.**

13. Survival: To be assessed during Follow Up every 6 weeks until PD or until a new antitumor therapy starts; every 6 months thereafter, up to 2 years from the end of treatment.
Following approval of Amendment 4, management of patients should be as per Section 9.5

14. Adverse Event Assessment (including neurological status evaluation): Events should be assessed and documented at each scheduled clinic visit since patient signs the Informed Consent: before treatment administration at baseline (baseline signs and symptoms), at the end of each treatment cycle, and 28 days after the last dose of study drug administration. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the events will be assessed by phone call if the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months). The phone call information will be reported in the medical chart and included in the CRF.
Patients must be followed for adverse events also until every ongoing drug-related toxicities and serious adverse events have resolved or the investigator assesses them as "chronic" or "stable". However, if the patient begins a new anticancer therapy before 28 days after the last dose of study drug administration, the adverse event reporting period will end at the time the new treatment is started. AEs will be recorded in the relevant CRF pages.
After cut-off date of 31 May 2017, AE/SAE data will be no longer collected into the CRF, but assessments information should continue to be collected into the patient's medical notes.
After protocol Amendment 5 approval, only SAEs will continue to be reported to CLIOSS Pharmacovigilance for patients still receiving treatment, up to 28 days after milciclib discontinuation, according to Section 9.5.

15. Concomitant Medications: all concomitant medications should be reported in the relevant CRF, including supportive care drugs, and drugs used for treating adverse events or chronic diseases. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the concomitant medications will be assessed by phone call if the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months). The phone call information will be collected in the medical chart and included in the CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**

16. Biomarker Status Assessment: retrieval of tumor paraffin embedded blocks (slides), obtained from consenting patients before study entry (if available) and characterization of baseline molecular features of p53, p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action.

17. Pharmacokinetics: blood samples may be collected if, in the opinion of the Investigator and of the Sponsor, an evaluation of PK parameters is needed for safety reasons.

18. Histological confirmation: histological confirmation of diagnosis of thymic carcinoma for all patients will be obtained by an Independent Review Committee during the course of the study, based on paraffin embedded tumor tissue slides retrieved from patients.

19. Enrollment: The Investigator is requested to sign a Request For Enrollment form. His/her signature is the guarantee that all the eligibility criteria are met. Patient Number is assigned by the Sponsor. Treatment must be administered within 7 days from patient enrollment.

20. PHA-848125AC administration: The study drug will be administered at 150 mg (flat dose) once daily for 7 consecutive days of each treatment cycle (from day 1 to Day 7). After an overnight fasting with free access to water, patients will take the study drug with a large glass of plain water without ice. A light breakfast can be served 1.5-2 hours later. PHA-848125AC capsules have to be stored under refrigerated conditions (5 ± 3 °C; 36-46 °F) and out of reach of children

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(*) Cycle 1, Day 1 assessments: hematology, blood chemistry, urinalysis and coagulation do not need to be repeated before treatment on Cycle 1, Day 1 if they were performed during screening within 1 week before first study drug administration.
Cycle > 1, Day 1 assessments: hematology and blood chemistry do not need to be repeated before treatment on Cycles> 1, Day 1 if no toxicity preventing re-treatment resulted from laboratory assessments done within the previous 4 days.

(§) AE FU: end of adverse events reporting period (to be done 28 days after last study drug administration) and follow up of unresolved drug related/serious AEs. **After data cut-off of 31 May 2017, AE/SAE data and related follow up will be no longer collected into the CRF, but assessments information should continue to be collected into the patient's medical notes. After approval of Amendment 4 to the protocol, only SAEs should be reported to CLIOSS Pharmacovigilance for patients still receiving study treatment, up to 28 days after discontinuation of milciclib, according to Section 9.5.**

2. ABBREVIATIONS AND DEFINITIONS OF TERMS

ADL	Activities of Daily Living
AE	Adverse Event
ALP	Alkaline Phosphatase
ALT/SGPT	Alanine aminotransferase
ANC	Absolute neutrophils count
APTT	Activated partial thromboplastin time
AST/SGOT	Aspartate aminotransferase
AUC	Area under the plasma concentration-time curve from time zero to infinity
BAX	BCL2-associated X protein
BCL-2	B-cell CLL/lymphoma 2 gene (BCL2)
BCL-XL	BCL2-like 1 (BCL2L1)
BID	Two times daily
BUN	Blood Urea Nitrogen
CDKs	Cyclin-dependent kinases
CL	Total plasma clearance
Cmax	Maximum plasma concentration
CNS	Central nervous system
CR	Complete Response
CRF	Case Report Form
DMBA	9,10-dimethyl-1,2-benzanthracene
DPYD	Dihydropyrimidine dehydrogenase
ECOG	Eastern Cooperative Oncology Group
GCP	Good clinical practice
EGFR	Epidermal growth factor receptor
FISH	Fluorescence in Situ Hybridization
FU	Follow-up
GI	Gastrointestinal
HER2/neu	v-erb-b2 erythroblastic leukemia viral oncogene homolog 2, neuro/glioblastoma derived oncogene homolog (ERBB2)
HLP	Hemolymphopoietic
IC_{50}	Concentration resulting in 50% inhibition
IEC	Independent Ethics Committee
IHC	Immunohistochemistry

IMP	Investigational Medicinal Product
IRB	Institutional Review Board
IV	Intravenous
Ki	Inhibition constant
KIT	V-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog
LC-MS/MS	HPLC coupled with tandem mass spectrometry
MedDRA	Medical Dictionary for Regulatory Activities
MTD	Maximum Tolerated Dose
NCI CTCAE	National Cancer Institute Common Terminology Criteria for Adverse Events
NMS	Nerviano Medical Sciences
NOAEL	No-observed-adverse-effect-level
NOEL	No-observed-effect-level
NSCLC	Non Small Cell Lung Cancer
OS	Overall Survival
P21	Cyclin-dependent kinase inhibitor 1A (CDKN1A)
P27	Cyclin-dependent kinase inhibitor 1B (CDKN1B)
P53	Tumor protein p53 (TP53)
P75	Nerve growth factor receptor (NGFR)
PCR	Polymerase Chain Reaction
PD	Progressive Disease
PFS	Progression-Free Survival
PFS-3	Progression-Free Survival at 3 months
PHA-848125AC	PHA-848125 maleate salt
PR	Partial Response
pRb	Retinoblastoma 1 (RB1)
PS	Performance Status
PT	Prothrombin time
PXR	Pregnane-X-Receptor
RP2D	Recommended Phase II Dose
SAE	Serious adverse event
SD	Stable Disease
SGOT/AST	Serum Glutamic-Oxaloacetic Transaminase
SGPT/ALT	Serum Glutamic Pyruvic Transaminase
SMRP	Soluble mesothelin-related proteins

$t_{1/2}$	Terminal half-life
TGI	Tumor Growth Inhibition
t_{max}	Time of occurrence of C_{max}
TRAMP	Transgenic Adenocarcinoma Mouse Prostate
TRKA	Thropomyosin Receptor Kinase A (NTRK1)
TYMS	Thymidylate synthase
ULN	Upper Limit of Normal
V_{ss}	Volume of distribution at steady-state
WBC	White Blood Cells
wks	Weeks
WHO	World Health Organization

3. BACKGROUND INFORMATION AND STUDY RATIONALE

3.1. Thymic Carcinoma

Thymic carcinomas are a category of heterogeneous thymic tumors with distinct molecular characteristics, the most aggressive behavior and poorest prognosis among thymomas [1]. Although rare, thymomas are nevertheless the most common neoplasms of the anterior mediastinal compartment, with an overall incidence in US (1973-1998) of 0.15 per 100000 person/years [2].
Patients with locally advanced or disseminated thymic malignancies are usually symptomatic, presenting with chest pain, shortness of breath, paralysis of the phrenic nerve, pleural effusion and superior vena cava syndrome. Immune disorders have also been associated with thymoma, the most common being myasthenia gravis [3].

There is no known cause for thymomas, however, in recent years, a few studies on molecular changes in thymomas and thymic carcinomas have been published. In particular, the expression of KIT and epidermal growth factor receptor (EGFR), targets, respectively, of imatinib and EGFR inhibitors (*ie*, erlotinib, gefitinib), have been tested by immunohistochemistry in a limited number of thymic malignancies: KIT was positive in 73% of thymic carcinomas and 5% of other thymomas, in contrast, EGFR expression was more often present in other thymomas (83%) than in thymic carcinomas (50%) [4]. Another study investigated HER2/neu amplification and expression of the apoptosis-related markers p53, BCL-2, BAX, BCL-XL, and p21 [5]; p53 and Bcl-2 were definitely more expressed in thymic carcinomas than thymomas and correlated with more advanced stages and unresectability, whereas no HER2/neu amplification was observed. Other markers, such as expression of thymidylate synthase and dihydropyrimidine dehydrogenase, which predict sensitivity to 5-fluorouracil-based chemotherapy, were not correlated with clinicopathological characteristics in a series of thymomas [6].

The loss of control of cell cycle checkpoints seems a common occurrence in thymomas, supporting the idea that functional cooperation between different cell cycle inhibitor proteins regulates cell growth control and tumor suppression. Univariate analyses on the expression of p21 and p27 in specimens from 25 encapsulated thymomas (using immunohistochemistry) suggest that negative expressions of p21 and p27 (natural inhibitors of CDKs) significantly correlates with poor prognosis for disease-free survival [7]. In addition, some members of the neurotrophin receptors family seem to play a significant role in this disease: in a study investigating the expression of neurotrophin receptors in thymic epithelial tumors on a quite large series of patients (N=99) [8], the pattern of TRKA expression was analyzed according to WHO classification for the histologic subtypes (A, AB, B1, B2, B3, C). All patients, except one, had immunostaining for TRKA expression in the tumors gradually increasing from type A to C. Conversely, none of the tumors showed TRKB or TRKC immunoreactivity. In addition, from type A to type C, there was a decreasing percentage of tumors with p75 immunostaining. These observations suggest a further elucidation on the specific role of TRKA and p75 in this disease.

Besides the above mentioned WHO histological classification of thymoma, the Masaoka staging system is commonly employed to evaluate invasiveness and to determine the therapy, since the optimal treatment for this disease depends on its clinical stage [1]. Locally advanced or metastatic thymomas are often treated with combined treatment modalities, including surgery, radiation and chemotherapy. There are no drugs officially approved for the treatment of thymoma and thymic carcinoma. However, several antineoplastic agents are used. Cisplatin/doxorubicin-based combination chemotherapy [PAC regimen (cisplatin, doxorubicin, cyclophosphamide) or ADOC regimen (doxorubicin, cisplatin, vincristine, cyclophosphamide)] seem to produce the best overall response rate and survival. Other combined and /or single agent chemotherapy with cisplatin, etoposide, ifosfamide, epirubicin, maytansine, octreotide and steroids are used as well [9-11]. Examining treatments for patients with advanced disease who had received one or more prior lines of therapy, published data generally indicates a limited activity, in fact single agent studies with gefitinib, imatinib or a combination study erlotinib+bevacizumab did not produce satisfactory results [12-14], although only two cases of activity of possible clinical benefit were observed with imatinib and sorafenib, due to the rare incidence of KIT mutations in those patients' tumors [15,16]. An optimal treatment strategy has yet to be determined and other drugs are warranted to improve the outcome of patients with advanced invasive tumors [10]. The unmet medical need for new agents for the treatment of thymic malignancies is therefore high, as demonstrated by the number of studies with experimental agents currently in progress.

Given the rarity of the thymic carcinoma subtype, prospective phase II studies of chemotherapy specifically targeting thymic carcinoma patients are rare. Combined therapies such as ADOC or VIP (cisplatin, ifosfamide, etoposide) achieved, as a first line treatment for the advanced disease, a median survival time around 19-20 months [17, 18]. In previously untreated, unresectable thymic carcinoma, first-line combination of carboplatin and paclitaxel achieved an overall response rate of 36% with a median survival time of 22.7 months and a median progression-free survival of 7.9 months [19].

In a single agent study with pemetrexed, on 23 previously treated evaluable patients with unresectable stage IV A and B disease, there were 4 objective responses (2 CRs and 2 PRs) (17% RR). Responses were obtained only in stage IVA thymoma patients, with a median duration of time to progression of 45.4 weeks; in thymic carcinoma patients the median duration of time to progression was much shorter (5.1 weeks) [20]. In other two recent series of previously treated patients with thymic malignancies, objective responses were reported in B2 and B2/3 tumors (no type C thymic carcinoma patients were enrolled in the first study) with a gemcitabine/ capecitabine combination [21] and with the histone deacetylase inhibitor (HDAC) belinostat [22]. Also in this second study, responses were obtained only in thymoma patients and no responses were seen in the 8 evaluable patients with thymic carcinoma. Thymic carcinoma is therefore confirmed as a type of cancer where prognosis is particularly poor and the course of the disease more aggressive compared to the other thymomas.

Given the current clinical scenario, in thymic carcinoma patients previously treated with chemotherapy, a single agent therapy is able to induce a median PFS of approximately 5.1 weeks [20], roughly corresponding to an estimated PFS rate at 3 months of about 17%. Therefore a 33% target PFS-rate at three months could be regarded as clinically interesting for a new investigational agent.

3.2. PHA-848125AC

3.2.1. Description

PHA-848125, N,1,4,4-tetramethyl-8-{[4-(4-methylpiperazin-1-yl)phenyl]amino}-4,5-dihydro-1H- pyrazolo[4,3-h]quinazoline-3-carboxamide, is a potent inhibitor of the kinase activity of the CDK2/Cyclin A complex (Ki = 32 nM), showing activity also towards closely related CDKs (ie, CDK1, CDK4, and CDK5) and TRKA.

The compound is formulated for oral administration as maleate salt (PHA-848125AC). Throughout this document, "PHA-848125" indicates the active compound as free base, which was used under various salt forms in preclinical studies.

Details regarding the results of the pre-clinical and clinical studies are contained in the PHA-848125AC Investigator Brochure, Version 7, 2011 [23].

3.2.2. Pharmacology

CDKs are serine/threonine kinases that, in concert with their activators (cyclins) and negative regulators, play a crucial role in the cell cycle progression [24,25]. CDK/Cyclin complexes regulate the phosphorylation and inhibitory binding of the pRb to members of the E2F transcription factors family, that are essential for the transcriptional regulation of a number of genes whose products control cell cycle progression. These include genes essential for the progression from the G1 into the S phase of the cell cycle and genes that are involved in the regulation of DNA replication.
Deregulation of CDKs activity, alterations of expression and/or genetic mutations of cyclins, CDKs, Cyclin Dependent Kinase Inibitors (CDKIs) and other components of the pRb pathway have been reported in more than 90% of human neoplasms [26]. For example, CDK2 activators, such as Cyclin E2 and A2 are found overexpressed in 50% of breast and lung cancer, and decreased levels of their natural inhibitors (*ie*, p21, p27) predict a poor prognosis in breast, prostate, colon, gastric, lung and esophageal cancer. Indeed, alterations in the Rb pathway lead to release of E2F transcription factors, high commitment to cell cycle progression and, ultimately, in uncontrolled proliferation. These findings strongly support a pharmacological inhibition of CDKs as an attractive strategy in the treatment of human cancers [27-32].

In fact, according to the postulated mechanism of action, as determined in biochemical assays, in *in vitro* experiments PHA-848125 was able to cause a block in G1 phase of the

cell cycle in tumor cells: it decreased the phosphorylation level of the Retinoblastoma protein (pRB), by inhibiting the CDK/Cyclin complexes, thus preserving the E2F transcription factors in an inactive state.
Additionally, PHA-848125 reduced the phosphorylation status of TRKA and of the proteins of the TRKA signaling pathway in cells expressing this receptor tyrosine kinase.

The antiproliferative effect of PHA-848125 was tested on a panel of 120 tumoral cell lines established from different solid tumors (ovary, breast, pancreas, lung, cervix, prostate, skin, brain, bone), leukemias and lymphomas. The IC50 values were in the range of 0.1-1 μM for 61 cell lines and in the range of 1-3 μM for 42 cell lines, indicating a broad spectrum of activity of the compound. Only 17 cell lines were poorly responsive to PHA-848125 with an IC50 value > 3 μM.

Significant anti-tumor activity was observed in all tested preclinical animal models with different oral treatment schedules of PHA-848125. In various human xenograft models, as well as in the TRAMP model, consistent tumor growth inhibition (up to 91%), was reported with a repeat daily treatment at tolerated doses (40 mg/kg bid). In the rat DMBA (9,10-dimethyl-1,2-benzanthracene)-induced mammary carcinoma model, comparable results (stasis and partial remission in 58 and 25% of the primary tumors, respectively) were obtained with repeat daily treatment at tolerated doses (up to 15 mg/kg bid). In the same model, intermittent treatment with a rest period was equally effective.
Inhibition of pRb phosphorylation status and modulation of the expression of genes regulated by E2F transcription factors (ie, cyclins and histones) were measured at mRNA and/or protein level, in a dose-dependent manner both in skin and tumors of mice treated with PHA-848125.

A statistically significant increase in survival time was obtained in two disseminated human leukemia models (AML and ALL) and in a human glioma model implanted intracranially.

In combination studies, PHA-848125 exhibited synergistic or more than additive activity when administered with docetaxel, topotecan or temozolomide, and additive effect when combined with bevacizumab, irinotecan, 5-FU and gemcitabine. Combinations were well tolerated.

3.2.3. Safety Pharmacology

Safety pharmacology studies were performed to investigate the effects of PHA-848125 on central nervous (CNS), respiratory and cardiovascular systems (all doses mentioned are expressed as free base).

Central Nervous System: no effects on general behavior (Irwin's test) and body temperature were observed in rats up to the highest single dose of 160 mg/kg and no alterations in the EEG were detected in adult rats given PHA-848125 for 5 consecutive days at 16 or 32 mg/kg/day.

Respiratory System: in a respiratory function study in rats, a dose-related shortening of the inspiratory time and an increase in peak inspiratory flow were observed from the dose of 80 mg/kg with recovery within 4 hours. The dose of 40 mg/kg was considered the NOEL.

Cardiovascular System: cardiovascular safety pharmacology assessments included *in vitro* evaluation for potential effects on cardiac ion channels (I_{kr}) and *in vivo* evaluations for effects on cardiovascular parameters and body temperature in conscious beagle dogs. A dose-related *in vitro* inhibition of the I_{kr} channel occurred from the concentration of 0.1 μM with a calculated IC_{50} of 1.32 μM (the IC_{50} for the antiproliferative activity in the A2780 cell line was 0.2 μM). In a telemetry study in dogs, a temporary moderate decrease in heart rate was observed from 0.5 to 3 h after treatment at the dose of 32 mg/kg. QT/QTc interval was not affected at any of the tested doses. No effect was seen at 16 mg/kg; this dose was considered the NOEL for cardiovascular function. No changes in ECG parameters (including QT and QTc) were observed in single and repeat-dose toxicity studies in dogs and monkeys.

3.2.4. Nonclinical Pharmacokinetics

Pharmacokinetics of PHA-848125 was investigated in the mouse, rat, dog and monkey after single IV and oral administration. Since the compound is intended for oral route administration, its pharmacokinetics was further studied after repeated oral administrations.

Following IV administration, PHA-848125 was characterized by moderate clearance in mice, rats and monkeys and high clearance in dogs. The clearance was mainly non renal as suggested by the very limited renal excretion of the unchanged drug. The volume of distribution was higher than the total body water in all the preclinical species, suggesting an extensive tissue distribution of the compound.

The results of the rat tissue distribution study conducted with [^{14}C]-PHA848125 indicated that apart from the GI tract, the highest levels of radioactivity were measured in the liver, spleen, skin, kidneys, adrenal glands and lachrymal glands. The blood to plasma ratio was between 0.8-1.12 indicating a moderate distribution of the radioactivity into the red blood cells. In rats and monkeys PHA-848125 distributed in the brain. Moreover, the study carried out in the rat using [^{14}C]-PHA848125, indicated that the distribution of the radioactivity was homogeneous in the different brain areas and cerebellum.

The *in vitro* plasma protein binding ranged from 78 to 90%, depending on the species.

