Preclinical Safety Evaluation of MTL-005, a Novel Metalloporphyrin Radiosensitiser

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Abstract

MTL-005 (copper meso-5, 15-Bis[3-[(1,2-Dicarba-closododecaboranyl)methoxy]phenyl]-meso-10,20-dinitroporphyrin) is under development as a radiosensitizer / radioenhancer for use in head and neck cancer. In viva it slows growth of murine squamous cell carcinomas and adenocarcinomas . Given i.p. to mice, it is cleared from the circulation and taken up by both normal tissues and tumors, with tumor concentrations up to 6 times higher than in muscle, skin, and brain. It concentrates in the liver and spleen and can be detected in both tumors and liver for at least 62 days post administration.

4 i.v. toxicology studies were carried out in rats and 2 in dogs. MTL-005 was dissolved in a mixture of dimethylacetamide, Solutol HS15 and PEG300 and diluted with 0.9% saline. Components of this vehicle are known to have toxic effects, especially in dogs. Rats were given 0, 20, 40 and 60 mg/kg (divided over 2 consecutive days)and dogs with 2 doses of 0, 1, 5 or 10 mg/kg given 14 days apart. Toxicities observed included body weight loss, reduced food consumption , dark discolouration of several organs, and reversible gastric epithelial hyperplasia (in rat at doses up to 30 mg/kg/day); and inflammatory changes at the infusion site, liver changes, and the death of 1 dog at 10 mg/kg. The vehicle may have contributed to the body weight, food consumption, and liver effects, as most of these signs were also observed in the vehicle controls. The MTD in rats was 40 mg/kg but a NOAEL could not be established due to the body weight losses observed both treated and control groups.

Dogs tolerated 1 or 5 mg/kg well but the vehicle contributed to the effects seen at 10 mg/kg. Liver histopathology findings and plasma chemistry changes associated with the vehicle were seen in all groups and liver histopathology findings persisted for 28 days. Injection site changes were reversible over 28 days. Again, a NOAEL could not be established .

These preclinical data were sufficient for regulatory approval of a Phase 1 clinical study of MTL-005 as an adjunct to radiotherapy in patients and this has now been initiated.

Safety Pharmacology

Although safety pharmacology studies are not mandated by ICH S9 to support studies in patients with late stage cancer disease, the core battery of single-dose, in-vivo safety pharmacology studies from ICH S7 have been performed, relating to the cardiovascular, respiratory and central nervous systems. The studies, which were GLP-compliant, showed no treatment-related adverse effect on any of the three systems, as detailed below.

ENDPOINT	SPECIES; N/GROUP*	ROUTE OF ADMINISTRATION	DOSES (mg/kg)	COMMENTS
Cardiovascular system	Dog; 5	IV infusion	0, 1, 5, 10	No treatment-related effects on haemodynamic or cardiographic intervals. Transient increase in blood pressure and heart rate noted in all groups and considered to be vehicle-related (histamine release mediated by Solutol)
Respiratory system	Rat; 5	IV bolus	0, 10, 15, 20	No treatment-related effects on respiratory parameters
Central nervous system	Rat; 5	IV bolus	0, 10, 15, 20	No treatment-related effects on locomotor activity, pain response or body temperature

Nonclinical Toxicity Studies

Dose-Range-Finding Studies (Non-GLP)

Non-GLP dose-range-finding (DRF)/maximum tolerated dose (MTD) studies, using IV infusion/bolus as the route of administration, were performed in SD rats and Beagle dogs.

The MTD in the rat was considered to be 30 mg/kg/day over 2 days and that in the dog 4 doses at 10 mg/kg administered over 2 weeks on Days 1 and 2 and 14 and 15.

