Supplementary Data

Inclusion criteria

1. Histologically proven advanced solid tumors, refractory to conventional treatment, or for which no conventional therapy existed or was declined by the patient.
2. Life expectancy of at least 12 weeks.
3. World Health Organisation (WHO) performance status of 0, 1 or 2.
4. Hematological and biochemical indices within the ranges shown below. These measurements were to be performed before the patient received the first dose of AT13148.

Laboratory Test	Value required
Hemoglobin	≥9.0 g/dL
Absolute neutrophil count	≥1.5 ×10^9/L
Platelet count	≥100 ×10^9/L
Fasting glucose	≤7 mmol/L
Either: Serum bilirubin or: Alanine aminotransferase and aspartate aminotransferase	≤1.5 × upper limit of normal (ULN) ≤2.5 × ULN unless raised due to tumor in which case up to 5 × ULN was permissible
Either: Calculated creatinine clearance Or: Isotope clearance measurement	≥50 mL/min ≥50 mL/min (uncorrected)

5. Adequate lung function indicated by a resting oxygen saturation level (on air) ≥94%, a carbon monoxide (CO)-transfer factor >60%.
6. 18 years or over.
7. Written (signed and dated) informed consent and capable of co-operating with treatment and follow-up.

Exclusion Criteria

1. Radiotherapy (except for palliative reasons), endocrine therapy (except luteinizing hormone releasing hormone agonists for prostate cancer), immunotherapy or chemotherapy during the previous 4 weeks (6 weeks for nitrosoureas, Mitomycin-C) and 4 weeks for investigational medicinal products) before the planned first dose of AT13148. (If the planned date of the first dose changed the Sponsor was to be contacted for advice).
2. Ongoing toxic manifestations of previous treatments. Exceptions to this were alopecia or certain Grade 1 toxicities, which in the opinion of the Investigator and the Sponsor did not exclude the patient.

3. Symptomatic brain metastases (if present they had to have been stable for >3 months).
4. Ability to become pregnant (or already pregnant or lactating). However, those female patients who had a negative serum or urine pregnancy test before enrolment and agreed to use two highly effective forms of contraception (oral, injected or implanted hormonal contraception and condom, had an intra-uterine device and condom, diaphragm with spermicidal gel and condom) during the trial and for 6 months afterwards were considered eligible.
5. Male patients with partners of child-bearing potential (unless they agreed to take measures not to father children by using one form of highly effective contraception [condom plus spermicide] during the trial and for 6 months afterwards). Men with pregnant or lactating partners were advised to use barrier method contraception (e.g. condom plus spermicidal gel) to prevent exposure to the foetus or neonate.
6. Major thoracic or abdominal surgery from which the patient had not yet recovered.
7. At high medical risk because of non-malignant systemic disease including active uncontrolled infection.
8. Known to be serologically positive for hepatitis B, hepatitis C or human immunodeficiency virus.
9. History of severe auto-immune disease or known allergy to peanuts.
10. Concurrent hypotension defined as a baseline supine blood pressure systolic <90 mmHg.
11. Patients receiving anti-hypertensive treatment or treatment with beta-blockers or rate-limiting calcium agents. A washout period of 5 × half-life of the drug was to be applied following withdrawal of any of these treatments.
12. Concurrent congestive heart failure, prior history of class III/IV cardiac disease (New York Heart Association), prior history of cardiac ischemia or prior history of cardiac arrhythmia. Coronary angioplasty or stenting in the previous 12 months.
13. Patients with a known left ventricular ejection fraction <50%. A multi-gated acquisition (MUGA) scan or echocardiogram (ECHO) had to be performed in all patients.
14. Concurrent diabetes requiring treatment (diet-controlled diabetes was acceptable).
15. Patients with prior bone marrow transplant or those having had extensive radiotherapy to >25% of bone marrow within 8 weeks of the trial.
16. A history of or underlying interstitial lung disease.
17. Any other condition which in the Investigator’s opinion did not make the patient a good candidate for the clinical trial.
18. Was a participant or planned to participate in another interventional clinical trial whilst taking part in this trial. Participation in an observational trial or interventional clinical trial which did not involve administration of an investigational medicinal product was acceptable.

Pharmacokinetic analytical method

AT13148 and the deuterated (D4) Internal Standard (IS) were extracted from 100 µL human plasma (harvested in lithium heparin) using protein precipitation with methanol. Samples were centrifuged and the supernatant was separated and analyzed.

Chromatography was carried out using Phenomenex Kinetex™ C18 X-B column (2.6 µM, 50mm, x 2.1 mm id) with a gradient mobile phase constituting of 0.1% formic acid and methanol. The flow rate was 0.6 ml/min and the run time 5 minutes. AT13148 and IS were ionized using electrospray interface in positive ion mode.

Detection of analytes was via tandem mass spectroscopy (MS/MS) in the multiple reaction monitoring (MRM) mode. The transitions m/z 296.16-117.10 and m/z 302.2-249.85 were monitored for AT13148 and IS respectively.

Using deuterated AT13148 as an internal standard, concentrations were calculated from a standard curve. The upper limit of quantification (ULOQ) was 1623 nM and the lower limit of quantification was 4.05 nM.