The bioavailability of PHA-848125 after oral administration was equal or higher than 30 % in all the preclinical species. PHA-848125 plasma levels increased largely in direct proportion with the dose. No relevant gender differences were observed in the preclinical species.

In dogs, the extent and rate of absorption of PHA-848125 was unaffected when the compound was administered in hard gelatin capsules or in 5% dextrose solution. No food effect was observed.

The elimination profile of total radioactivity following a single oral administration of [^{14}C]-PHA-848125AC in rats suggested that the main route of elimination was *via* faeces, accounting for about 70 % of the dose within 96 hours post dose. The urinary excretion of the radioactivity represented 16% of the dose. In all preclinical species, after IV and oral treatment, the amount of parent compound excreted unchanged in urine was less than 6% of the administered dose. In the rat treated by oral route, the excretion of PHA-848125 in bile accounted for about 15% of the administered dose. Overall, the excretion data suggested that the compound was mainly eliminated via metabolic transformations.

Metabolism studies (*in vitro* and *in vivo*) showed that the major route of metabolism of PHA-848125 was the N-oxidation of the N-methyl piperazine moiety to give the metabolite M1 (NMS-867734). More than one enzymatic pathway is involved in the metabolism of this compound, decreasing the risk for unwanted clinical relevant drug-drug interactions. From a qualitative point of view no species differences were observed in the metabolism of PHA-848125 and no unique metabolite was detected in human microsomes. Given these results, both monkeys and dogs represented suitable nonrodent species for the conduct of repeat-dose toxicological studies. The plasma profile of NMS-867734 was investigated in rats and monkeys showing a metabolite/parent AUC ratio of 0.2-0.4 and 0.05, respectively.

PHA-848125 did not activate human and mouse PXR, suggesting that the compound is unlikely to increase the CYP3A mediated enzyme activities by increasing its gene transcription. PHA-848125 showed some competitive inhibition on CYP3A4 (Ki = 2.4 μM and 18.7 μM using triazolam and testosterone as substrate, respectively) at concentrations much higher than those needed for the pharmacological activity (Ki = 32 nM for CDK2/Cyclin A complex).

3.2.5. Toxicology

PHA-848125 was characterized in single and repeat-dose toxicity studies in rats, dogs and monkeys. PHA-848125 was also characterized by *in vitro* genotoxicity and dermal and ocular irritation studies.

Single-dose toxicity studies were carried out administering PHA-848125 by oral route in rats, dogs and monkeys. Effects related to the mechanism of action of the compound were seen in the hemolymphopoietic (HLP) system, the gastrointestinal (GI) tract and the male reproductive organs in all species. Regression was observed at the end of the 2-week recovery period for all the changes except for the testes, as expected considering the time of maturation of the seminiferous epithelium. Additional toxicities, apparently not related to the mechanism of action of the compound, were the vascular effects (congestion and hemorrhages in several organs) and the CNS toxicity observed in dogs.

In rats, no mortality was observed up to the highest administered single dose of 160 mg/kg. The dose of 40 mg/kg (mean AUC = 69 μM·h) was the NOAEL in rats after single oral administration.

In dogs, one female given the highest dose of 69.1 mg/kg was sacrificed 9 h after treatment due to the severe clinical signs of CNS toxicity (tremors, incoordination, convulsions). In this animal, the AUC(0-8h) measured before death was 31.69 μM·h. In addition, this dog showed hemorrhages in several organs and multifocal edema in the brain. The dose of 34.5 mg/kg (mean AUC = 17 μM·h) was the MTD in dogs after single oral administration.

In monkeys, no mortality or significant signs of toxicity were observed after the administration of single oral doses up to 69.1 mg/kg (mean AUC = 31 μM·h).

Repeat-dose toxicity studies were conducted with PHA-848125 administered by oral route daily for 4/7 days, 4 weeks or 3 months period. Noteworthy effects of PHA-848125 in the repeat-dose toxicity studies included changes in the HLP system, GI tract and male reproductive organs in all species. Effects on female reproductive organs were also seen in rats and dogs. The effects on the HLP system were dose-related and were observed at all doses tested. In dogs, GI toxicity was seen at all doses with dose-relationship independently of the treatment schedule. All the above changes recovered after drug withdrawal, with the exception of the effects on the male reproductive system, which were still present at the end of the recovery period.

Additional toxicities, that are considered not related to the mechanism of action of the compound, were CNS, ocular and renal toxicities. In addition, hemorrhages in different organs were observed in dogs and monkeys

Clinical signs of CNS toxicity, characterized by tremors, increased reactivity, incoordination and convulsions were noted at high doses given as single administration in dogs or repeated administrations in rats and monkeys. This effect, observed at daily AUC ≥ 30 μM·h, recovered after drug withdrawal and was not supported by any morphological change in the brain.

Ocular toxicity was observed in rats after prolonged administration (≥ 1 month) of PHA-848125. Bilateral retinal atrophy occurred from the dose of 16 mg/kg/day in females (daily AUC = 27 μM·h) and at 24 mg/kg/day in males (daily AUC = 38 μM·h) in the 4-week study. The same finding was seen in rats given the dose of 16 mg/kg/day for 3 cycles of 3 weeks + 1 week rest or 5 cycles of 2 weeks + 1 week rest (mean daily AUC from 25.5 to 34.5 μM·h). This effect did not regress after the 2- or 4-week recovery periods. The dose of 8 mg/kg/day, given for 4 consecutive weeks or cyclically for 3 months, corresponding to mean daily AUC values of 9.8 and 11.7 μM·h, respectively, was the NOEL for ocular changes. No retinal changes were seen in rats following administration of PHA-848125 for 7 consecutive days up to the highest dose (51.8 mg/kg/day) or a daily AUC of about 50 μM·h.

Other effects, observed at lethal doses, involved the kidneys (cortical tubular nephropathy) in dogs at the dose of 8 mg/kg/day administered as two 2-week cycles (daily AUC 15 μM·h) and at 12 mg/kg/day as two 1-week cycles (daily AUC ≥ 15 μM·h). Some changes were also seen in monkeys at the toxic dose of 15.97 mg/kg/day given for 7 days (daily AUC 71 μM·h in the only surviving animal). Renal changes did not completely regress at the end of the recovery period.

Congestions and hemorrhages in different organs occurred following repeated administrations of PHA-848125 at high toxic lethal doses in dogs (34.5 mg/kg/day for 4 days; daily AUC ≥ 27 μM·h) and in monkeys (15.97 mg/kg/day for 4 and 7 consecutive days; daily AUC > 30 μM·h).

PHA-848125 was not genotoxic in the Ames test and did not induce any structural or numerical chromosome aberrations in human peripheral blood lymphocytes *in vitro*.

In the dermal and ocular irritation tests, PHA-848125 was not irritating to the skin of rabbits, while it induced severe irritant reactions when applied to the eye of rabbits for 1 h. A complete recovery occurred within 10-16 days.

3.2.6. Drug-Drug Interactions

As detailed in Section 3.2.4, more than one enzymatic pathway is involved in the metabolism of PHA-848125AC, decreasing the risk for unwanted clinical relevant drug-drug interactions. Also, PHA-848125 did not activate human and mouse PXR, suggesting that the compound is unlikely to increase the CYP3A mediated enzyme activities by increasing its gene transcription.

Significant drug-drug-interactions are therefore not expected.

3.2.7. Phase I clinical trials

The phase I clinical program of PHA-848125AC as a single agent is ongoing with three studies testing four different schedules (daily x 7 every 2 weeks; 4 days on / 3 days off x 3 weeks every 4 weeks; daily x 21 days every 4 weeks; daily x 14 days every 3 weeks), being conducted in US and Europe. Both a flat dose approach and a BSA-based approach have been investigated.

The clinical experience gained so far (July 2009) on overall 90 patients treated for 277 cycles indicated that the safety profile of the compound is characterized by a dose-limiting neurological toxicity and, to a lesser extent, by gastrointestinal toxicity. Nausea and/or vomiting and /or diarrhea were mostly of grade 1- 2 in severity and were manageable with appropriate therapy, including hydration. Haematological toxicity, mainly represented by neutrophils decrease, was reported in some patients only. A prolonged transaminitis was occasionally observed with the 7 days on / 7 days off schedule, however, with a treatment

schedule encompassing 14 consecutive days every 3 weeks, alterations of liver function tests were more frequently reported (starting from the dose of 54 mg/m^2/day for 14 days). Ocular toxicity was never encountered so far, with the exception of a worsening of ERG during treatment compared to baseline in one diabetic patient treated at 24 mg/m^2/day for 21 consecutive days and in a second patient treated at 72 mg/m^2/day for 14 consecutive days every 3 weeks (dose then reduced to 48 mg/m^2/day in the two subsequent cycles). Both patients, however, did not show any clinical symptomatology.
The neurological toxicity consisted in tremors and ataxia. In study CDKO-125a-001, where the same schedule of administration proposed in the present study was studied (*ie* 7 days on /7 days off, flat dose) and which therefore represents the best reference study for this compound, on 22 patients treated for 112 cycles, mild, short-lasting tremors were observed, starting from the dose of 100 mg/day and increasing in frequency and in severity at 200 mg/day and 300 mg/day. Grade 3 ataxia was repeatedly observed starting from the dose of 200 mg/day, where a mean Day 7 $AUC_{(0-24h)}$ of 29.2 μM·h was reported, but occurred also in one patient (grade 2) after 7 cycles of treatment at 150 mg/day (in this patient treatment was continued at a reduced dose of 100 mg/day, with no further episodes of the event). Tremors and ataxia were reversible in all cases in up to 7-8 days, upon drug discontinuation. The MTD with the 7 days on /7 days off schedule was attained at the dose of 200 mg/day. The Phase II recommended dose (RP2D) with this schedule of administration was identified in 150 mg/day. At the RP2D, where a mean Day 7 $AUC_{(0-24h)}$ of 25.3 μM·h was measured, only tremors (up to grade 2) were reported, plus the above mentioned grade 2 ataxia after 7 treatment cycles. Overall, at this dose level, toxicity consisted mainly of mild/moderate nausea, fatigue, diarrhea and tremors, all effects easily manageable and reversible.
In the same study, for the patients treated under the second schedule (4 days on / 3 days off x 3 weeks every 4 weeks, flat dose) ataxia was never reported, but grade 1 tremors were common findings at doses below 200 mg/day, reaching grade 2-3 in severity and qualifying for DLTs at 200 mg/day (mean Day 18 $AUC_{(0-24h)}$ of 33.3 μM·h).

As for efficacy, one out of three thymic carcinoma patients treated at the dose of 150 mg/day (4 days on /3 days off for 3 weeks every 4 weeks) showed a confirmed partial response after 11 treatment cycles (the patient is on study for 22 cycles [22 months] at 150 mg/day at the time of writing this document), and another patient showed an unconfirmed partial response after 7 cycles of treatment (the patient is on study for 7+ cycles [7+ months] at 150 mg/day at the time of writing this document). The third patient was taken off study after 2 cycles for disease progression. In addition, a stable disease for more than 4 months was reported in a NSCLC patient (on study for 12 cycles at 200 mg/day, dose reduced to 100 mg/day since cycle 10, 7 days on /7 days off schedule) and another stable disease for more than 12 months was observed in a colorectal cancer patient (on study for 27 cycles at 150 mg/day, dose reduced to 100 mg/day since cycle 8, 7 days on /7 days off schedule).

3.2.8. Pharmacokinetics in Humans

The pharmacokinetics of PHA-848125AC and its major metabolite, NMS-867734 (M1), has been investigated in the three ongoing phase I clinical trials CDKO-125a-001, CDKO-125a-

002, CDKO-125a-003. Results for PHA-848125AC are summarized in Table 1, Table 2 and Table 3, respectively.

In particular, in the CDKO-125a-001 study, first schedule, which is the same schedule adopted in the present study, the pharmacokinetic profile of PHA-848125AC was characterized by:

- achievement of maximal plasma concentration at 2 - 4 hours post dosing, after single and repeated administration
- terminal half-life in the range of 30 and 40 hours
- good dose proportionality in systemic exposure, in the range of 50-300 mg/day
- similar exposure to the compound after subsequent cycles of treatment
- inter-patient variability limited
- AUC of the major metabolite lower than that of the parent compound, accounting for 30-50%. Its half-life is comparable to or slightly longer than that of the parent compound.

Of note, at the RP2D of 150 mg/day, which is the dose to be administered to patients in the present study:

- on day 1 in cycle 1, maximal plasma concentration was 0.66±0.24 μM (n=6), attained on average at 2.5 h post-dosing. The corresponding daily AUC was 8.86±2.64 μM·h (n=6).
- after seven days of treatment in cycle 1, maximal plasma concentration was 1.47±0.51 μM (n=6), attained on average at 2.5 h post-dosing and corresponding to a daily AUC of 25.31±8.56 μM·h (n=6).
- the day 7/day 1 $AUC_{(0\text{-}24h)}$ ratio was around 3, in line with what was expected considering the schedule and the half-life of the compound that at this dose level was on average 33.3 hours.

Overall, at similar doses administered with different schedules, comparable PHA-848125AC PK profile was observed across studies.

For more details on PHA-848125AC, please refer to Investigator Brochure for PHA 848125AC, Version 7, 2011 [23].

Table 1. PHA-848125AC pharmacokinetic data from CDKO-125a-001 study

CDKO-125a-001 first schedule (7 days on, 7 days off q2wks)						
Day	**Dose (mg/day)**	**No. of Pts**	**Cmax (μM)**	**AUC0-t(24h) (μM· h)**	**tmax (h)**	**t1/2,λz (h)**
1	50	3	0.2±0.06	2.56±0.52	3.35±1.17	N/A
7	50	3	0.4±0.15	6.84±2.21	3.33±1.15	32.8±12.2
1	100	6	0.44±0.07	6.19±2.26	3.5±2.51	N/A
7	100	6	1.2±0.47	19.9±10.5	2.76±1.89	38.2±9.28
1	150	6	0.66±0.24	8.86±2.64	2.5±1.22	N/A
7	150	6	1.47±0.51	25.3±8.54	2.5±1.22	33.3±7.08
1	200	6	0.56±0.11	8.41±1.68	4±1.26	N/A
7	200	6	1.68±0.17	29.2±4.5	4.33±1.51	31.8±4.68
1	300	1	1.15	22.3	2	N/A
7	300	1	5.12	83.6	4	31.3
CDKO-125a-001 second schedule (4 days on, 3 days off x 3wks q4wks)						
Day	**Dose (mg/day)**	**No. of Pts**	**Cmax (μM)**	**AUC0-t(24h) (μM· h)**	**tmax (h)**	**t1/2,λz (h)**
1	150	3	0.42±0.1	6.67±1.72	4±0	N/A
18	150	3	1.08±0.07	19.5±4.27	3.13±1.96	41±3.01
1	180	1	0.55	6.67±1.72	2	N/A
18	180	1	1.22 (Day 4)	6.67±1.72	4	--
1	200	4	0.79±0.5	9.58±7.34	2±0	N/A
18	200	3	2.33±0.58	33.3±2.56	1.71±0.44	25.9±8.38
N/A: not applicable						

Table 2. PHA-848125AC pharmacokinetic data from CDKO-125a-002 study

CDKO-125a-002 (14 days on, 7 days off q3wks)						
Day	**Dose (mg/m^2/day)**	**No. of Pts**	**Cmax (μM)**	**AUC0-t(24h) (μM· h)**	**tmax (h)**	**t1/2,λz (h)**
1	18	5	0.103±0.042	1.7±0.487	4.8±1.09	N/A
14	18	4	0.248±0.064	4.08±1.23	2.5±1	40.1±6.68
N/A: not applicable						

Table 3. PHA-848125AC pharmacokinetic data from CDKO-125a-003 study

CDKO-125a-003 first schedule (21 days on, 7 days off q4wks)						
Day	**Dose (mg/m^2/day)**	**No. of Pts**	**Cmax (μM)**	**AUC0-t(24h) (μM· h)**	**tmax (h)**	**t1/2,λz (h)**
1	16	7	0.22±0.28	2.54±2.92	2.86±1.57	N/A
21	16	6	0.25±0.08	4.07±1.29	2.96±1.15	33.6±10
1	24	6	0.2±0.09	2.48±0.92	1.78±1.26	N/A
21	24	5	0.42±0.2	6.24±2.44	2.43±2.05	37.1±5.77
CDKO-125a-003 second schedule (14 days on, 7 days off q3wks)						
Day	**Dose (mg/m^2/day)**	**No. of Pts**	**Cmax (μM)**	**AUC0-t(24h) (μM· h)**	**tmax (h)**	**t1/2,λz (h)**
1	24	3	0.18±0.05	2.32±0.54	2.3±1.5	N/A
14	24	3	0.45±0.22	7.1±3.94	1.7±0.6	41.2±4.26
1	48	3	0.31±0.09	4.5±1.43	2.8±1.4	N/A
14	48	3	0.7±0.44	1.27±7.9	3.3±1.2	28.9±1.6
N/A: not applicable						

3.3. Rationale for the use of PHA-848125AC in Thymic Carcinoma

Chemotherapy was shown to have significant antitumor activity against unresectable, recurrent or metastatic thymomas and thymic carcinomas, but an optimal treatment strategy has not been determined and other drugs are needed to improve the outcome of patients with advanced invasive tumors [10].

In recent years, a few studies on molecular changes in thymomas point to alterations that may be of use in selecting patients for targeted therapies [4-8]. Unfortunately for some of them, such as KIT and EGFR inhibitors, the reported results seem to be discouraging [12-14]. In addition, despite a lack of correlation of thymidilate synthase expression with thymoma clinicopathological characteristics, pemetrexed obtained interesting responses in stage IVA thymomas, but not in thymic carcinomas [20].

The pyrazoloquinazoline PHA-848125 is a compound counteracting the effects of some of the other so far identified, but not yet exploited, molecular alterations found in thymoma and thymic carcinoma, e.g. underexpressed p21 and p27 and overexpressed TRKA [7,8]. These targets are not yet been investigated in therapeutic intent clinical trials.

In fact, p21 and p27 encode natural potent cyclin-dependent kinase inhibitors and their ablation in the cancer cell results in an activation of cyclin E/CDK2 or cyclin D/CDK4 complexes and thus in uncontrolled cell cycle progression and cell proliferation [24-32], while TRKA activation promotes tumor growth [33], probably through MAPK pathway activation. PHA-848125 potently inhibits both the CDK/Cyclin complexes and TRKA,

proposing in this way to be a "selective dual inhibitor" and appearing particularly attractive in this disease.

The preliminary results in early clinical trials on thymic carcinomas patients (2 out of 3 patients with clinical benefit) support the hypothesis that PHA-848125 could be of therapeutic value for patients with this tumor type.

An exploratory analysis on the relationship of molecular features of p53 (natural regulator of p21), p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action with treatment efficacy variables will be possibly performed in tumor biopsies of consenting patients obtained before study entry.

4. TRIAL OBJECTIVES AND ENDPOINTS

4.1. Objectives

4.1.1. Primary Objective

- Assessment of the antitumor activity of PHA-848125AC as second-line treatment in patients with recurrent or metastatic, unresectable thymic carcinoma previously treated with chemotherapy. Antitumor activity will be evaluated on the basis of the progression-free survival status at 3 months.

4.1.2. Secondary Objectives

- Assessment of additional measures of tumor control to further characterize the efficacy profile of PHA-848125AC in recurrent or metastatic, unresectable thymic carcinoma previously treated with chemotherapy.
- Evaluation of the safety profile of repeated administrations of PHA-848125AC in patients with recurrent or metastatic, unresectable thymic carcinoma previously treated with chemotherapy.

4.1.3. Exploratory Objective

- Relationship of baseline molecular features in tumor biopsies of p53, p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action with treatment efficacy.

4.2. Endpoints

4.2.1. Primary Endpoint

- Progression-free survival rate at 3 months (PFS-3 rate). The PFS-3 rate, will be calculated as the proportion of evaluable patients (see Section 12.2) known to be alive

and progression-free at ≥ 3 months since study treatment start out of the total number of evaluable patients.

4.2.2. Secondary Endpoints

- Confirmed Objective Response Rate (CR + PR) according to RECIST guideline (version 1.1).
- Disease Control Rate (Confirmed Objective Response Rate + ≥ 6 weeks SD rate).
- Progression-free survival, calculated as the time from the date of treatment start to the date of first documentation of objective progression or of death due to any cause, whichever comes first.
- Duration of Response, measured from the time measurement criteria are first met for CR/PR (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded on study).
- Overall Survival (OS), i.e. the time from the date of treatment start to the date of death from any cause.
- Overall safety profile, evaluated on the basis of laboratory and clinical safety parameters (i.e. hematology and blood chemistry, urinalysis, vital signs, ophthalmologic examinations and adverse events emerging during the trial). The National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE) Version 3.0 will be used for the severity grading of adverse events and of hematological and blood chemistry abnormalities.

4.2.3. Exploratory Endpoint

- Baseline characterization of selected biomarkers (p53, p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action) in tumor tissue of consenting patients. Assessments will be done on paraffin embedded blocks, obtained from all patients before study entry, by IHC and other possible techniques, such as FISH and PCR.

5. TRIAL DESIGN AND DESIGN RATIONALE

In consideration of the exploratory nature of the study, the Simon's optimal 2 stage design is adopted for this single-arm, open-label, multicentre phase II clinical trial of PHA-848125AC administered to patients with recurrent or metastatic, unresectable thymic carcinoma previously treated with chemotherapy (only one prior systemic therapy allowed).

The intent of the study is to assess the antitumor activity of PHA-848125AC and ultimately to improve the outcome of patients with thymic carcinoma who have already exploited one chemotherapy option. To this purpose, the progression free survival rate at 3 months is considered to be an appropriate primary endpoint.

Patients will receive repeated cycles of PHA-848125AC administered orally once daily at the flat dose of 150 mg/day, according to a 7 days on / 7 days off schedule in 2-week cycles. A treatment cycle will therefore comprise 7 days of PHA-848125AC administration (Days 1 to 7) followed by 7 days of rest (Days 8 to 14) for a total of a 14 days period (2-week cycle).

The 150 mg/day dose has been selected based on the results of the first single agent Phase I study in solid tumor patients (CDKO-125a-001), where PHA-848125AC was administered with the same schedule, at escalating dose levels, ranging from 50 to 300 mg/day: the dose of 150 mg/day was assessed as the dose recommended for Phase II investigations (RP2D); this dose was associated with mild/moderate reversible adverse events, mostly represented by maximum grade 2 nausea and/or vomiting and/or diarrhea and maximum grade 2 neurological effects.

Patients may continue on study treatment until disease progression, patient refusal or withdrawal of patient consent, or the occurrence of unacceptable toxicity and will be followed up for survival up to the end of the study and in any case for no more than 2 years from the end of treatment.

Patients remaining on treatment longer than 6 cycles of treatment are allowed to come back to the site once every 3 cycles (once every 1.5 month) for safety evaluation, drug dispensing, oncologic assessment and ocular examination. At the end of each cycle instead of the site visit the patient is required to perform the laboratory tests locally. Also a phone interview will be performed by the Investigator to clarify the toxicity experienced by the patient during the observation period. The Investigator must document in writing the results of the phone call with the patient in the medical chart. Data are to be reported in the CRF.
Only after evaluation of the laboratories' results and of the patient's safety performed by phone, the Investigator can decide if the patient may continue with the treatment. Otherwise, if the Investigator deems necessary to physically visit the patient (especially for the evaluation of his/her neurological status) or he has any doubt about the capability of the self-evaluation for a specific patient, it is left to the Investigator's discretion the possibility to require that the patient reaches the site before continuing the treatment.

A cut-off date is planned on 31 May 2017 in order to proceed with clinical database closure and preparation of the clinical study report.

The management of patients after the cut-off date of 31 May 2017 should be conducted as per Section 9.5.

The total number of evaluable patients for the primary efficacy analysis ranges from 17 (if the trial stops at the end of the 1st stage) to 54 (if the trial proceeds up to the completion of the 2nd stage). Accounting for a 10%-15% proportion of non evaluable patients, up to 20 patients could be required in the 1st stage of the study and up to 60 patients could be required for completing the trial (1st and 2nd stage).

For statistical considerations, see Section 12.

5.1. Confirmation of histological diagnosis

Histological confirmation of thymic carcinoma for all patients will be obtained by an Independent Review Committee during the course of the study, based on paraffin embedded tumor tissue slides retrieved from the patients.

6. SUBJECT SELECTION

This clinical trial can fulfill its objectives only if appropriate subjects are enrolled. The following eligibility criteria are designed to select subjects for whom protocol treatment is considered appropriate. All relevant medical and non-medical conditions should be taken into consideration when deciding whether this protocol is suitable for a particular subject. Eligibility criteria may not be waived by the investigator and are subject to review in the case of a Good Clinical Practices (GCP) or a regulatory authority audit. Any questions regarding a subject's eligibility should be discussed with NMS Study Management prior to enrollment.

6.1. Subject Inclusion Criteria

Subjects must meet all of the following inclusion criteria to be eligible for enrollment into the study:

1. Signed and dated IRB/IEC-approved Informed Consent.
2. Histologically or cytologically proven diagnosis of unresectable B3 thymoma or thymic carcinoma recurrent or progressing after prior chemotherapy (only one prior systemic therapy allowed).
3. Presence of measurable disease defined as at least one lesion that can be accurately measured by CT scan in at least one dimension, as > 10 mm for non nodal lesions (longest diameter to be recorded) and ≥ 15 mm for lymph nodal lesions (short axis to be recorded). (CT scan is the desirable method for lesion measurement. Other measurement techniques [*eg* MRI] are acceptable [but not for lung lesions] provided that the size of the measurable lesion is twice the slice thickness of the MRI).
   Tumor lesions situated in a previously irradiated area, or in an area subjected to other loco-regional therapy, are usually not considered measurable unless there has been demonstrated progression in the lesion.
4. Age ≥ 18 years.
5. ECOG performance status 0-1.
6. Estimated life expectancy of at least 3 months.
7. Negative pregnancy test (if female in reproductive years).

8. Agreement upon the use of effective contraceptive methods (hormonal or barrier method of birth control, or abstinence) prior to study entry and for the duration of study participation, if men and women of child producing potential.
9. Adequate liver function:
   - Total Serum Bilirubin ≤ 1.5 x upper limit of normal (ULN)
   - Transaminases (AST/ALT) ≤ 2.5 ULN (if liver metastases are present, then ≤ 5 ULN is allowed)
   - ALP ≤ 2.5 ULN (if liver and/or bone metastases are present, then ≤ 5 ULN is allowed).
10. Adequate renal function:
    - Serum Creatinine ≤ ULN
      or
      Creatinine Clearance calculated by Cockcroft and Gault's formula > 60 mL/min.
11. Adequate hematologic status:
    - ANC ≥ 1,500 cells/mm^3
    - Platelet Count ≥ 100,000 cells/mm^3
    - Hemoglobin ≥ 9.0 g/dL.
12. At the time of start of treatment, at least 2 weeks must have elapsed since completion of prior chemotherapy, minor surgery, radiotherapy (provided that no more than 25% of bone marrow reserve has been irradiated).
13. With the exception of alopecia, resolution of all acute toxic effects of any prior chemotherapy, surgery or radiotherapy to NCI CTC (Version 3.0) grade ≤ 1 and to the baseline laboratory values as defined in Inclusion Criteria Number 9, 10, 11.
14. Able and willing to comply with scheduled visits, therapy plans, and laboratory tests required in this protocol.
15. Capability to swallow capsules intact (without chewing, crushing, or opening).

6.2. Subject Exclusion Criteria

The presence of any of the following will exclude a subject from study enrollment:

1. Any of the following in the past 6 months: myocardial infarction, uncontrolled cardiac arrhythmia, unstable angina, coronary/peripheral artery bypass graft, symptomatic congestive heart failure, cerebrovascular accident or transient ischemic attack, pulmonary embolism, deep vein thrombosis.
2. Grade >1 retinopathy as determined by an ophthalmologist.
3. Known brain metastases.
4. Major surgery, other than diagnostic surgery, within 4 weeks prior to treatment.
5. Active, uncontrolled bacterial, viral, or fungal infections.
6. Known infection with HIV, active hepatitis B or hepatitis C.
7. Pregnant or breast feeding women.
8. Previous (within the last 5 years) or current malignancies at other sites, except for adequately treated basal cell or squamous cell skin cancer or in situ carcinoma of the cervix uteri.
9. Current enrollment in or participation in another therapeutic clinical trial within 4 weeks preceding treatment start.
10. Diabetes mellitus uncontrolled.
11. Gastrointestinal disease (e.g. Crohn's disease, ulcerative colitis, or short gut syndrome) that would impact on drug absorption.
12. Patients under treatment with anticoagulants or with coagulation disorders or with signs of hemorrhage at baseline.
13. Patients with previous history or current presence of neurological disorders, including epilepsy (although controlled by anticonvulsant therapy), Parkinson's disease and extra-pyramidal syndromes.
14. Other severe acute or chronic medical or psychiatric condition or laboratory abnormality that may increase the risk associated with study participation or may interfere with the interpretation of study results and, in the judgment of the Investigator, would make the patient inappropriate for entry into this study or could compromise protocol objectives in the opinion of the Investigator and/or the Sponsor.

7. SCHEDULE OF EVENTS

The following table summarizes information on the timing of study assessments.

SCHEDULE OF EVENTS							
Protocol Activities and Forms to Be Completed	**Pre Treatment**		**Treatment Cycles**			**Post Treatment**	
	≤ 21 Days	**≤ 7 Days**	**Week 1 Day 1 (*)**	**Week 2 between Days 11-14**	**Last Cycle**	**AE FU (§)**	**Follow up**
Informed Consent **1**	X						
Medical/ Oncologic History & Physical Examination **2**	X			X	X		
Vital Signs **3**		X	X				
ECG **4**	X		As medically indicated		X		
Chest X-Ray **5**	X				X		
Ophthalmologic exam. **6**	X			X (even cycles)	X		
Laboratory Assessments							
Serum/ Urine Pregnancy Test **7**		X					
Urinalysis **8**		X	X **(*)**	X (even cycles)	X		
Hematology **9**		X	X **(*)**	X			
Blood Chemistry **10**		X	X **(*)**	X			
Coagulation **11**		X	X **(*)**	X (even cycles)	X		
Tumor Assessments							
Tumor Imaging: chest/abdomen/pelvic CT scan or MRI **12**	X			q6wks	X		q6wks
Survival **13**							q6wks (+ q6mos)
Other Clinical Assessments							
Adverse Event Assessment (including neurological status evaluation) **14**		X	X			X	
Concomitant Medications **15**		X	X				
Special Studies							
Biomarker Status Assessment **16**	X						
Pharmacokinetic Blood Sampling **17**			As needed				
Histological confirmation **18**	X						
Study Treatment							
Enrollment **19**		X					
PHA-848125AC Administration **20**			Once daily for 7 cons. days (D1 to D7)				
Footnotes for Schedule of Events							
1. Informed Consent: Every patient must sign the informed consent to participate in this trial before starting any trial related procedures. After signature, a Patient Screening Number is assigned centrally by CLIOSS on behalf ofTiziana Life Sciences, PLC.							
2. Medical/Oncology History & Physical Examination: Includes oncologic history, history of other disease processes (active, controlled or resolved) and concomitant illnesses (all of them at pretreatment only). Physical examination will be done at pretreatment, and, on treatment, at the end of each cycle (see Section 11.4.3 Other safety assessments). For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the physical examination will be done in concomitance with the visit to the site (once every 3 cycles, 1.5 months). Oncologic and non-oncologic medical history, including prior antitumor treatments/procedures, are to be reported in the relevant case report form (CRF). **Following approval of Amendment 4, management of patients should be as per Section 9.5**							

3. Vital signs: Blood pressure/pulse (supine): at baseline and on Day 1 of each treatment cycle, before treatment administration. Height (in cm or inches): at baseline only. Weight (in kilograms or pounds): at baseline and on Day 1 of each treatment cycle. ECOG performance status: at baseline and on Day 1 of each treatment cycle. Temperature (in °C or in °F): at baseline and on Day 1 of each treatment cycle. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, vital signs (including blood pressure/pulse, weight, ECOG performance status and temperature) will be done in concomitance with the visit to the site (once every 3 cycles, 1.5 months). **Following approval of Amendment 4, management of patients should be as per Section 9.5**
4. ECG: 12-lead study. To be done at baseline, at the end of the last cycle and to be repeated during treatment as medically indicated.
5. Chest X-Ray: Mandatory at screening visit and at the end of last cycle as safety assessment.
6. Ophthalmologic Examination: to be conducted at baseline, at the end of even cycles (between days 11 and 14) and at the end of last cycle. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, opthalmological examination will be done in concomitance with the visit to the site (once every 3 cycles, 1.5 months). Includes visual acuity test and fundoscopic examination by an ophthalmologist. Additional assessments will be performed if clinically indicated. Ophthalmologic evaluations will be possibly done by the same ophthalmologist for a given patient. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
7**. Pregnancy Test:** For women of reproductive potential. Must be done within 7 days prior to initial study treatment.
8. Urinalysis: pH, dipstick for glucose, protein and blood. To be done at baseline and at the end of last cycle. During treatment, should be performed at the end of even cycles (between days 11 and 14). For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the urinalysis tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
9. Hematology (local laboratory): Hemoglobin, erythrocytes (RBC), white blood cell (WBC) with differential count (neutrophils, lymphocytes, monocytes, eosinophils, basophils, differential other cells), platelets (PLTs). To be done at baseline and between Day 11-14 of each cycle. In case of toxicity preventing re-treatment, hematology should be re-assessed on Day 1 of the next cycle before starting treatment. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the hematological tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
10. Blood Chemistry (local laboratory)**:** sodium, potassium, chloride, magnesium, blood urea nitrogen (BUN) or urea, creatinine, albumin, AST/SGOT, ALT/SGPT, Alkaline Phosphatase (ALP), total bilirubin, creatinine clearance (calculated by Cockroft and Gault's formula), amylase and lipase. To be done at baseline and between Day 11-14 of each cycle. In case of toxicity preventing re-treatment, blood chemistry should be re-assessed on Day 1 of the next cycle before starting treatment. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the biochemical tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
11. Coagulation: INR (International Normalized Ratio), activated partial thromboplastin time (APTT in seconds). To be done at baseline, at the end of last cycle and, during treatment, at the end of even cycles (between Days 11 and 14). For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the coagulation tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
12. Tumor imaging: chest/abdomen/pelvic CT scan/ MRI : At baseline, to be done within 21 days before Day 1; during the treatment period (irrespectively from cycles duration), the first one to be done 6 weeks (between days 42 and 48) after first drug administration, **the second one to be done 3 months (between days 92 and 98) after first drug administration**, following ones to be repeated every 6 weeks (between days 42 and 48) after the previous one, and at end of last cycle (if not done in the previous 4 weeks). During Follow Up, to be performed every 6 weeks, until PD or until a new antitumor therapy starts. Patients with responding tumors (complete or partial response) must have response confirmed by CT scan at least 4 weeks after the 1st documentation of response. The same method of assessment and the same technique should be used to characterize and follow the same lesion at baseline and during treatment. CT scan is the desirable method for lesion measurement. Other measurement techniques (eg MRI) are acceptable (but not for lung lesions) provided that the size of the measurable lesion is twice the slice thickness of the MRI. **After data cut-off of 31 May 2017, the patients should perfom CT scan according to routine clinical practice and/or at investigator's discretion.**
13. Survival: To be assessed during Follow Up every 6 weeks until PD or until a new antitumor therapy starts; every 6 months thereafter, up to 2 years from the end of treatment. **Following approval of Amendment 4, management of patients should be as per Section 9.5**

14. Adverse Event Assessment (including neurological status evaluation): Events should be assessed and documented at each scheduled clinic visit since patient signs the Informed Consent: before treatment administration at baseline (baseline signs and symptoms), at the end of each treatment cycle, and 28 days after the last dose of study drug administration. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the events will be assessed by phone call if the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months). The phone call information will be reported in the medical chart and included in the CRF. Patients must be followed for adverse events also until every ongoing drug-related toxicities and serious adverse events have resolved or the investigator assesses them as “chronic” or “stable”. However, if the patient begins a new anticancer therapy before 28 days after the last dose of study drug administration, the adverse event reporting period will end at the time the new treatment is started. AEs will be recorded in the relevant CRF pages. **After cut-off date of 31 May 2017, AE/SAE data will be no longer collected into the CRF, but assessments information should continue to be collected into the patient’s medical notes.** **After protocol Amendment 5 approval, only SAEs will continue to be reported to CLIOSS Pharmacovigilance for patients still receiving treatment, up to 28 days after milciclib discontinuation, according to Section 9.5.**
15. Concomitant Medications: all concomitant medications should be reported in the relevant CRF, including supportive care drugs, and drugs used for treating adverse events or chronic diseases. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the concomitant medications will be assessed by phone call if the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months). The phone call information will be collected in the medical chart and included in the CRF. **Following approval of Amendment 4, management of patients should be as per Section 9.5**
16. Biomarker Status Assessment: retrieval of tumor paraffin embedded blocks (slides), obtained from consenting patients before study entry (if available) and characterization of baseline molecular features of p53, p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action.
17. Pharmacokinetics: blood samples may be collected if, in the opinion of the Investigator and of the Sponsor, an evaluation of PK parameters is needed for safety reasons.
18. Histological confirmation: histological confirmation of diagnosis of thymic carcinoma for all patients will be obtained by an Independent Review Committee during the course of the study, based on paraffin embedded tumor tissue slides retrieved from patients.
19. Enrollment: The Investigator is requested to sign a Request For Enrollment form. His/her signature is the guarantee that all the eligibility criteria are met. Patient Number is assigned by the Sponsor. Treatment must be administered within 7 days from patient enrollment.
20. PHA-848125AC administration: The study drug will be administered at 150 mg (flat dose) once daily for 7 consecutive days of each treatment cycle (from day 1 to Day 7). After an overnight fasting with free access to water, patients will take the study drug with a large glass of plain water without ice. A light breakfast can be served 1.5-2 hours later. PHA-848125AC capsules have to be stored under refrigerated conditions (5 ± 3 °C; 36-46 °F) and out of reach of children
(*) Cycle 1, Day 1 assessments: hematology, blood chemistry, urinalysis and coagulation do not need to be repeated before treatment on Cycle 1, Day 1 if they were performed during screening within 1 week before first study drug administration. **Cycle > 1, Day 1 assessments**: hematology and blood chemistry do not need to be repeated before treatment on Cycles> 1, Day 1 if no toxicity preventing re-treatment resulted from laboratory assessments done within the previous 4 days.
(§) AE FU: end of adverse events reporting period (to be done 28 days after last study drug administration) and follow up of unresolved drug related/serious AEs. **After data cut-off of 31 May 2017, AE/SAE data and related follow up will be no longer collected into the CRF, but assessments information should continue to be collected into the patient’s medical notes.** **After approval of Amendment 4 to the protocol, only SAEs should be reported to CLIOSS Pharmacovigilance for patients still receiving study treatment, up to 28 days after discontinuation of milciclib, according to Section 9.5.**

8. ENROLLMENT PROCEDURES

The informed consent should be signed and dated before starting any trial specific procedures. After signature of informed consent, the patient will be registered with a patient's screening number centrally assigned by CLIOSS on behalf of Tiziana (details reported in the Study Manual). Upon completion of screening evaluation, the patient, if eligible, will be enrolled and assigned a patient number to be carried out during the trial.

In order to obtain screening number and then patient number for each patient, the Investigator will fill in a registration/enrollment form and fax it to the CLIOSS study management (details reported in the Study Manual). Investigator's signature on the registration/enrollment form is the guarantee that all the eligibility criteria are met. A reply by fax will be sent within 1 working day. The time between enrollment and initiation of treatment should not be longer than 7 days.

Enrollment will be competitive among the participating sites.

CLIOSS will inform all sites on a regular basis about the enrollment status.

9. TREATMENT

Only qualified personnel familiar with procedures that minimize undue exposure to themselves and to the environment should undertake the preparation, handling, and safe disposal of chemotherapeutic agents.

9.1. Trial Products

9.1.1. Description

PHA-848125AC is formulated as hard gelatin capsules containing 10 mg, 50 mg and 100 mg, respectively, of active ingredient.

The three different strengths are distinguishable each other by the different color and capsules sizes: 10 mg capsules are size 4 hard gelatin capsules, opaque Swedish orange cap and opaque caramel body; 50 mg capsules are size 2 hard gelatin capsules, opaque Swedish orange cap and body; 100 mg capsules are size 1 hard gelatin capsules, opaque white cap and body. All the described capsules contain a pale yellow to yellow powder.

PHA-848125AC 10 mg, 50 mg and 100 mg capsules are packaged in white High Density Polyethylene (HDPE) bottles with twist off plastic caps.

New capsule formulations have been developed in order to have a formulation suitable for an automatic filling process, whereas the original formulations are filled with a semiautomatic capsule filling machine.

For more details on the new formulations, see PHA-848125AC Investigator Brochure, Version 7, 2011 [23].

9.1.2. Drug Preparation/Administration/Dispensing

PHA-848125AC will be administered as home-based treatment. The Investigator (or delegate) will dispense to the patient the doses of PHA-848125AC covering no more than two cycles at a time during the first three months of treatment (6 cycles) and covering three cycles at a time after three months of treatment (for patients who continue treatment after 6 cycles and come back to the site every 3 cycles). The daily doses will be made out of a number of capsules, according to the dose (in mg/day) at which the patient has been assigned for a given cycle.

The study medication has to be stored under refrigerated conditions (5 ± 3 °C; 36-46 °F), in the original packaging, out of the reach and sight of children.

Original IMP Packaging

As each capsule of PHA-848125AC is individually packaged in a bottle (the primary packaging), the daily dose of PHA-848125AC will consist of a number of bottles packed together in an outer packaging (bag) and the 7 bags will be stored in a weekly container (box). At each cycle, patients will take the assigned daily dose for 7 consecutive days (from Day 1 to Day 7) and will be instructed not to take the study drug for the subsequent 7 days (i.e. Days 8-14), so to ensure a 7-day resting period.

A secondary packaging (box) will be used to store stocks of bottles in the pharmacy of the participating institutions only.

Primary, secondary and outer packaging, and weekly container will be labeled with the relevant information.

New IMP Packaging

A new IMP packaging, currently under preparation, will be introduced and will consist of one PHA-848125AC capsule of each strength individually packaged in a labeled bottle (the primary packaging) and each bottle inserted in one labeled small single carton (the secondary packaging).

The Sponsor has considered to simplify the original IMP packaging above described, so that no repacking activities for patient doses preparation should occur at clinical trial sites. The original IMP packaging will be used in the clinical trial until the new IMP packaging will be available.

The different IMP strengths (50 mg and 100 mg) will be easily distinguished by using different colors for labels stuck on primary and secondary packaging.

As each capsule of PHA-848125AC is individually packaged in a bottle/single carton, the daily dose of PHA-848125AC will consist of a number of bottles/cartons (e.g. for a full dose of 150 mg/day: 1 bottle containing one 50 mg capsule in a single small carton and 1 bottle containing one 100 mg capsule in a single small carton). At each cycle, patients will take the assigned daily dose for 7 consecutive days (from Day 1 to Day 7) and will be instructed not to take the study drug for the subsequent 7 days (i.e. Days 8-14), so to ensure a 7-day resting period.

9.1.3. Patient Education & Information

All patients will receive their doses of PHA-848125AC together with a patient's diary, which includes also the instructions relevant to study drug administration. The site personnel handing the study medication to patients must ensure that they fully understand that the treatment period (cycle) comprises 14 days, consisting of 7 days of daily administration (from Day 1 to Day 7), followed by a 7-day resting period, from Day 8 to Day 14. Extension of treatment period to Days >7 is not allowed, even in case of missed doses.

Additionally, the patients should be informed:

- to take the study drug every day as prescribed by their doctor. Some patients may find particularly hard to remember the idea of repeated short courses of treatment with 'gaps' between them.
- to take the study drug at the same time each day (1.5-2 hours before breakfast with a glass of plain water without ice; water is allowed during fasting period).
- if the patient forgets to take the study drug at the usual time, to take it between meals at any time during the same day and record the events on the patient's diary.
- if the patient misses one day, to take the normal amount the next day, if scheduled. Not to double the daily prescribed dose of the study drug and to track the number of capsules missed and the reason for it in the patient's diary.
- if vomiting occurs during or after having taken the daily dose, not to take extra capsules that day and to take the normal amount the next day, if scheduled. Not to double the daily prescribed dose of the study drug and record the events in the patient's diary.
- to adhere to the principles of safe handling and storage of the study medication. Breaking/crushing or opening the capsules must be avoided. In such cases, record of the broken capsules should be kept in the patient's diary. In case of opening of the capsules, to avoid contact or inhalation. In case of skin contact, to wash the affected area with plenty of water or soap and water. In case of eye-contact, to rinse thoroughly with plenty of water and seek medical advice as soon as possible.
- to return all packaging (bottles, bags and carton boxes for the original IMP packaging and bottles and single cartons for the new IMP packaging) as well as not-taken capsules (if applicable) and the patient diary to the site personnel.

As much of this information as possible should first be given at the pre-treatment visit and reinforced on subsequent visits.

9.2. Procedure for Handling Drug Spills

Precautions for drug handling of cytotoxic agents should be followed for PHA-848125AC. In case of opening of the capsules, avoid contact or inhalation. In case of skin contact, the affected area should be washed with plenty of water or soap and water. Any contaminated clothing should be removed. In case of eye-contact, rinse thoroughly with plenty of water. Seek medical advice as soon as possible.

9.3. Storage and Stability

PHA-848125AC is to be stored under refrigerated conditions (5 ± 3 °C; 36-46 °F) and in the original packaging, out of the reach and sight of children.

To ensure optimal environmental and patient exposure protection, the packaging has been designed as a single use one.

On the basis of the available supportive stability results, a provisional shelf-life of 48 months is assigned to both 50 mg and 100 mg capsules, when stored at 5 ± 3 °C (36-46 °F) in the original packaging.

Concurrent stability studies on the clinical batches of the current formulation will be carried out in order to evaluate the chemical and physical parameters, and the expiry date will be extended accordingly with the stability results. Any unexpected finding will be promptly communicated to the investigators and to the Health Authorities.

9.3.1. Source of Drug

NerPharMa (NMS Group), on behalf of the Sponsor Tiziana Life Sciences PLC will supply PHA-848125AC (milciclib maleate) free of charge. Additional supply may be obtained by contacting CLIOSS (see Study Manual for contact information).

9.3.2. Drug Accountability

PHA-848125AC must not be used outside the context of this protocol. Under no circumstances should the Investigator or site personnel supply study product to other Investigators or clinics, or allow the supplies to be used other than as directed by this protocol without prior authorization from the Sponsor.

Adequate records on receipt, use, return, loss, or other disposition of medication must be maintained. A specific drug accountability form supplied by the Sponsor or computer records used by the pharmacy at the investigational site, can be used to provide drug accountability information. In either case, information describing study drug supplies and

their disposition, subject by subject, must be provided, signed by the Investigator (or the pharmacist or other person who dispensed the drug) and collected by the study monitor. Requisite data includes relevant dates, quantities, batches or code numbers, and subject identification for subjects who received study drug.

At the end of the study and upon authorization of the Sponsor, the study drug may be destroyed at the site as dictated by the appropriate standard procedures of the participating institutions. Destruction must be documented. Alternatively, all unused product will be collected by the local monitor and returned to NerPharMa.

If disposed locally, all used bottles and containers of study product should be discarded according to the standard institutional policy.

Study drug handling and accountability after the cut-off date of 31 May 2017 should continue as described above.

9.4. Treatment Administration

9.4.1. Treatment Dose and Schedule

PHA-848125AC will be administered orally at the dose of 150 mg/day (flat dose) once daily for 7 consecutive days of each treatment cycle. A treatment cycle will comprise 7 days of PHA-848125AC administration (Day 1 to 7) followed by 7 days of rest for a total of 14 days (2 weeks) period.

After an overnight fasting, with free access to water, patients will take the study drug with a large glass of plain water without ice. A light breakfast can be served 1.5-2 hours after study drug intake.

9.4.2. Duration of Treatment

Each patient will remain on treatment until disease progression, patient refusal, consent withdrawal, or the occurrence of unacceptable toxicity or of any other criteria for withdrawal as detailed in Section 10.

At the end of the treatment period, all patients will be followed for survival every 6 months up to 2 years after the end of treatment. Anyhow, if a patient stops the treatment before any documentation of PD, the follow up will be initially performed at 6-week intervals and will include a complete oncological assessment up to documentation of PD or the start of another antitumor therapy, then it will be continued for survival only every 6 months.

Patients still on treatment after the cut-off date of 30 April 2017, will be managed according to Section 9.5.

9.4.3. Dose Modifications

Dose modifications may occur as a consequence of drug-related toxicities, during a cycle or at the start of a new cycle. In the event of multiple toxicities, dose modifications should be applied based upon the most severe toxicity observed and attributable to PHA-848125AC.

As far as dose reductions are concerned, a dose reduction to the dose level of 100 mg/day (i.e. –33%) is recommended in case of poor tolerability of the full dose of 150 mg/day.

The dose of 100 mg/day (mean Day 7 AUC 19.9 μM·h) was successfully used for 20 cycles in keeping disease control in a patient with colorectal cancer, initially treated at 150 mg/day for 7 cycles in the CDKO-125a-001 study.

Doses reduced for drug-related toxicity should not be re-escalated. Any exception is to be discussed between the Investigator and the Sponsor.

Table 4 describes the recommended dose modifications during the treatment cycles and at the start of any new cycle.
Note: the dose adjustments in the third column are intended as referred to the dose of Day 1 of the previous cycle.

Table 4. Criteria for PHA-848125AC Dose Modifications

Toxicity (Graded as per NCI CTCAE Criteria, Version 3.0)	**During Treatment Cycle**	**Dose Adjustment for Next Treatment Cycle (after recovery from toxicities to grade ≤1, or to baseline values) ¶**
Hematologic		
Grade 0 or Grade 1	Maintain dose level	Maintain dose level
Grade 2 Neutropenia (ANC < 1500-1000/mm^3) and/or Grade 2 Thrombocytopenia (platelet count <75000-50000/mm3)	If occurs during treatment, maintain daily dose level Monitor until resolved to grade ≤1	Maintain dose level
Grade 3 Neutropenia (ANC <1000-500/mm^3)	If occurs during treatment, decrease daily dose by one dose level Monitor until resolved to grade ≤1	Maintain dose level
Grade 3 Thrombocytopenia (platelet count <50000-25000/mm3), uncomplicated or lasting less than 5 days	If occurs during treatment, decrease daily dose by one dose level Monitor until resolved to grade ≤1	Maintain dose level
Grade 4 Neutropenia (ANC < 1000/mm^3) lasting < 7 days	If occurs during treatment, decrease the daily dose by one dose level Monitor until resolved to grade ≤1	Maintain dose level
Grade 4 Neutropenia (ANC < 1000/mm^3) lasting 7 days or more	If occurs during treatment, interrupt the treatment Monitor until resolved to grade ≤1	↓ 1 dose level
Febrile neutropenia (ANC <1,000/mm3 and fever of unknown origin ≥38.5°C)		
Neutropenic infection: grade ≥ 3 (i.e. infection documented clinically or microbiologically with grade 3 or grade 4 neutropenia)		
Grade ≥ 3 Thrombocytopenia (platelet count <50000/mm^3) uncomplicated lasting 5 days or more, or associated with bleeding and/or requiring transfusion		
Any other grade 4 hematological toxicity		
Nausea and/or Vomiting		
Grade 1 or 2 in absence of antiemetics	If occurs during treatment, maintain daily dose level and add antiemetics, if needed. If occurs during rest period, add antiemetics, if needed	Maintain dose level with antiemetics §, if needed
Grade 3 in absence of antiemetics	If occurs during treatment, decrease the daily dose by one dose level, add antiemetics. If occurs during rest period, add antiemetics Monitor until resolved to grade ≤1	Maintain dose level with antiemetics §
Grade ≥3 in presence of antiemetics §	If occurs during treatment, interrupt treatment Monitor until resolved to grade ≤1	↓ 1 dose level

Toxicity (Graded as per NCI CTCAE Criteria, Version 3.0)	**During Treatment Cycle**	**Dose Adjustment for Next Treatment Cycle (after recovery from toxicities to grade ≤1, or to baseline values) ¶**
Diarrhea		
Grade 1 or 2 (in absence or presence of management of the event §)	If occurs during treatment, maintain daily dose level, add/adjust antidiarrheal treatment, if needed. If occurs during rest period, add/adjust antidiarrheal treatment, if needed	Maintain dose level
Grade ≥3 despite optimal management of the event §	If occurs during treatment, interrupt treatment Monitor until resolved to grade ≤1	↓ 1 dose level
Neurologic		
Grade 0, 1 or no worsening compared to baseline	Maintain dose level	Maintain dose level
Grade 2 tremors	If occurs during treatment, interrupt treatment	↓ 1 dose level or **discontinue** study treatment (Investigator to discuss with the Sponsor)
Grade > 2 tremors or any other Grade ≥ 2 neurological toxicity	**Discontinue study treatment** (Investigator to discuss with the Sponsor)	**Discontinue study treatment** (Investigator to discuss with the Sponsor)
Ocular/Visual		
Grade 0	Maintain dose level	Maintain dose level
Increase of 1 or more grades retinopathy by ophthalmologic examination compared to baseline	**Discontinue study treatment**	**Discontinue study treatment**
Other non hematological toxicity (except alopecia)		
Grade 0, 1	Maintain dose level	Maintain dose level
Grade 2	If occurs during treatment, maintain or decrease the daily dose by 1 dose level, if clinically indicated Monitor until resolved to grade ≤1	Maintain dose level
Grade ≥ 3	If occurs during treatment, interrupt treatment Monitor until resolved to grade ≤1	↓ 1 dose level
Failure to recover		
Failure to recover to grade ≤ 1 toxicity (except alopecia) or to baseline values, if grade 2 is allowed at study entry, after delaying the initiation of next cycle by > 2 weeks	Monitor until resolved to grade ≤1	↓ 1 dose level (Investigator to discuss with the Sponsor)

¶ : the dose adjustments in the third column are intended as referred to the dose of Day 1 of the previous cycle.

§ For prophylaxis and management of the events, see Concomitant Medications and Other Therapy, Section 9.4.9.

Abbreviations: ANC = absolute neutrophil count; CTCAE = Common Terminology Criteria for Adverse Events, NCI = National Cancer Institute

9.4.4. Retreatment and Dose Delay

A new cycle of treatment may begin when the ANC is ≥ 1500/mm^3, the platelet count is ≥ 100,000/mm^3, and the non-hematologic toxicities are ≤ grade 1, except for alopecia (for which a grade > 1 is allowed) and for ocular toxicity (for which a worsening retinopathy of 1 or more grades compared to baseline implies study treatment discontinuation) or are recovered to baseline values, if grade 2 is allowed at study entry. If these conditions have not been met, treatment should be delayed for 1 or 2 weeks to allow for recovery. If after a maximum of a 2-week delay all toxicities (except for alopecia and for toxicities for which grade 2 is allowed at study entry) are ≤ grade 1, then proceed with treatment as outlined in the Dose Modification Table 4. An increased delay > 2 weeks is to be discussed between the Investigators and the Sponsor on a case by case basis.

9.4.5. Overdose Instructions

There are no known antidotes for milciclib maleate PHA-848125AC. In the case of an overdose of PHA-848125AC, neurological toxicity (tremors and ataxia) and gastrointestinal toxicity are among the expected effects. The Sponsor study management should be contacted to discuss the details of any overdose.

9.4.6. Unblinding

Not applicable in this study.

9.4.7. Assessment and Management of Potential CNS Toxicity

Signs of CNS toxicity, including tremors, hyperactivity and ultimately convulsions have been observed during the toxicology evaluations of PHA-848125AC administered at high single or high repeated doses attaining daily systemic exposures ≥ 30μM.h. In all species, this pattern of toxicity was reversible and not accompanied by morphological/pathological findings.

In humans, reversible tremors and ataxia have been reported in the CDKO-125a-001 study (see Section 3.2.7). Resolution of both events in approximately 7-8 days was reported, upon study treatment discontinuation.

Tremors which can be induced by other drugs include enhanced physiologic tremor, rest tremor, and action tremor. Signs and symptoms of drug-induced tremors depend on the drug used and on a patient's predisposition to its side effects. Cessation of the causative agent may improve drug-induced tremor symptoms. Table 5 lists drugs that may induce tremor, along with the types of tremors and neurologic signs they produce.

Table 5. Drug-induced Tremors and Corresponding Neurologic Signs

Drug/Drug Class	Tremor Type	Neurologic Signs
Neuroleptics	Rest, postural	Extrapyramidal
Metoclopramide	Rest, postural	Extrapyramidal
Lithium	Rest, postural, action	Extrapyramidal
Theophylline	Postural	None
Bronchodilators	Postural, action	None
Valproate	Postural	Rarely parkinsonism
Amiodarone	Postural	Rarely parkinsonism

During visits, patients treated with PHA-848125AC will undergo regular clinical examination, which includes the assessment of clinical tremors and associated signs and symptoms. Patients should be examined during rest, when assuming various positions and when moving. Tremors will be graded according to NCI CTCAE version 3.0 grading system as follows:

Tremor Grades, NCI CTCAE Version 3.0
0. Absent
1. Mild and brief or intermittent but not interfering with function
2. Moderate tremor interfering with function, but not interfering with ADL
3. Severe tremor interfering with ADL
4. Disabling

PHA-848125AC dose modifications for neurologic adverse events attributable to the study drug are detailed in Table 4.

Pharmacological management of neurological adverse events, such as tremors and/or convulsions, should refer to institutional and/or published interventional protocols [35,36].

9.4.8. Assessment of Potential Ocular Toxicity

Ocular toxicity has been reported in rats treated for prolonged period of time (daily treatments for 3 consecutive weeks + 1 week of rest x 3 cycles or daily treatments for 4 consecutive weeks) at doses associated with daily AUCs close to 25 μM·h, as well as in rats given daily treatments for 2 consecutive weeks +1 week of rest for 5 cycles at doses associated to AUC of about 32 μM·h.
No ocular toxicity was observed in rats with a 7-day repeat treatment schedule up to doses associated to a daily AUC of about 50μM·h or up to doses associated to a daily AUC of about 10 μM·h in the 4-week schedule and up to doses associated to daily AUC of about 12 μM·h. in the 3-week schedule.

Despite clinical manifestations of ocular toxicity were not observed in the clinical studies so far, a potential ocular toxicity cannot be excluded. During visits, patients treated with PHA-848125AC will undergo regular clinical examinations, i.e. ophthalmologic examination (visual acuity test and funduscopic examination), at the end of even cycles (see Schedule of Events in Section 7 and also Section 11.4.3).

Retinopathy will be graded according to NCI CTCAE version 3.0 grading system as follows:

Retinopathy Grades, NCI CTCAE Version 3.0	
0.	Absent
1.	Asymptomatic
2.	Symptomatic with moderate decrease in visual acuity (20/40 or better)
3.	Symptomatic with marked decrease in visual acuity (worse than 20/40)
4.	Blindness (20/200 or worse)

Grade 1 retinopathy is allowed at study entry since it is an asymptomatic event occurring in relation to common medical conditions such as hypertension [37,38]. It is recognized that several patients in their 50s and 60s with underlying hypertension may present asymptomatic retinopathy. Such a status is to be regarded as a condition which does not prevent patients' enrollment in the study, being not clinically significant.

Ophtalmologic evaluations will be possibly done by the same ophthalmologist for a given patient.
The increase of 1 or more grades retinopathy by ophthalmologic examination will require immediate study drug discontinuation.

Management of ocular adverse events should refer to institutional interventional protocols.

9.4.9. Assessment of Potential, Thrombotic Microangiopathy/Hemolytic Uremic Syndrome

Two patients, both treated in the CDKO-125a-004 combination study with gemcitabine ('A Phase I Dose-Escalation Study of Oral PHA-848125AC Administered in Combination with Gemcitabine in Adult Patients with Advanced/Metastatic Solid Tumors'), developed Thrombotic Microangiopathy/Hemolytic Uremic Syndrome (TMA/HUS).
The two cases were considered to be probably related to gemcitabine and unlikely related to PHA-848125AC by the participating Investigator, and occurred in patients with very prolonged and high exposure to both drugs (after 14 and 21 months on treatment, respectively); both patients recovered in the opinion of the Investigator, and only one of them required plasmapheresis. No other cases of TMA/HUS were reported in any of the other studies completed or ongoing with PHA-848125AC (all single agent studies). Anyway, taking into consideration the small number of patients treated so far at this stage of development of PHA-848125AC (about 200 patients), a contribution of the study drug on

the two observed TMA/HUS events cannot be completely ruled out. As a consequence, special attention should be paid to the following events:

- Progressive increase of anemia up to CTC Grade ≥ 3 no otherwise explained
- Progressive platelets decrease up to CTC Grade ≥ 2
- Increase of creatinine (shift to CTC Grade 2) or calculated creatinine clearance diminished by ≥ 25% vs baseline
- Presence of hematuria or proteinuria
- Appearance of high blood pressure in a patient with unknown history or with high blood pressure previously well controlled with medications
- Appearance of oedema concomitant to proteinuria

Whenever ≥ 2 of the above listed alterations appear concomitantly for the first time, or at any time if the Investigator judges it appropriate, study drug administration is to be temporarily held, until further evaluations are done. The latter ones consisting of the search of schistocytes in the peripheral blood smear, the count of reticulocytes in the peripheral blood, coagulation test, total LDH and 24-hour proteinuria [39]. Routine Hematology, Blood Chemistry and Urinalysis tests foreseen by study protocol, should also be repeated at the time when this evaluation is performed.

If based on the above examinations MTA/HUS can be ruled out, study drug administration can be resumed, adopting the dose modifications outlined in Table 4 for drug-related toxicities, as applicable.

In case of suspicion or evidence of MTA/HUS, study drug has to be discontinued.

9.4.10. Concomitant Medications and Other Therapy

All concomitant medications must be entered into the CRF.
After the data cut-off of 31 May 2017 and protocol Amendment 4 is approved at the investigational site, data will be no longer collected into the CRF, and information on concomitant medications should continue to be recorded into the patient's medical notes only.
Directives for supportive care are outlined below.

9.4.10.1. Antiemetics

Prophylactic therapy with antiemetics is allowed starting from the first onset of emesis. Pharmacological management of nausea and/or vomiting may refer to institutional and/or published guidelines for treatment [40,41].

9.4.10.2. Antidiarrheals

Treatment with loperamide might be started at the occurrence of grade 1 diarrhea (increase of < 4 stools per day over baseline; mild increase in ostomy output compared to baseline). Loperamide should be taken in the following manner: 4 mg at the first onset of diarrhea, then 2 mg every 2 hours. This therapy should continue for 12 hours after the last liquid stool. Patients may take loperamide 4 mg every 4 hours during the night. In no instance should loperamide be administered for more than 48 consecutive hours because of the risk of paralytic ileus.
Other antidiarrhetic agents and/or antibiotics used by the Institutions are allowed.
Patients should be instructed to refer to the center in case of diarrhea lasting > 24 hours despite optimal supportive care or diarrhea with fever.

9.4.10.3. Antiacids

Patients with chronic/ intensive use of H2-receptor antagonists, proton pump inhibitors, antacids, should take these drugs at a time point far from PHA-848125AC intake (at least 3-4 hours) during the day, or the night before treatment, to avoid any potential effect on PHA-848125AC absorption.

9.4.10.4. Hematopoietic Growth Factors

Hematopoietic growth factors may be used as medically indicated in patients with relevant neutropenic complications, such as tissue infection, sepsis, etc., at the Investigator's discretion. Red blood cell and/or platelets transfusion should be considered at the Investigator's discretion in patients with significant anemia and/or thrombocytopenia.

9.4.10.5. Anticoagulants

Concomitant anticoagulant treatment is not recommended.

9.4.10.6. Inhibitors or Inducers of CYP3A4

Significant drug-drug interactions with inhibitors or inducers of CYP3A4 are not expected (see Section 3.2.4). Therefore, there are no specific contraindications to the use of inhibitors or inducers of CYP3A4.

9.4.10.7. Steroids

Concurrent use of steroids for treatment of a pre-existing autoimmune disorder or as antiemetic therapy is allowed.

9.4.10.8. Other Permitted Concomitant Medications

Therapies considered necessary for the patient's well being may be given at the discretion of the Investigator, i.e. chronic treatments for concomitant medical conditions, as well as agents required for life-threatening medical problems, analgesics etc. Patients should be advised to contact the treating physician before starting any new drug.

9.4.10.9. Concomitant Radiotherapy

Palliative radiotherapy to specific sites is permitted if considered medically necessary by the treating physician. However, the irradiated area should be as small as possible, and involve ≤20% of the bone marrow reserve [Appendix 2]. During the period of irradiation and for 2 weeks after, chemotherapy should be withheld. If irradiation-related toxicities (other than xerostomia) have not normalized to pre-irradiation levels after these 2 weeks of rest, the patient should be removed from the study. Radiotherapy should be avoided for at least 5 days after the last dose of PHA-848125AC.

9.4.10.10. Other Anticancer or Experimental Therapy

No other approved or investigational anticancer treatment will be permitted during the study period including chemotherapy, biological response modifiers, hormones or immunotherapy. No other investigational drug may be used during therapy on this protocol. Simultaneous participation in other clinical treatment study protocols is not allowed.

9.5. Management of Patients after Data Cut-Off

Patients still receiving milciclib maleate at the cut-off date of 31 May 2017 can continue with their treatment, and will continue with their protocolled scheduled visits to the site until they meet any discontinuation criteria as per Section 10.
However, after protocol Amendment 4 is approved at the investigational site, data will be no longer collected into the CRF, and assessments information should continue to be recorded into the patient's medical notes only. After protocol Amendment 4 approval, the patients should attend visit according to routine clinical practice and/or at investigator's discretion, until they meet any discontinuation criteria as per Section 10.

After cut-off date of 31 May 2017, SAEs will continue to be reported as per protocol for patients still receiving study treatment, up to 28 days after discontinuation of milciclib.

Drug supply will be granted by the Sponsor free of charge, and accountability should continue to be performed until the patient stops study treatment completely.

The end of the study is defined as the date of the last visit of the last patient, occurring when all patients have discontinued study treatment.

10. SUBJECT WITHDRAWAL FROM STUDY PARTICIPATION

Patients may continue with therapy unless any of the following occurs:

- Disease progression at any time
- Unacceptable toxicity dictating cessation of treatment
- Changing in medical status of the patient (including pregnancy) such that the Investigator believes that patient safety will be compromised or that it would be in the best interest of the patient to stop treatment
- Withdrawal of consent (includes any reason why the patient decides to discontinue completely his/her study participation and does not wish that the Sponsor follows him/her to collect information regarding disease status. In this case no further evaluation should be performed and no attempts should be made to collect additional data)
- Patient's refusal to continue the study treatment (includes any patient who does not want to be treated anymore in the frame of this study but accepts that the Sponsor continues to follow him/her in order to collect information regarding disease status). Applicable only for patients treated pre-Amendment 4.
- Non-compliance by the patient with protocol requirements. Applicable only for patients treated pre-Amendment 4.
- Patient lost to follow up. If a patient does not return for a scheduled visit, every effort should be made to contact the patient. In any circumstance, every effort should be made to document patient outcome. Applicable only for patients treated pre-Amendment 4.

11. ASSESSMENTS

11.1. Timing of Assessments

Section 7 summarizes information on the timing of study assessments.

11.2. Efficacy Assessments

The determination of antitumor efficacy will be based on objective tumor assessments made according to the RECIST guideline (version 1.1) [34].

11.2.1. Tumor Imaging

Tumor imaging by chest/abdomen/pelvic CT scan or MRI: the baseline tumor assessment by CT scan or MRI has to be performed within 3 weeks prior to the treatment start; during the treatment period (irrespectively of cycles duration) the first assessments will be performed 6 weeks (between days 42 and 48) after first drug administration, **the second one 3 months (between days 92 and 98) after first drug administration**, the following ones every 6 weeks (between days 42 and 48) after the previous one (or whenever a clinical

deterioration will be observed) and at end of last cycle (if not done in the previous 4 weeks). For patients with responding tumors (complete or partial response), response confirmation must be performed no less than 4 weeks after the criteria for response are first met.

In addition, in patients withdrawn from study treatment for causes other than disease progression, tumor response will have to be reassessed at the time of study drug discontinuation, unless a tumor assessment was performed within the previous 4 weeks. In these patients, regular tumor imaging assessments are to be performed every 6 weeks during follow-up, until PD or until a new antitumor therapy starts.

The same method and technique (CT scan, spiral CT scan or MRI) should be used to characterize each identified and reported lesion at baseline and during follow-up. Imaging-based evaluation, over clinical examination, is the required technique when both could be used to assess the antitumor effect of the treatment.
CT scan is the desirable method for lesion measurement. CT scans should have a maximum slices thickness of 5 mm and a minimum size for a measurable lesion is twice that: 10 mm (even if slice thickness is <5 mm). If scanners with slice thickness > 5 mm are used, to qualify for a measurable lesion, the minimum lesion size must have a longest diameter twice the actual slice thickness. MRI is acceptable (but not for lung lesions), provided that the size of the measurable lesion is twice the slice thickness of the MRI.

All patients' files and scans must be available for source verification.

11.2.1.1. Measurability of Tumor Lesions

At baseline, tumor lesions will be categorized by the Investigator as measurable or non-measurable by the RECIST guideline (version 1.1) criteria as described below.

Measurable:
Tumour lesions: Must be accurately measured in at least one dimension (longest diameter in the plane of measurement is to be recorded) with a minimum size of:
• 10 mm by CT scan (CT scan slice thickness no greater than 5 mm). In case of MRI, the size of the measurable lesion must be twice the slice thickness of the MRI and a minimum of 10 mm.
• 10 mm caliper measurement by clinical exam (lesions which cannot be accurately measured with calipers should be recorded as non-measurable).
• 20 mm by chest X-ray.
Malignant lymph nodes: To be considered pathologically enlarged and measurable, a lymph node must be ≥15 mm in short axis when assessed by CT scan (CT scan slice thickness recommended to be no greater than 5 mm). At baseline and in follow-up, only the short axis will be measured and followed.

Non-Measurable:
All other lesions, including small lesions (longest diameter <10 mm or pathological lymph

nodes with ≥10 to <15 mm short axis) as well as truly non-measurable lesions. Lesions considered truly non-measurable include: leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, abdominal masses/abdominal organomegaly identified by physical exam that is not measurable by reproducible imaging techniques.

Special considerations regarding lesion measurability:
Bone lesions, cystic lesions, and lesions previously treated with local therapy require particular comment:
Bone lesions:
• Bone scan, PET scan or plain films 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, that can be evaluated by cross sectional imaging techniques such as CT or MRI can be considered as measurable lesions if the soft tissue component meets the definition of measurability described above.
• Blastic bone lesions are non-measurable.
Cystic lesions:
• Lesions that meet the criteria for radiographically defined simple cysts should not be considered as malignant lesions (neither measurable nor non-measurable) since they are, by definition, simple cysts.
• "Cystic lesions" thought to represent cystic metastases can be considered as measurable lesions, if they meet the definition of measurability described above. However, if non-cystic lesions are present in the same patient, these are preferred for selection as target lesions.
Lesions with prior local treatment:
• Tumor lesions situated in a previously irradiated area, or in an area subjected to other loco-regional therapy, are usually not considered measurable unless there has been demonstrated progression in the lesion.

When effusions are known to be a potential adverse effect of treatment, the cytological/histological confirmation of the neoplastic origin of any effusion that appears or worsens during treatment when the measurable tumor has met criteria for response or SD is mandatory to differentiate between response or SD and PD.

11.2.1.2. Recording Tumor Measurements

Target lesions. When more than one measurable lesion is present at baseline, all lesions up to a maximum of five lesions total (and a maximum of two lesions per organ) representative of all involved organs should be identified as target lesions and will be recorded and measured at baseline (this means in instances where patients have only one or two organ sites involved a maximum of two and four lesions respectively will be recorded). Target lesions should be selected on the basis of their size (lesions with the longest diameter), be representative of all involved organs, 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 which can be measured reproducibly should be selected.
Pathological nodes which are defined as measurable and may be identified as target lesions must meet the criterion of a short axis of ≥15 mm by CT scan. Only the short axis of these nodes will contribute to the baseline sum. The short axis of the node is the diameter normally used by radiologists to judge if a node is involved by solid tumor. Nodal size is normally reported as two dimensions in the plane in which the image is obtained (for CT scan this is almost always the axial plane; for MRI the plane of acquisition may be axial, sagittal or coronal). The smaller of these measures is the short axis. For example, an abdominal node which is reported as being 20 mm × 30 mm has a short axis of 20 mm and qualifies as a malignant, measurable node. In this example, 20 mm should be recorded as the node measurement.
A sum of the diameters (longest for non-nodal lesions, short axis for nodal lesions) for all target lesions will be calculated and reported as the baseline sum diameters. If lymph nodes are to be included in the sum, then as noted above, only the short axis is added into the sum. The baseline sum diameters will be used as reference to further characterize any objective tumor regression in the measurable dimension of the disease.

Non-target lesions. All other lesions (or sites of disease) should be identified as non-target lesions and should also be recorded at baseline, including pathological lymph nodes with short axis ≥10 mm but <15 mm (nodes that have a short axis <10 mm are considered non-pathological and should not be recorded or followed).
Measurements are not required and these lesions should be followed as “present”, “absent”, or in rare cases “unequivocal progression”. In addition, it is possible to record multiple non-target lesions involving the same organ as a single item on the case record form (e.g. “multiple enlarged pelvic lymph nodes” or “multiple liver metastases”).

11.2.1.3. Target Lesions Tumor Response

Complete response (CR) is defined as disappearance of all target lesions and reduction in short axis to <10 mm of any pathological lymph nodes (whether target or non-target).

Partial response (PR) is defined as at least 30% decrease in the sum of the diameters of the target lesions, taking as a reference the baseline sum diameters.

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

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

Special notes on the assessment of target lesions
Lymph nodes: lymph nodes identified as target lesions should always have the actual short axis measurement recorded (measured in the same anatomical plane as the baseline examination), even if the nodes regress to below 10 mm on study. This means that when lymph nodes are included as target lesions, the 'sum' of lesions may not be zero even if complete response criteria are met, since a normal lymph node is defined as having a short axis of <10 mm. For PR, SD and PD, the actual short axis measurement of the nodes is to be included in the sum of target lesions.
Target lesions that become "too small to measure": while on study, all lesions (nodal and non-nodal) recorded at baseline should have their actual measurements recorded at each subsequent evaluation, even when very small (e.g. 2 mm). However, sometimes lesions or lymph nodes which are recorded as target lesions at baseline become so faint on CT scan that the radiologist may not feel comfortable assigning an exact measure and may report them as being 'too small to measure'. When this occurs it is important that a value be recorded on the case report form. If it is the opinion of the radiologist that the lesion has likely disappeared, the measurement should be recorded as 0 mm. If the lesion is believed to be present and is faintly seen but too small to measure, a default value of 5 mm should be assigned. (Note: It is less likely that this rule will be used for lymph nodes since they usually have a definable size when normal and are frequently surrounded by fat such as in the retroperitoneum; however, if a lymph node is believed to be present and is faintly seen but too small to measure, a default value of 5 mm should be assigned in this circumstance as well). This default value is derived from the 5 mm CT slice thickness (but should not be changed with varying CT slice thickness). The measurement of these lesions is potentially non-reproducible, therefore providing this default value will prevent false responses or progressions based upon measurement error. To reiterate, however, if the radiologist is able to provide an actual measure, that should be recorded, even if it is below 5 mm.
Lesions that split or coalesce on treatment: when non-nodal lesions "fragment", the longest diameters of the fragmented portions should be added together to calculate the target lesion sum. Similarly, as lesions coalesce, a plane between them may be maintained that would aid in obtaining maximal diameter measurements of each individual lesion. If the lesions have truly coalesced such that they are no longer separable, the vector of the longest diameter in this instance should be the maximal longest diameter for the "coalesced lesion".

11.2.1.4. Non Target Lesions Tumor Response

While some non-target lesions may actually be measurable, they need not be measured and instead should be assessed only qualitatively at the time points specified in the protocol.

Complete response (CR) is defined as the disappearance of all non-target lesions and normalization of tumor marker level. All lymph nodes must be non-pathological in size (<10 mm short axis).

Non-CR/Non-PD is defined as a persistence of one or more non-target lesion(s) and/or maintenance of tumor marker level above the normal limits.

Progressive disease (PD) is defined as unequivocal progression of existing non-target lesions. (Note: the appearance of one or more new lesions is also considered progression).

Special notes on assessment of progression of non-target disease
To achieve "unequivocal progression" on the basis of the non-target disease, there must be an overall level of substantial worsening in non-target disease such that, even in presence of SD or PR in target disease, the overall tumor burden has increased sufficiently to merit discontinuation of therapy. A modest increase in the size of one or more non-target lesions 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 PR of target disease will therefore be extremely rare.

11.2.1.5. Confirmation of Tumor Response

To be assigned a status of PR or CR, changes in tumor measurements in patients with responding tumors must be confirmed ≥ 4 weeks after the criteria for response are first met by the same tumor imaging technique used at baseline and on treatment (*eg* CT scan or MRI).
In case of SD, measurements must have met the SD criteria at least once after study entry at the minimum interval of 6 weeks.

11.2.1.6. Determination of Overall Response by RECIST (version 1.1)

When both target and non-target lesions are present, individual assessments will be recorded separately. The overall assessment of response will involve all parameters as depicted in Table 6.

Table 6. Response Criteria

Target Lesions [1]	Non-Target Lesions[2]	New Lesions [3]	Overall Response
CR	CR	No	CR
CR	Non-CR/Non-PD	No	PR
CR	Not evaluated	No	PR
PR	Non-PD or not all evaluated	No	PR
SD	Non-PD or not all evaluated	No	SD
Not all evaluated	Non-PD	No	NE
PD	Any response	Yes or No	PD
Any response	PD	Yes or No	PD
Any response	Any response	Yes	PD

[1] Measurable lesions only
[2] May include measurable lesions not followed as target lesions or non-measurable lesions
[3] Measurable or non-measurable lesions
Abbreviations: CR = complete response, PD = progressive disease, PR = partial response, SD = stable disease, NE = inevaluable

The best overall response is the best response recorded from the start of the study treatment until the end of treatment taking into account any requirement for confirmation (achievement of both measurement and confirmation criteria, depending on the nature of the study and the protocol requirement). The patient's best overall response assignment will depend on the findings of both target and non-target disease and will also take into consideration the appearance of new lesions. In the case of SD, follow-up measurements must have met the SD criteria at least once after study entry at a minimum interval of 6 weeks.

Table 7. Best overall response when confirmation of CR and PR required

Overall response First time point	Overall response Subsequent time point	BEST overall response
CR	CR	CR
CR	PR	SD, PD or PR[a]
CR	SD	SD provided minimum criteria for SD duration met, otherwise, PD
CR	PD	SD provided minimum criteria for SD duration met, otherwise, PD
CR	NE	SD provided minimum criteria for SD duration met, otherwise NE
PR	CR	PR
PR	PR	PR
PR	SD	SD
PR	PD	SD provided minimum criteria for SD duration met, otherwise, PD
PR	NE	SD provided minimum criteria for SD duration met, otherwise NE
NE	NE	NE

CR = complete response, PR = partial response, SD = stable disease, PD = progressive disease, and NE = inevaluable.
[a] If a CR is truly met at first time point, then any disease seen at a subsequent time point, even disease meeting PR criteria relative to baseline, makes the disease PD at that point (since disease must have reappeared after CR). Best response would depend on whether minimum duration for SD was met. However, sometimes 'CR' may be claimed when subsequent scans suggest small lesions were likely still present and in fact the patient had PR, not CR at the first time point. Under these circumstances, the original CR should be changed to PR and the best response is PR.

Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be reported as "symptomatic deterioration" (see Study Manual specific data collection instructions).
Of note, every effort should be made to document the objective progression even after discontinuation of treatment. Symptomatic deterioration is not a descriptor of an objective response: it is a reason for stopping study therapy. The objective response status of such patients is to be determined by evaluation of target and non-target disease.

In some circumstances, it may be difficult to distinguish residual disease from normal tissue. When the evaluation of a CR depends upon this determination, it is recommended that the residual lesion be investigated by fine needle aspirate or biopsy before assigning a status of CR.
For equivocal findings of progression (e.g. very small and uncertain new lesions; cystic

changes or necrosis in existing lesions), treatment may continue until the next scheduled assessment. If at the next scheduled assessment, progression is confirmed, the date of progression should be the earlier date when progression was suspected.

11.3. Outcomes Research Assessments

Not applicable in this study.

11.4. Safety Assessments

11.4.1. Adverse Event Assessment

11.4.1.1. Definition of Adverse Events

11.4.1.1.1. Adverse Event

According to ICH definition an AE is defined as any untoward medical occurrence in a patient or a clinical trial subject administered a medicinal product and which does not necessarily have to have a causal relationship with the use of the product. An adverse event can therefore be any unfavorable and unintended sign (e.g. an abnormal laboratory finding), symptom, or diagnosis temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

Any untoward medical occurrence, which occurs outside the period of subject follow-up defined in the protocol (28 days after the last treatment administration for the present study, see Section 11.4.1.3), is not considered an AE. Symptoms or medically significant laboratory or instrumental (e.g. electrocardiograph) abnormalities of a pre-existing condition should not be considered an AE. However, occurrences of new symptoms, laboratory or instrumental abnormalities, as well as worsening of pre-existing ones, are considered AEs.

In this trial, the following should not be reported as adverse event:

- The general wording “Progression of the disease”.
  Note: the specific symptoms of the disease worsening have to be considered as adverse events.
- Uncomplicated and asymptomatic abnormal laboratory findings.
  Note: abnormal laboratory findings have to be considered as adverse events when they cause treatment discontinuation or when require clinical intervention (e.g., hospitalization for further investigation or management of the laboratory abnormality).

11.4.1.1.2. Serious Adverse Events

A serious adverse event is an adverse event that falls into one or more of the following categories:

- Results in death
- Is life-threatening, i.e., is an event which, in the view of the Investigator, places the subject at immediate risk of death from the event as it occurred (it does not include an event which hypothetically might have caused death if it was more severe)
- Requires inpatient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity, where disability is defined as a substantial disruption of a person's ability to conduct normal life functions, either reported or defined as per clinical judgment
- Is a congenital anomaly/birth defect (if exposure to product just before conception or during pregnancy resulted in an adverse outcome in the child)
- Is any other important medical event, i.e., may not result in death, be life-threatening, or require hospitalization, but based upon appropriate medical judgment, it may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in the points above. Examples of such events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, and blood dyscrasias or convulsions that do not result in inpatient hospitalization.

A non-serious adverse event is any adverse event that does not meet the criteria listed above for a serious adverse event or the outcome (i.e., subject treatment, life-threatening condition, hospitalization, recovery) cannot be determined with the information provided.

Each adverse event has to be classified by the Investigator as serious or non-serious. This classification of seriousness of the event determines the reporting procedures to be followed.

In this trial the following do not have to be classified as Serious Adverse Events:

- Admission to hospital required by the protocol.
- Events definitely related to disease progression.

All deaths occurring during the reporting period have to be reported as SAEs, even if due to disease progression.

11.4.1.2. Eliciting Adverse Event Information

The Investigator has to report all directly observed adverse events and all adverse events spontaneously reported by the trial subject using concise medical terminology. In addition, each trial subject will be questioned about adverse events at each clinic visit following the signature of the informed consent. The question asked will be "Since your last clinical visit have you had any health problems?"

11.4.1.3. Adverse Event Reporting Period

Adverse event reporting period:

The adverse event reporting period for this trial begins upon signing of informed consent form and ends 28 days after the last treatment administration.

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However, if the patient begins a new anticancer therapy before 28 days after the last dose of study treatment administration, the adverse event reporting period will end at the time the new treatment is started.

All adverse events that occur in trial subjects during the adverse event reporting period must be reported to CLIOSS (delegated by the Sponsor), whether or not the event is considered related to the study treatment.

Adverse event follow up after the end of the reporting period:

The following events should be followed after the end of the reporting period (i.e., 28 days after the last treatment administration):

1. All serious adverse events with outcome “not recovered” or “unknown” at the end of the reporting period.

2. Those non-serious events whose relationship with investigational study treatment has been classified as “Unlikely”, “Possible”, “Probable” and “Definite” with outcome ‘ not recovered’ at the end of the reporting period.

Such events should be followed until they resolve, the patient begins a new anticancer therapy or the Investigator determines, whenever possible, that they have become “chronic” or “stable”. Resolution of such events is to be documented on the appropriate report form.

In addition, if, after the end of the reporting period, suspected serious adverse reactions or deaths are reported to the Investigator and he/she believes that they are related to the investigational product, it is the Investigator’s responsibility to report this suspected serious adverse reactions to CLIOSS Pharmacovigilance. Such suspected serious adverse reactions will be reported using a Serious Adverse Event Report Form or any other way chosen by the Investigator.

After the cut-off date of 31 May 2017 and protocol Amendment 4 approval, SAEs will continue to be reported to CLIOSS Pharmacovigilance for patients continuing treatment, until 28 days after study treatment is discontinued, according to Section 9.5.

11.4.1.4. Reporting Requirements

Each adverse event has to be classified by the Investigator as SERIOUS or NON SERIOUS. The seriousness of the event determines the reporting procedures to be followed. If a serious adverse event occurs, the CLIOSS Pharmacovigilance (delegated by the Sponsor) has to be notified, by Fax (+39 0331 581681) or by e-mail (drugsafety@clioss.com) using the designated form, within 24 hours of awareness of the event by the Investigator. The initial report should be followed by submission of more detailed adverse event information within 5 calendar days after the Investigator first became aware of the serious adverse event. Reporting requirements for adverse events are summarized in the following table.

Table 8. Reporting requirements for adverse events

Gravity	Reporting Time	Type of Report
SERIOUS	Within 24 hours	Initial report on designated serious adverse event form (SAER-F)
	Within 5 calendar days	Follow-up/Final report on designated serious adverse event form (SAER-F)
NONSERIOUS	Per case report form submission procedure	Study case report forms

Serious adverse events should also be recorded on appropriate section of the case report form.

In the rare event that the Investigator does not become aware of the occurrence of a serious adverse event immediately (for example, if an outpatient trial subject initially seeks treatment elsewhere), the Investigator should report the event within 24 hours after learning of it and document his/her first awareness of the adverse event.

Non serious adverse events have to be recorded on the study case report forms.

11.4.1.5. Recording Adverse Events in the Case Report Forms

Information on adverse events must be evaluated by a physician and recorded in a source document such as the hospital file. Adverse events are to be recorded on the case report forms as above specified. The Investigator will also be asked to assess the relationship between the adverse event and the investigational medication.

- Preexisting Conditions

In this trial, a pre-existing condition (i.e., a disorder present before an adverse event occurs and recorded on the pretreatment medical history/physical examination form) should not be reported as an adverse event unless the condition worsens or episodes increase in frequency during the adverse event reporting period.

- Baseline Signs and Symptoms

All Signs and Symptoms (either tumor-related or not tumor-related) collected at baseline should not be reported anymore during the study except in case they worsen in severity or in frequency.

- Procedures

Diagnostic and therapeutic non-invasive and invasive procedures, such as surgery, should not be reported as adverse events. However, the medical condition for which the procedure was performed should be reported if it meets the definition of adverse event. For example, an acute appendicitis that begins during the adverse event reporting period should be reported as the adverse event and the resulting appendectomy should be recorded in the source documents.

After cut-off date of 31 May 2017, AE/SAE data will be no longer collected into the CRF, but assessments information should continue to be collected into the patient's medical notes.

11.4.1.6. Grading of Adverse Event Severity

The severity of adverse events will be graded using the Common Terminology Criteria for Adverse Events (CTCAE, Version 3.0) of the US National Cancer Institute. See the website: http://ctep.info.nih.gov/reporting/ctc.html. For each event, the highest severity grade attained should be reported.
Schedule of adverse events safety assessment is reported in Section 7, Schedule of Events.

For events not reported in the CTCAE Version 3.0, the Investigator will use the grade or adjectives reported in Table 9:

Table 9. Grading of Adverse Event Severity for Events not reported in the CTCAE Version 3.0

Grade	Adjective	Description
Grade 1	Mild	Does not interfere with patient's usual function
Grade 2	Moderate	Interferes to some extent with patient's usual function
Grade 3	Severe	Interferes significantly with patient's usual function
Grade 4	Life-threatening	Results in threatening to life or in an incapacitating disability
Grade 5	Death	Results in death

Note the distinction between the seriousness and the intensity of an adverse event. Severe is a measure of intensity; thus, a severe reaction is not necessarily a serious reaction. For example, a headache may be severe in intensity but would not be classified as serious unless it met one of the criteria for serious events listed in paragraph (see Section 11.4.1.1.2).

11.4.1.6.1. Relationship to the Investigational product

The causality between each adverse event and the investigational product should be classified according to the following terms:

Unrelated: the event is a pre-dose event or is definitely due to causes separate from the administration of the investigational product, i.e.: documented pre-existing condition, concomitant medication, the subject's clinical state or the event is judged as not related and does not fall under any of the above points.

Unlikely: an event for which the exclusion of drug causality seems most plausible, whilst other drugs or underlying disease provide plausible explanation.

Possible: a clinical event, including laboratory test abnormality, with a reasonable temporal sequence from administration of the investigational product and with a known response pattern to the investigational product, but the event could have been caused by subject's clinical state, other therapy administered or diagnostic/interventional procedure. Information on drug withdrawal may be lacking or unclear.

Probable: a clinical event, including laboratory test abnormality, with a reasonable temporal sequence from administration of the investigational product and with a known response pattern to the investigational product, but the event cannot be reasonably explained by the known characteristics of the subject's clinical state, other therapy administered or diagnostic/interventional procedure and for which there is evidence of partial or complete disappearance of the event after withdrawal of the product (positive dechallenge).

Definite: a clinical event that follows a reasonable temporal sequence from administration of the drug or in which the drug level has been established in body fluids or tissues; that follows a known response pattern to the suspected drug; and that is confirmed by improvement on withdrawal (positive dechallenge) or reducing the dose, and reappearance of the event after reintroduction of the drug (positive rechallenge).

11.4.1.7. Exposure In Utero

The Investigator will be asked to submit an Exposure in Utero Form in the cases reported below:

- a trial patient becomes or is found to be pregnant while receiving an investigational medication/product or within **90 days** of its discontinuation
- a partner of a male patient becomes or is found to be pregnant, while the patient is receiving an investigational medication/product or within **90 days** of discontinuing the investigational medication/product.

This must be done irrespective of whether an adverse event has occurred and within 24 hours of awareness of the pregnancy. The information submitted should include the estimated date of delivery (see below for information related to induced termination of pregnancy). However if the patient begins a new anticancer therapy before **90 days** after the last dose of study treatment administration, the exposure *in utero* reporting period will end at the time the new treatment is started.

The Investigator will follow the patient until completion of the pregnancy or until pregnancy termination (i.e., induced abortion) and then notify the CLIOSS Pharmacovigilance of the outcome within 5 days or as specified below. The Investigator will provide this information as a follow up to the initial Exposure in Utero form. The reason(s) for an induced abortion must be specified.

If the outcome of the pregnancy meets the criteria for immediate classification as a serious adverse event (i.e., spontaneous abortion, stillbirth, neonatal death, or congenital anomaly [including that in an aborted fetus]), the Investigator should follow the procedures for reporting serious adverse events, i.e., report the event to Nerviano Medical Sciences (as described in Section 11.4.1.4, Reporting Requirements).

In the case of a live birth, the "normality" of the newborn can be assessed at the time of birth (i.e., no minimum follow-up period of a presumably normal infant must pass before an Exposure in Utero Form can be completed). The "normality" of an aborted fetus can be assessed by gross visual inspection unless pre-abortion laboratory findings are suggestive of a congenital anomaly.

Additional information about pregnancy outcomes that are classified as serious adverse events follows:

- "Spontaneous abortion" includes miscarriage and missed abortion.
- All neonatal deaths that occur within 1 month of birth should be reported, without regard to causality, as serious adverse events. In addition, any infant death after 1 month that the investigator assesses as possibly related to the in utero exposure to the investigational medication should also be reported.

11.4.2. Laboratory Safety Assessments

Laboratory safety assessments will include repeated evaluation of hematology and biochemistry parameters, urinalyses and pregnancy test that after the cut-off date of 31 May 2017 will be done according to routine clinical practice and/or at investigator's discretion. The results of these exams should be recorded only in the patient's medical notes..

Hematology tests include: hemoglobin, erythrocytes (RBC), white blood cell (WBC) with differential count (neutrophils, lymphocytes, monocytes, eosinophils, basophils, differential other cells), platelets (PLTs).
They will be performed during screening period and repeated during treatment between the Days 11 and 14 of all cycles. If toxicity preventing re-treatment results from the Day 11-14 assessment, hematology should be repeated as needed to monitor for recovery.
Hematology tests will be done also on Day 1 of cycles, before study drug administration, only in case toxicity preventing re-treatment resulted from the assessment done within the previous 4 days at the end of the prior cycle.
For patients remaining on treatment for a period of time more than 6 cycles and come back

to the site every 3 cycles, the hematological tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF.

Blood chemistry tests include: sodium, potassium, chloride, magnesium, blood urea nitrogen (BUN) or urea, creatinine, albumin, AST/SGOT, ALT/SGPT, Alkaline Phosphatase (ALP), total bilirubin, creatinine clearance (calculated by the Cockroft and Gault's formula), amylase and lipase.
They will be performed during screening period and repeated during treatment between the Days 11 and 14 of all cycles. If toxicity preventing re-treatment results from the Day 11-14 assessment, blood chemistry should be repeated as needed to monitor for recovery.
Blood chemistry tests will be done also on Day 1 of cycles, before study drug administration, only in case toxicity preventing re-treatment resulted from the assessment done within the previous 4 days at the end of the prior cycle. For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the biochemical tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF.

Coagulation tests include: INR (International Normalized Ratio) and activated partial thromboplastin time (APTT, in seconds). To be done at baseline, at the end of last cycle and, during treatment, at the end of even cycles (between Days 11 and 14). For patients remaining on treatment for a period of time more than 6 cycles and come back to the site every 3 cycles, the coagulation tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF.

Urinalysis includes: pH, dipstick for glucose, protein and blood. It will be performed at baseline and at the end of last cycle. During treatment, should be performed at the end of even cycles (between Days 11 and 14). For patients remaining on treatment for a period of time more than 6 cycles and come back to the site very 3 cycles, the urinalysis tests may be done locally when the visit to the site is not planned (one visit planned to the site every three cycles, 1.5 months) and must be sent to the site for safety evaluation and reported on CRF.

Serum/urine pregnancy test is indicated only for women of reproductive potential and is to be carried out within 7 days prior to the first treatment administration.

The timing and specific requirements for each laboratory safety evaluation are given in Section 7, Schedule of Events.

Specific analyses to be performed in presence of events indicative of, or suggestive of, MTA/HUS (see Section 9.4.9): search of schistocytes in the peripheral blood smear, count of reticulocytes in the peripheral blood, coagulation test, total LDH and 24-hour proteinuria.

Routine Hematology, Blood Chemistry and Urinalysis tests, foreseen by the study protocol, should also be repeated at the time when this evaluation is performed.

11.4.3. Other Safety Assessments

The following safety assessments will also be performed, according to timing reported in Section 7, Schedule of Events:

Physical examination will be done at pretreatment and on treatment at the end of each cycle. Any abnormality at pretreatment should be reported in the Baseline Signs and Symptoms CRF page and on treatment in the AE CRF page. For patients still on treatment at the cut-off date of 31 May 2017, assessments data should be recorded only in the patient's medical notes.

Vital Signs: Blood pressure/pulse (supine) is to be monitored at baseline and on Day 1 of each treatment cycle, before treatment administration; height (in cm or inches) will be measured at baseline only; weight (in kg or pounds) will be assessed at baseline and on Day 1 of each treatment cycle; ECOG performance status will be monitored at baseline and on Day 1 of each treatment cycle; temperature (in °C or °F) will be measured at baseline and on Day 1 of each treatment cycle.

ECG (12-lead) will be carried out at the pretreatment visit and at the end of last cycle. To be repeated during treatment as medically indicated.

Chest X-ray will be carried out, for safety assessment, at the pretreatment visit and at the end of last cycle.

Ophthalmologic examination will include visual acuity test and fundoscopic examination and will be carried out at the pretreatment visit, at the end of even cycles (between Days 11 and 14) and at the end of last cycle by an ophthalmologist. Additional assessments will be performed if clinically indicated.
Ophthalmologic evaluations will be done by the same ophthalmologist for a given patient. For patients still on treatment at the cut-off date of 31 May 2017, data should be recorded only in the patient's medical notes.

Neurological status evaluation: at each scheduled clinical visit (at baseline, at the end of each treatment cycle, and 28 days after the last dose of study drug administration) patients will undergo regular clinical examination, including the assessment of clinical tremors and associated signs and symptoms. Patients should be examined during rest, when assuming various positions and when moving (see also Section 9.4.7, Assessment and Management ofPotential CNS Toxicity). Any sign and symptom of neurological toxicity should be recorded in the Adverse Events CRF pages of the relevant cycle. For patients still on treatment at the cut-off date of 31 May 2017, no AEs will be collected in the CRF, and all evaluations should be recorded only in the patient's medical notes.

11.5. Other Assessments

11.5.1. Confirmation of histological diagnosis

Histological confirmation of diagnosis of thymic carcinoma for all patients will be obtained by an Independent Review Committee during the course of the study, based on paraffin embedded tumor tissue slides retrieved from patients.

11.5.2. Pharmacokinetics

No blood sampling is expected during the study for pharmacokinetics analysis. Blood samples may be collected if, in the opinion of the Investigator and of the Sponsor, an evaluation of the systemic exposure of PHA-848125AC is needed for safety reasons. In such case the IRB will be notified.

If applicable, the actual time of sample collection has to be recorded in the Case Report Form.

11.5.3. Laboratory Exploratory Studies on Tumor Specimens

11.5.3.1. Collection of Tumor Specimen(s)

Paraffin-embedded tissue blocks or derived slides containing the original diagnostic tumor samples from consenting patients will be procured from the institutions where the diagnostic procedures were performed.

Tumor blocks or derived slides will be shipped by a commercial courier under controlled temperature conditions in protective packaging.

11.5.3.2. Molecular characterization of patient's tumors

Molecular analysis of p53, p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-848125AC mechanism of action on paraffin-embedded tissues of consenting patients will be performed in designated facilities in NMS and NIH/NCI.

12. STATISTICAL METHODS

12.1. Sample Size Calculation

In consideration of the exploratory nature of the study, the Simon's optimal 2-stage design [42] is adopted for this single-arm, open-label, multicentre phase II clinical trial.

The total number of evaluable patients for the primary efficacy analysis ranges from 17 (if the trial stops at the end of the 1st step) to 54 (if the trial proceeds up to the completion of the 2nd step). Accounting for a 10%-15% proportion of non evaluable patients, up to 20

patients could be required in the 1st stage of the study and up to 60 patients could be required for completing the trial (1st and 2nd stage).

The primary endpoint of the study is the progression-free survival status at 3 months and the primary efficacy analysis will be performed on the proportion of successes (i.e. patients alive and in a progression-free status at 3 months since treatment start) out of the total number of evaluable patients (PFS-3 rate).

Considering a 33% PFS-3 rate as clinically interesting, against a clinically uninteresting hypothesis of a PFS-3 rate no higher than 17%, the system of hypotheses to be tested is:

$$H_0: p \leq p_0, \quad p_0 = 0.17 \qquad vs. \qquad H_1: p \geq p_1, \quad p_1 = 0.33$$

The analysis will be performed at the overall level $\alpha = 0.05$ (1-sided).
The design below outlined will provide 80% power (1-β) not to reject the treatment as insufficiently effective, if the true PFS-3 rate is 33% or higher.

In the first stage, 17 evaluable patients are to be assessed. If ≤ 3 successes are observed, the study treatment will be considered as providing insufficient evidence of efficacy and the trial will be terminated.

If at least 4 successes are observed in the 1st stage, patients' enrollment will proceed up to an overall enrollment of 54 evaluable patients. At the final analysis, ≥ 14/54 successes (PFS-3 rate ≥ 25.9%) will be required to reject the null hypothesis and suggest that the drug might have an interesting level of efficacy. The progression free survival at 3 months will be evaluated based on the antitumor activity evaluated during the oncologic assessment at 3 months after first drug administration. The oncologic assessment will be performed preferably between 92-98 days from treatment start, but all patients with assessments performed up to 134 days will be considered evaluable for the primary end-point.

H_0 *vs.* H_1	α (1-sided)	power (1-β)	1st Stage (*)		2nd Stage		
			Pts	**STOP and Reject drug**	**Pts**	**Reject drug**	**Do not reject drug**
$p_0 \leq 0.17$ *vs.* $p_1 \geq 0.33$ *(PFS-3 rate)*	0.05	0.80	17 evaluable pts	≤ 3 / 17 successes	54 evaluable pts	≤ 13 / 54 succ. (PFS-3 rate ≤24.1%)	≥ 14 / 54 succ. (PFS-3 rate ≥ 25.9%)
(*) Probability of Early Termination: PET=0.675, if the drug is actually ineffective; Probability to continue the trial > 0.863, if the true PFS-3 rate is at least 33%:							
Expected sample size assuming ineffective drug: 29							
No. of enrolled patients: ≤20 patients in the 1st stage, ≤60 patients overall (if 10%-15% inevaluable patients)							

12.2. Definition of Analyzed Study Populations

For the purpose of the analysis, the following patient populations are defined and the endpoints, to be analyzed in these populations, are specified:

- *Screened Patients:* This population will include all subjects who are screened about their eligibility for the trial, regardless of whether or not they will be enrolled in the study. This population will be evaluated in the analysis of patients' disposition.
- *Enrolled Patients:* This population will include all subjects who are enrolled in the trial, regardless of whether subjects receive the study drug or not. This population will be evaluated in the analysis of patients' disposition.
- *Treated Patients:* The treated patient population consists of all enrolled patients who actually receive at least one study drug administration. This population will be evaluated in the analysis of patient disposition, baseline characteristics, treatment efficacy and safety and treatment exposure.
- *Patients Evaluable for Efficacy Analysis:* This is the patient population for the primary efficacy analysis of PFS-3 rate and consists of all treated patients who fulfill the following additional conditions:
  - They have received histological confirmation of thymic carcinoma by an Independent Review Committee
  - They receive at least 80% of drug in the first two cycles overall.
  - They have baseline and ≥1 on-treatment tumor/oncologic assessment(s) or die before tumor re-assessment.

  If deemed of clinical interest, patient disposition, baseline characteristics, treatment efficacy and safety and treatment exposure will be analyzed also in this population.

12.3. Analyses

12.3.1. Study Conduct and Subject Disposition

Patients' disposition will be presented in frequency distribution tables and individual data listings. All screened patients will be included in this evaluation. Violations of eligibility criteria at study entry as recorded in the relevant sections of the CRF will be documented. Reasons for stopping treatment will be summarized as frequency distribution in the treated patient population and, if clinically interesting, in other patient subsets (e.g. evaluable patients).

12.3.2. Baseline Characteristics

Descriptive statistics of the baseline characteristics will be generated in the treated patient population and, if clinically interesting, in other patient subsets (e.g. evaluable patients). Frequency distributions will be presented for the categorical / categorized variables.

Summary statistics including mean, standard deviation, median, minimum, maximum and the number of assessed patients will be calculated, as appropriate, for the quantitative variables. Individual data will be presented in listings.

12.3.3. Treatment Administration/Compliance

The treatment exposure and the compliance with study treatment will be descriptively analyzed in the treated patient population and, if clinically interesting, in other patient subsets (e.g. evaluable patients). Descriptive statistics (e.g. min, max, mean, standard deviation, and median value) will be calculated on a per-patient basis for the following variables: the number of cycles administered, the overall duration of treatment, the actual and total doses administered, and the absolute and relative dose intensity. Frequency distributions of patients and/or cycles will be used to describe dose modifications, delays and omissions, as well as the reasons for deviation from the planned therapy. These data will be presented as reported in the relevant CRF sections.

12.3.4. Efficacy Analyses

Primary Efficacy Endpoint

For the primary efficacy analysis, the primary endpoint, *i.e.* the PFS-3 rate, will be calculated as the proportion of evaluable patients (see Section 12.2) known to be alive and progression-free at ≥ 3 months since study treatment start out of the total number of evaluable patients (see Section 4.2.1). Decision rules will be applied in the primary efficacy analysis as described in Section 12.1, i.e.:

- At least 4 / 17 evaluable patients alive and progression free at ≥ 3 months are required not to early terminate the trial and reject the drug at the end of the first step.
- At least 14 / 54 evaluable patients alive and progression free at ≥ 3 months are required at the end of the second step to reject the null hypothesis of a PFS-3 rate as low as 17%, at the 1-sided α-level of 0.05.

Supportive analyses of the primary endpoint will include the estimation of the PFS-3 rate together with its exact, two-tail, 95% confidence interval and the estimation of the PFS curve by the Kaplan-Meier method [43] in both the evaluable and the treated patient population.

Other Efficacy Endpoints

The secondary efficacy endpoints outlined in section 4.2.2 and including the confirmed objective response rate, the disease control rate, the duration of response and the overall survival will be analyzed in both the evaluable and the treated patient population as follows:

Objective tumor response rate: Point and 95% confidence interval estimates will be calculated for the objective tumor response rate (confirmed CRs or PRs). The analysis will

be performed in the evaluable and in the treated patient populations. The estimates of the rates of unconfirmed tumor objective responses will also be provided and considered as supportive.

Disease control rate: Point and 95% confidence interval estimates will be calculated for the disease control rate (confirmed CRs / PRs and SD ≥ 6 weeks). The analysis will be performed in the evaluable and in the treated patient populations.

Time-to-event endpoints: For the other secondary endpoints, duration of response (in patients achieving a confirmed objective tumor response by RECIST version 1.1 criteria) and overall survival (OS), individual calculated times will be presented in patients' data listings together with the identification of censored vs. failure status. Kaplan-Meier curves will be estimated for OS in both the evaluable and the treated patient populations.

All collected efficacy data will be presented in individual patients' data listings.

12.3.5. Outcomes Research Analyses

Not applicable in this study.

12.3.6. Safety Analyses

Safety data will be evaluated in the treated patient population and, if clinically interesting, in other patient subsets (e.g. evaluable patients).

The adverse events (AEs) will be coded by the Medical Dictionary for Regulatory Activities (MedDRA) and their severity graded according to the NCI Common Terminology Criteria for Adverse Events (CTCAE) version 3.0. The MedDRA preferred term and system organ class (SOC) will be used to identify and group events in data listings and descriptive analyses. All reported baseline signs and symptoms and on-treatment adverse events will be reported in the patients' data listings. The analysis will address all the events that are recorded on treatment (i.e. from cycle 1, on). The raw incidence of each sign/symptom, of the subsets of events grouped by system organ class and, in general, the raw incidence of any AE, will be calculated. In this analysis, every patient will be accounted for on the basis of the worst CTCAE grade reported during the whole period, within the considered category (*e.g.* the analyzed sign/symptom or system organ class). The following subsets of AEs will also be specifically considered: serious AEs, AEs with CTCAE grade 3-5, AEs with a relationship to study treatment classified by the Investigator as possible or probable or definite and AEs reported as leading to discontinuation from treatment.

Raw incidence of death will be calculated. Deaths will also be described in terms of relationship to the study treatment and according to the time from treatment stop.

Laboratory test values will be graded according to the NCI CTCAE scale, v3.0, whenever possible. For each laboratory test included in the NCI CTCAE system, the incidence of

abnormalities will be evaluated by considering the worst occurrence for each patient throughout the whole treatment period. The frequency distributions of cycles according to the worst CTCAE grade will also be calculated for selected parameters, as clinically indicated. Preliminary conversions of reported values from the originally reported units to a set of predefined units will be applied to facilitate the comparison. For selected parameters descriptive analysis of shifts relative to baseline will be performed if deemed of clinical relevance. Coagulation tests will be evaluated with respect to the corresponding baseline values and to the recorded normal ranges. Urinary parameters will be documented in patient data listings.

Ophthalmologic examinations will be summarized in terms of normal / abnormal findings. Shift tables comparing patients' worst assessments after treatment initiation to the corresponding baseline assessments will be built.

All collected safety data (including vital signs, ECG, ophthalmologic evaluations, laboratory tests and adverse events) will be presented in individual patients' data listings. Identification of clinically relevant values and calculation of changes from baseline will be provided in the listings for those parameters and conditions deemed of clinical relevance.

12.3.7. Analyses of Other Endpoints

12.3.7.1. Baseline Expression of Molecular Markers in Tumor Biopsies

The collection of these data is foreseen only in the consenting patients with available tumor samples.
Descriptive statistics of molecular features of p53, p21, p27, cyclin D1, p75, TRKA and other genes/proteins involved in the PHA-84125AC mechanism of action in the available tumor samples will be presented. Exploratory analyses on the relationship between these markers and treatment efficacy variables (e.g. PFS-3, PFS, objective tumor response, OS) will be performed only if sufficient data are collected.

12.3.7.2. Pharmacokinetics

Plasma levels versus sampling times will be presented in tabular and graphic form, if applicable.

12.3.7.3. Concomitant Medications and Post-Treatment Anti-tumor Therapies

Individual patients' data listings will be prepared to document the administration of concomitant medications.
Patients starting antitumor therapies during follow-up will be identified in a patients' data listing.

12.4. Interim Analysis Plan

Not Applicable in this study.

12.5. Data Monitoring Committee

Not applicable.

13. END OF STUDY

After approval of Amendment 4, the end of the trial is defined as the last visit of the last patient, occurring when all patients have discontinued study therapy.

14. QUALITY CONTROL AND QUALITY ASSURANCE

Monitoring visits to the trial site will be made periodically during the trial to ensure that GCPs and all aspects of the protocol are followed. Source documents will be reviewed for verification of agreement with data on case report forms. The investigator/institution guarantees direct access to source documents by the Sponsor and appropriate regulatory authorities.

The trial site may also be subject to review by the institutional review board (IRB)/independent ethics committee (IEC) to quality assurance audits performed by the Sponsor, and/or to inspection by appropriate regulatory authorities to assure compliance with proper trial conduct.

It is important that the investigator(s) and their relevant personnel are available during the monitoring visits and possible audits or inspections and that sufficient time is devoted to the process

15. DATA HANDLING AND RECORD KEEPING

15.1. Case Report Forms

A case report form is required and should be completed for each included subject. The completed original case report forms are the sole property of Tiziana Life Sciences, PLC and should not be made available in any form to third parties, except for authorized representatives of appropriate regulatory authorities, without written permission from Tiziana Life Sciences.

It is the investigator's responsibility to ensure completion and to review and approve all case report forms. Case report forms must be signed by the investigator or by an authorized staff member. These signatures serve to attest that the information contained on the case report forms is true. At all times, the investigator has final personal responsibility for the accuracy and authenticity of all clinical and laboratory data entered on the case report forms. Subject

source documents are the physician's subject records maintained at the study site. In most cases, the source documents will be the hospital's or the physician's chart. In cases where the source documents are the hospital or the physician's chart, the information collected on the case report forms must match those charts. In some cases, a portion of the source documents for a given study/site may be the case report forms. The investigator must agree which items will be recorded in the source documents and for which items the case report form will stand as the source document.

For patients still on treatment at the cut-off date of 31 May 2017, data should be recorded only in the patient's medical notes.

15.2. Record Retention

To enable evaluations and/or audits from regulatory authorities or Tiziana Life Sciences, PLC, the investigator agrees to keep records, including the identity of all participating subjects (sufficient information to link records, case report forms and hospital records), all original signed informed consent forms, copies of all case report forms, source documents, and detailed records of treatment disposition. The records should be retained by the investigator according to local regulations or as specified in the Clinical Trial Agreement, whichever is longer.

If the investigator relocates, retires, or for any reason withdraws from the study, the Sponsor should be prospectively notified. The study records must be transferred to an acceptable designee, such as another investigator, another institution, or to Tiziana Life sciences, PLC. The investigator must obtain the Sponsor written permission before disposing of any records. .

For patients still on treatment at the cut-off date of 31 May 2017, data should be recorded only in the patient's medical notes.

16. ETHICS

16.1. Institutional Review Board (IRB)/Independent Ethics Committee (IEC)

Approval of the trial protocol, protocol amendments, informed consent forms, and other relevant documents (eg, advertisements), if applicable, must be obtained from the IRB/IEC according to applicable regulatory requirements.

The only circumstance in which an amendment may be initiated prior to IRB/IEC approval is where the change is necessary to eliminate apparent immediate hazards to the subjects. In that event, the investigator must notify the IRB/IEC and the local Nerviano Medical Sciences site personnel as soon as the change has been implemented.

16.2. Ethical Conduct of the Trial

The trial will be performed in accordance with International Conference on Harmonization Good Clinical Practice guidelines, the Declaration of Helsinki (1996 Version), and applicable local regulatory requirements and laws.

16.3. Subject Information and Consent

It is the responsibility of the Investigator to give each subject (or the subject's acceptable representative) full and adequate verbal and written information regarding the objective and procedures of the trial and the possible risks involved. This information must be provided to the subject prior to undertaking any trial-related procedure. The subjects must be informed about their right to withdraw from the trial at any time. Written subject information, approved by the IRB/IEC, must be given to each subject before any trial-related procedure is undertaken. The written subject information must not be changed without prior approval by NMS and the IRB/IEC. Furthermore, it is the responsibility of the Investigator to obtain signed and dated informed consent from all subjects, and a signature from the persons conducting the informed consent discussion, prior to undertaking any trial-related procedure.

A recommended informed consent sample is provided in Appendix 1 of this protocol.

17. SPONSOR DISCONTINUATION CRITERIA

Tiziana Life Sciences, PLC reserves the right to discontinue the trial prior to inclusion of the intended number of subjects, but intends only to exercise this right for valid scientific or administrative reasons. After such a decision, the Investigator must contact all participating subjects within a time period set by the Sponsor. In any case, for patients on treatment, the Sponsor will guarantee the supply of PHA-848125AC until the patients will benefit from this product. As directed by the Sponsor, all study materials must be collected and all case report forms completed to the greatest extent possible.

18. DISSEMINATION AND PUBLICATION OF RESULTS

The conditions regulating dissemination of the information derived from this clinical study are described in the Clinical Trial Agreement. Presentations, abstracts, publications of the study results are to be submitted to the Sponsor for internal review procedure, at least 1 month before submission.

19. REFERENCES

1. NCI PDQ [Physician Data Query] ®, last modified 05/08/2008

2. Malignant thymoma in the United States: demographic patterns in incidence and associations with subsequent malignancies" Int. J. Cancer, 2003; 105(4): 546-51

3. Wright C. Management of thymomas. Crit. Rev. Oncol. Hematol., 2008; 65(2): 109-20

4. Henley JD, Cummings OW, Loehrer PJ Sr. Tyrosine kinase receptor expression in thymomas. J Cancer Res Clin Oncol 2004; 130:222–224.

5. Pan CC, Chen CH, Wang LS, Lee JY, Chiang H. Expression of apoptosis-related markers and HER-2/*neu* in thymic epithelial tumours. Histopathol 2003; 43:165–172.

6. Sasaki H, Fukai I, Kiriyama M, Kaji M, Yano M, Yamakawa Y, Fujii Y. Thymidylate synthase and dihydropyrimidine dehydrogenase mRNA levels in thymoma. Surg Today 2003; 33:83–88.

7. Baldi A, Ambrogi V, Mineo D, Mellone P, Campioni M, Citro G, and Mineo TC. Analysis of Cell Cycle Regulator Proteins in Encapsulated Thymomas. Clin Cancer Res 2005;11(14) July 15, 2005

8. Kim DJ, Yang WI, Kim SH, Park IK, Chung KY. Expression of neurotrophin receptors in surgically resected thymic epithelial tumors. Eur. J. Cardiothorac. Surg., 2005; 28(4): 611-6

9. Kondo K. Optimal therapy for thymoma. J. Med. Invest., 2008; 55(1-2): 17-28

10. Yokoi K, Matsuguma H, Nakahara R, Kondo T, Kamiyama Y, Mori K, Miyazawa N. Multidisciplinary treatment for advanced invasive thymoma with cisplatin, doxorubicin, and methylprednisolone. J. Thorac. Oncol. 2007; 2(1): 73-8

11. Giaccone G. Treatment of malignant thymoma. Curr Opin Oncol, 2005; 17:140-146

12. Kurup A, Burns M, Dropcho S, Pao W, Loehrer PJ. Phase II study of gefitinib treatment in advanced thymic malignancies. JCO 23 (16S, Part I of II, June 1 Supplement), ASCO 2005: Abs. 7068

13. Salter JT, Lewis D, Yiannoutsos C, Loehrer PJ, Risley L, Chiorean EG. Imatinib for the treatment of thymic carcinoma. JCO 26: ASCO 2008 (May 20 Supplement): Abs. 8116

14. Bedano PM, Perkins S, Burns M, Kessler K, Nelson R, Schneider BP, Risley L, Dropcho S, Loehrer PJ. A Phase II trial of erlotinib plus bevacizumab in patients with recurrent thymoma or thymic carcinoma. JCO 26: ASCO 2008 (May 20 Supplement): Abs. 19027

15. Ströbel P, Hartmann M, Jakob A, Mikesch K, Brink I, Dirnhofer S, Marx A. Thymic carcinoma with overexpression of mutated KIT and the response to imatinib. N Engl J Med. 2004 Jun 17;350(25):2625-6.

16. Bisagni G, Rossi G, Cavazza A, Sartori G, Gardini G, Boni C. Long lasting response to the multikinase inhibitor by 43-9006 (Sorafenib) in a heavily pretreated metastatic thymic carcinoma. J Thorac Oncol. 2009 Jun;4(6):773-5.

17. Koizumi T, Takabayashi Y, Yamagishi S, Tsushima K, Takamizawa A, Tsukadaira A, Yamamoto H, Yamazaki Y, Yamaguchi S, Fujimoto K, Kubo K, Hirose Y, Hirayama J, Saegusa H. Chemotherapy for advanced thymic carcinoma: clinical response to cisplatin, doxorubicin, vincristine and cyclophosphamide (ADOC chemotherapy). Am J Clin Oncol, 2002; 25(3): 266-8

18. Magois E, Guigay J, Blancard PS, Margey J, Milleron B, Lher P, Jounieaux V. Multimodal treatment for thymic carcinoma: Report of nine cases. Lung Cancer, 2008; 59 (1): 126-32

19. Igawa S, Murakami H, Takahashi T, Nakamura Y, Tsuya A, Naito T, Kaira K, Ono A, Shukuya T, Tamiya A, Endo M, Yamamoto N. Efficacy of chemotherapy with carboplatin and paclitaxel for unresectable thymic carcinoma. Lung Cancer 2009 Apr 29 [Epub ahead of print]

20. Loehrer PJ, Yiannoutsos C, Dropcho S, Burns M, Helft P, Chiorean EG, Nelson RP. A Phase II trial of pemetrexed in patients with recurrent thymoma or thymic carcinoma. JCO 24 (18S, Part I, June 20 Supplement): ASCO 2006, Abs. # 7079

21. Palmieri G, Merola C, Petillo L, Merola G, Federico P, Leopardo D, Marino M, Montella L. Gemcitabine-capecitabine in platinum-refractory patients with thymic epithelial tumors. ASCO 2009, Abs. # 7590

22. Giaccone G, Rajan A, Carter C, Kelly R, Berman A, Spittler J, Espinoza-Delgado I, Lee M, Trepel J, Loehrer P. Phase II study of the histone deacetylase inhibitor belinostat in thymic malignancies. ASCO 2009, Abs.# 7589

23. Investigator Brochure for PHA-848125AC, Version 7. Nerviano Medical Sciences; 2011. Document No CDKO-125a-IB7.

24. Sherr CJ, Roberts JM. Living with or without cyclins and cyclin-dependent kinases. Genes Dev 2004;18(22):2699-2711.

25. Malumbres M, Barbacid M. To cycle or not to cycle: a critical decision in cancer. Nat Rev Cancer 2001;1(3):222-231.

26. Shapiro GI. Cyclin-Dependent Kinase Pathways As Targets for Cancer Treatment. J Clin Oncol 2006;24(11):1770-1783.

27. Malumbres M, Barbacid M. Cell cycle kinases in cancer. Curr Opin Genet Dev 2007;17(1):60-65.

28. Benson C, Kaye S, Workman P, Garret M, Walton M, de Bono J. Clinical anticancer drug development: targeting the cyclin-dependent kinases. Br J Cancer 2005;92(1):7-12.

29. Swanton C. Cell-cycle targeted therapies. Lancet Oncol 2004;5(1):27-36.

30. Slingerland J, Pagano M. Regulation of the Cdk inhibitor p27 and its deregulation in cancer. J Cell Physiol 2000;183:10-17.

31. Desdouets C, Bréchot C. p27: a pleiotropic regulator of cellular phenotype and a target for cell cycle dysregulation in cancer. Pathol Biol 2000;48:203-210.

32. Adams PD. Regulation of the retinoblastoma tumor suppressor protein by cyclins/cdks. Biochimica et Biophysica Acta 2001;1471:M123-133.

33. Hondermarck H. Nerve growth factor: the dark side of the icon. Am J Pathol. 2008 Apr;172(4):865-7.

34. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, Rubinstein L, Shankar L, Dodd L, Kaplan R, Lacombe D and Verweij J. New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). Eur J Cancer. 2009 Jan;45(2):228-47.

35. Marik PE. The management of Status Epilepticus, Chest 2004;126:582-591.

36. Working Group on Status Epilepticus. Treatment of convulsive status epilepticus. Recommendations of the Epilepsy Foundation of America's Working Group on Status Epilepticus. JAMA 1993;270:854-859.

37. Marmor MF, Holder GE, Seeliger MW, and Yamamoto S. Standard for Clinical Electroretinography (2004 update). Document approved by the International society for Clinical Electrophysiology of Vision in Nagoya, Japan on April 4, 2003. Documenta Ophthalmologica 2004;108:107-114.

38. Wong TY, Mitchell P. Hypertensive Retinopathy. N Engl J Med 2004;351:2310-2317.

39. Desramé J, Duvic C, Béchade D et al. Is the frequency of hemolytic uremic syndrome as a complication of gemcitabine underestimated? The role of systematic screening. Gastroenterol Clin Biol. 2006 Feb;30(2):332-4.

40. Kris MG, Hesketh PJ, Somerfield MR, Feyer P, Clark-Snow R, Koeller JM, Morrow GR, Chinnery LW, Chesney MJ, Gralla RJ, Grunberg SM. American Society of Clinical Oncology guideline for antiemetics in oncology: update 2006. J Clin Oncol 2006;24(18): 2932-2947.

41. The Antiemetic Subcommittee of the Multinational Association of Supportive Care in Cancer (MASCC). Prevention of chemotherapy– and radiotherapy-induced emesis: results of the 2004 Perugia International Antiemetic Consensus Conference. Ann Oncol 2006;17:20-28.

42. Simon R. Optimal two-stage design for phase II clinical trials. CCT 1989; 10:1-10.

43. Kaplan EL, Meier P. Nonparametric estimation from incomplete observations. J Am Stat Assoc 1958; 53:457-481.

APPENDICES

Appendix 1. Recommended Informed Consent Form

Appendix 2. The Distribution of Active Bone Marrow in the Adult

Appendix 1. Recommended Informed Consent Form

Patient Information and Informed Consent

Phase II study of oral PHA-848125AC in patients with thymic carcinoma previously treated with chemotherapy.

Protocol No. CDKO-125a-006

Institution/Hospital:

Investigator:

Sponsor: Tiziana Life Sciences, PLC

You are being asked if you want to take part in a research study using an experimental drug called PHA-848125AC. Whether you take part or not is entirely your decision. You may decide to participate to the study or not to take part at all. If you decide to participate to the study, you may end your participation at any time. In any case, your decision will not affect your regular medical care or any benefit to which you are otherwise entitled. If there is anything you do not understand about this study after reading this information, please ask your study doctor or study staff member.

Why is this study being done?

PHA-848125AC is a new drug selected for clinical evaluation because of its antitumor activity in experimental models. This drug acts by blocking specific proteins (called cyclin-dependent kinases) involved in the control of cell division. In the case of tumor cells such control is altered and leads to abnormal cell replication. Experiments have shown that the addition of PHA-848125AC may block this uncontrolled proliferation. Treatment with PHA-848125AC might possibly have a positive effect on your disease.

This drug was evaluated in a number of clinical studies in the US and in Europe, with different schedules of administration. As of November 2011, PHA-848125AC has been administered to approximately 200 patients with various solid tumors. Currently, two studies are ongoing, including this one.
The most relevant side effects encountered so far in the evaluation in humans were transient nausea and/or vomiting and /or diarrhea and neurological toxicity, mainly represented by transient tremors and ataxia (incoordination). Fatigue has been frequently reported. See also section “Risks” here below.

Two objective tumor responses were obtained so far in the US study, in two out of three patients affected by thymic carcinomas.

This is the first study with PHA-848125AC specifically dedicated to patients with thymic carcinoma.

This study is called phase II clinical study. Patients will be all treated at the same dose of PHA-848125AC; this dose has been selected based on results of the first clinical study in solid tumors patients where patients treated at this dose had mild/moderate reversible adverse events, represented by nausea, vomiting, diarrhea and tremors.
The dose of PHA-848125AC will never be increased and could be reduced based on the tolerability of the patient.

The purpose of this study is to help researchers to:

- evaluate if PHA-848125AC can be effective as a treatment for thymic carcinoma

- get additional information on the side effects of PHA-848125AC.

If you decide to take part in this clinical study, you will get PHA-848125AC every day for 7 consecutive days and then you will not take the drug from day 8 to day 14. A period of 14 days (2 weeks) is called a cycle. You will take the study medicine by oral route. The number of cycles you may receive will depend both on the observed side effects and the evolution of your disease.

During your stay in the study you will be also followed regularly for the status of your disease.

This study is conducted in US and Europe. A total of about 60 patients will participate.

This is what will happen if you decide to be in this study

Your doctor will talk to you about your disease, ask you questions about your health, and give you a physical exam. Blood will be taken from one of your veins and you will be asked to give a urine sample for laboratory tests. Electrocardiogram and ocular examination will be carried out. The ocular examination includes a fundoscopic examination and a visual acuity test. The funduscopic examination will consist of projection of a light from an ophthalmoscope through the pupil to view the back (fundus) of the eyeball. The examination will take few minutes and will not be painful. The exam may be preceded by drops in the eye, to dilate pupils. Visual acuity test measures the smallest letters that you can read on a standardized chart at a standard distance.

During the course of this study, blood samples will be periodically taken. These regular blood tests, as well as other examinations, will be performed to check that the study medicine is not adversely affecting your bone marrow, kidneys, liver and other organs. At each sampling, the volume of blood taken for these analyses will be about 5 mL.
There is the possibility, for safety reasons, that a few additional 5 ml blood samples may be required by the Investigator and the Sponsor to investigate the presence of PHA-848125AC

in your body.
Part of your tumor tissue (paraffin embedded block), previously taken at the time of diagnosis of your disease, will be used to confirm histological diagnosis by an independent group of experts.

You will also have other tests and activities to monitor your disease and the possible effects of the treatment as shown in the following table:

Schedule of Visits/Tests

Activity	Before you begin the study	During the study	End of treatment	After end of treatment period
Discussion With Nurse/Study Doctor	**X**	**X**	**X**	**X**
Medical/Oncology history/Physical exam	**X**	**X**	**X**	
Confirmation of histological diagnosis		**X**		
Vital signs	**X**	**X**	**X**	**X**
Pregnancy Test	**X**			
Blood Samples	**X**	**X**	**X**	
Urine Samples	**X**	**X**	**X**	
Chest X-Ray and ECG	**X**		**X**	
Ophthalmologic visit	**X**	**X**	**X**	
Other exams to follow your disease (CT scan or MRI)	**X**	**X**	**X**	**X**
Anything the study doctor believes is medically needed	**As needed**			

Your doctor will ask you to record your daily drug intake in a diary.

The effect of the study medicine on your disease will be assessed periodically by a CT scan or MRI.

The administration of PHA-848125AC will continue as long as you can tolerate the experimental compound and if there are no indications that the disease is worsening. If unacceptable side-effects occur, the study medicine will be stopped and appropriate medical care will be provided. If your disease becomes worse during treatment, you will be told, the treatment will be stopped and alternate medical care will be provided.

Experimental Aspects of the Trial

Analysis of the basal molecular markers of the tumor

Alteration of some genes is very common in human cancers, including thymic carcinoma.

It would be of value to know if the cells of your tumor have this kind of alteration, to study the relationship between the affected genes and the study treatment efficacy.

To do so, a sample of your tumor tissue is needed: it is possible that a sample of your tumor has already been collected in the past, if you had a surgery or a biopsy, or that it may be requested by your doctor to understand the status of your disease before entering this study.

If you give your consent (or you consented for this study earlier), part of your tumor sample will be analyzed also in this study to determine some molecular features of p53, p21, p27, cyclin D1, p75, TRKA, and other genes/proteins involved in the PHA-84125AC mechanism of action.

The possible correlation between these molecular features and the effect of PHA-848125AC on the disease may be of help to support in the future the choice of the best treatment option for patients having thymic carcinoma.

Your Responsibilities

If you decide to be in this study, you will have to:

- Keep all scheduled appointments.
- Tell your doctor about any other medicines that you take, even if it is medicine you buy without a prescription or it is an herbal remedy.
- Tell your doctor about any medical problems you have.
- Adopt effective contraceptive methods if you are sexually fertile while on the study and for 90 days after you leave the study.
- Avoid, if you are female, becoming pregnant while on the study and for 90 days after you leave the study.
- Avoid, if you are male, father a child while on the study and for 90 days after you leave the study.
- Avoid breast-feeding while you are taking the study medicine.

If you do not follow the items listed above, you may be removed from the study.

Other Treatments

There may be other ways to treat your disease. Your doctor can tell you more about these other treatments and their side effects.

Benefits

The medicine that you are given might help you with your disease, but this cannot be guaranteed. However, even if you does not benefit personally from being in this study, having you take part in this study will help researchers find out if PHA-848125AC will help other people who are affected by thymic carcinoma.

Of note, two objective tumor responses were obtained so far in a previous study performed in the US, in two out of three patients affected by thymic carcinomas.

If you are in this clinical study, you will get free medical examinations and laboratory tests that are not part of the routine care for your condition. You will not have to pay for the medicine.

Risks

The medicine you receive may cause side effects in human beings. Since PHA-848125AC is an experimental drug, these side effects in humans are not fully known. If you have any side effects, your doctor may stop or temporarily interrupt the treatment, or give you other drugs to make them less serious and to make you more comfortable.

The most common side effects seen in animals after treatment with PHA-848125AC were: anemia, thrombocytopenia, neutropenia, lymphopenia, diarrhea, nausea and vomiting. These events were almost completely reversible. Ocular and central nervous system (CNS) toxicity were reported in animals at high doses or after prolonged administration, as well as congestions and/or hemorrhages (including hemorrhagic diarrhea, hemorrhages in the heart), gastrointestinal (GI) tract ulceration, bleeding and infection.

The most relevant side effects encountered so far in the evaluation in humans were transient nausea and/or vomiting and /or diarrhea and neurological toxicity, mainly represented by transient tremors and ataxia (incoordination) and less frequent episodes of dizziness. The neurological symptoms gradually improved in all patients after temporary suspension of the study drug. Fatigue is a side effects reported in about half of patients. Hematological toxicity was sporadically reported, consisting especially in mild/moderate anemia (low hemoglobin) and neutropenia (low neutrophils). A prolonged elevation in liver enxymes was occasionally observed with the schedule adopted in the present study (7 days on / 7 days off), however, with a more prolonged treatment schedules (eg a schedule encompassing 14 consecutive days every 3 weeks), alterations of liver function tests were more frequently reported. Some episodes of transient elevation of lipase (a pancreatic enzyme), without clinical symptoms, have been reported, as well as mild transient elevations of creatinine (a parameter indicative of renal function status). There could be the possibility that the daily dose of study drug is reduced after recovery of adverse effects in order to continue study treatment at a better tolerated dose.
Ocular toxicity was never encountered so far, with the exception of a worsening of the

electroretinography examination during treatment compared to baseline in one diabetic patient treated at 24 mg/m^2/day for 21 consecutive days and in a second patient treated at 72 mg/m^2/day for 14 consecutive days (dose then reduced to 48 mg/m^2/day in the two subsequent cycles). Both patients, however, did not show any ocular clinical symptoms. Two cases of TMA/HUS (Thrombotic Microangiopathy/Haemolytic Uraemic Syndrome) occurred in two different patients, both treated for more than a year, in a study administering PHA-848125AC together with Gemcitabine, a drug approved for the treatment of different tumor types, which is known to produce HUS. Both patients recovered.

There may be some side effects that researchers do not yet know about. If researchers learn important new information that could change your decision to continue to take part in the study, your study doctor will tell you in a timely manner. PHA-848125AC might not work for you or might not work as well as other medicines you have received in the past.

Your study doctor will be checking you closely to see if you are having any side effects. Your study doctor may give you medicines to help with side effects. Many side effects stop after the treatment is stopped. Your study doctor may also change the schedule or the amount of study treatment to reduce side effects.

The risks of having blood taken from your vein include pain, bruising, or infection at the site where the blood was taken, and fainting.

Stopping Early

You may decide to leave the study at any time without any disadvantage to you. You are not obliged to provide reason for your withdrawal.

Your doctor may also decide to remove you from the study if you fail to follow instructions, for medical reasons, or for other reasons. There is also the possibility the study could be stopped by the Sponsor before your participation is complete. If this would happen and if this is because of non-medical reasons, and you are benefiting by the study treatment, the supply of PHA-848125AC will be guaranteed. From that moment on, the treatment will be under the responsibility of your doctor.

If you leave the study for any reason, you will be asked to return to the study doctor to bring back any left study medicine and to complete the final study activities. All study information that is collected about you until the time you leave the study and refuse further evaluations will be kept and used by the Sponsor.

Pregnancy

You must avoid becoming pregnant while you are in this study. If you become pregnant while you are in the study or within 90 days after completing your participation, you must

tell your study doctor. Your doctor must then report the outcome of your pregnancy to the Sponsor.

Similarly, if you are male, you must avoid making a partner become pregnant while you are in the study. If you make a partner pregnant while you are in the study, or within 90 days after completing your participation, you must tell the study doctor. The study doctor must then report this and also the outcome of the pregnancy to the Sponsor.

Compensation

Every effort will be made to prevent research related injury. If any injury is due to your participation in this study you will receive emergency care. The Sponsor has insurance that covers any research-related injury you should suffer. The term "research-related injury" means physical injury caused by the product or procedures required by the trial which are different from the medical treatment you would have received if you had not participated in the trial.

You will not be paid for participating in this trial. You will not need to pay for the study medicine and for any tests or procedures which are "research-related".

Confidentiality and Data Privacy

Your doctor will record the study information described above on case report forms that will be sent to the study Sponsor. In these case report forms, you will only be identified with a study subject number. All records in which your name appears will be kept by your doctor and be kept confidential. Your name will never appear on any Sponsor forms or in a report or publication.

The study Sponsor will use the information recorded on the case report form to support new approvals for medical use of the PHA-848125AC and for any required regulatory reporting of side effects. The information may be transferred and exported by the Sponsor to other entities within the Sponsor organization, to its business partners, and to regulatory authorities, on a global basis.

Authorized persons from the Sponsor, governmental regulatory agencies, and/or the ethics committees may need to look at the medical records kept by your doctor to verify the information on the case report forms.

By signing this informed consent form you agree to allow this review of your records as well as to the processing and transfer of your personal information as described above.

Storage and Analysis of Biological Samples

Eventual blood samples collected for safety reason to investigate the presence of PHA-848125AC in your body will be sent to the Sponsor for analysis in Accelera Srl and storage in a secure place to ensure protection of your confidentiality.

Tumor samples to be analyzed for the characterization of molecular markers will be sent to the Sponsor for analysis and storage in a secure place to ensure protection of your confidentiality.

If You Have Questions

For questions about your responsibilities or study activities, please contact:

_____ at _____

For questions about your rights as a participant in this study, please contact

_____ at _____

For questions about possible injury due to activities in this study, please contact

_____ at _____

Patient's Statement

- I voluntarily agree to participate in this study
- I understand that the study Sponsor may stop the study at any time
- I have read and understood this statement of informed consent and the risks described
- I understand that I will receive a signed and dated copy of this consent form
- I understand that I may withdraw my consent at any time
- I also voluntary agree ☐ or not agree ☐ to have my tumor tissue analyzed for molecular features.
- I have had a chance to ask questions and understand the answers given to all of my questions

Signature of study patient: _____ Date: _____

Printed name of study patient: _____

****

Signature of person conducting informed consent discussion: _____

Printed name of person conducting informed consent discussion: _____

Date: _____

Appendix 2. The Distribution of Active Bone Marrow in the Adult

The distribution of bone marrow in the adult

Site	Marrow Weight (g)	Fraction Red Marrow - Age 40	Red Marrow Weight -Age 40 (g)	% Total Red Marrow
Head			**136.6**	**13.1**
Cranium	165.8	0.75	124.3	
Mandible	16.4	0.75	12.3	
Upper Limb Girdle			**86.7**	**8.3**
2 Humerus, head & neck	26.5	0.75	20.0	
2 Scapulae	67.4	0.75	50.5	
2 Clavicles	21.6	0.75	16.2	
Sternum	39.0	0.6	**23.4**	**2.3**
Ribs			**82.6**	**7.9**
1 pair	10.2	All 0.4	4.1	
2	12.6		5.0	
3	16.0		6.4	
4	18.6		7.4	
5	23.8		9.5	
6	23.6		9.4	
7	25.0		10.0	
8	24.0		9.6	
9	21.2		8.5	
10	16.0		6.4	
11	11.2		4.5	
12	4.6		1.8	
Vertebrae (Cervical)			**35.8**	**3.4**
1	6.6	All 0.75	5.0	
2	8.4		6.3	
3	5.4		4.1	
4	5.7		4.3	
5	5.8		4.4	
6	7.0		5.3	
7	8.5		6.4	
Vertebrae (Thoracic)			**147.9**	**14.1**
1 pair	10.8	All 0.75	8.1	
2	11.7		8.8	
3	11.4		8.5	
4	12.2		9.1	
5	13.4		10.1	
6	15.3		11.5	
7	16.1		12.1	
8	18.5		13.9	
9	19.7		14.8	
10	21.2		15.9	
11	21.7		16.3	
12	25.0		18.8	

The distribution of bone marrow in the adult

Site	Marrow Weight (g)	Fraction Red Marrow - Age 40	Red Marrow Weight -Age 40 (g)	% Total Red Marrow
Vertebrae (Lumbar)			**114.1**	**10.9**
1 pair	27.8	All 0.75	20.8	
2	29.1		21.8	
3	31.8		23.8	
4	32.1		24.1	
5	31.4		23.6	
Sacrum	194.0	0.75	**145.6**	**13.9**
Lower limb girdle			**273.0**	**26.1**
2 os coxae	310.6	0.75	233.0	
2 femoral head & neck	53.0	0.75	40.0	
Total	**1497.7**		**1045.7**	**100.0**

R.E. ELLIS - The Distribution of Active Bone Marrow in the Adult. Phy. Med. Biol. 5, 255 – 258, 1961