STUDY	N/GROUP	POSOLOGY	COMMENTS
Rat MTD	Phase B: 5 M/F	4 doses of 40 mg/kg over 2 weeks on Days 1 and 2 and 15 and 16 (infusion)	Bodyweight loss; decreased food consumption; decreased thymus and testis weight; darkened tissue appearance testicular and prostatic atrophy; lymphoid depletion; pigmented macrophages and thymic atrophy. Cmax 600-1300 µg/mL
Rat DRF/MTD	LD: 5 F,HD: 5M/F	Doses of 20 and 30 mg/kg on two consecutive days with a 3-day recovery period (bolus)	Slight bodyweight loss and reduced food consumption – partial recovery by D5. Dark discolouration of several organs. No histopathology undertaken. MTD 30 mg/kg for 2 days. For 30 mg/kg dose on D1: Cmax for males = 600-800 µg/mL; females 700-900 µg/mL
Dog DRF/MTD	Phase B: 1 M/F	4 doses at 10 mg/kg administered over 2 weeks on Days 1 and 2 and 14 and 15 (infusion)	Minimal changes in bodyweight and food consumption; markedly increased ALP, ALT and ASP; high triglycerides and changed albumin/globulin ratio, suggestive of changes in hepatic function. No histopathology. MTD considered to be10 mg/kg four times over 2 weeks.

GLP-Compliant Dog Toxicity Study

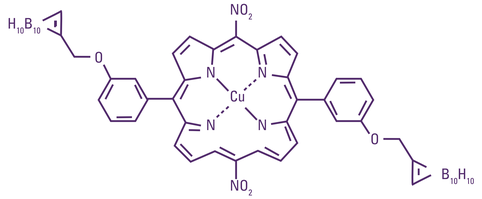
Three groups of 3 male and 3 female beagle dogs were initially scheduled to receive MTL 005 by IV infusion at dose levels of 1, 5 or 10 mg/kg on Days 1 and 2 (dose 1) and 14 and 15 (dose 2) over 2 weeks. A control group received the vehicle for the same duration as for the animals treated with the high dose. An additional 2 males and 2 females were assigned to the control and high-dose group for a subsequent 28-day reversibility assessment.

Following administration on Days 1 and 2, adverse reactions including decreased body weight were observed in both control and high-dose animals. In addition, high plasma ALT activities on Day 11 (males)/13 (females) were also observed in some control and high-dose animals. Consequently, only those animals that were fit for treatment on Day 14 were dosed using a reduced infusion period of 12 minutes. In order not to jeopardise the reversibility assessment this commenced earlier (from Day 3) for control females and animals at 10 mg/kg. The control males commenced recovery from Day 16.

DAY	NUMBER OF ANIMALS DOSED / NUMBER OF ANIMALS SURVIVING							
	Group 1M (Control)	Group 2M (1 mg/kg)	Group 3M (5 mg/kg)	Group 4M (10 mg/kg)	Group 1F (Control)	Group 2F (1 mg/kg)	Group 3F (5 mg/kg)	Group 4F (10 mg/kg)
1	5/5	3/3	3/3	5/5	5/5	3/3	3/3	5/5
2	5/5	3/3	3/3	5/5	5/5	3/3	3/3	5/5
14	2/5	3/3	3/3	0/3	3/3	3/3	3/3	1/3
15	2/5	3/3	3/3	0/3	0/3	3/3	3/3	0/3

Introduction

MTL-005 (copper meso-5, 15-Bis[3-[(1,2-Dicarba-closododecaboranyl)methoxy]phenyl]-meso-10,20-dinitroporphyrin; structure shown below) is under development as an enhancer of radiotherapy, initially for use in head and neck cancer. MTL-005 does not have a direct pharmacodynamic effect, but it combines two features that enhance tumour responses to radiotherapy: porphyrin moieties are known to be tumour-seeking and a number of metalloporphyrins act as radiosensitizers.



MTL-005 significantly enhances the effectiveness of radiotherapy of the SCCVII squamous cell carcinoma and EMT6 mammary carcinoma implanted in mice, markedly slowing tumour growth and increasing the number of controlled tumors. Given i.p. to mice, it is cleared from the circulation and taken up by both normal tissues and tumors, with tumor concentrations up to 6 times higher than in muscle, skin, and brain. It also concentrates in the liver and spleen and can be detected in both tumors and liver for at least 62 days post administration.

The present non-clinical safety studies, consistent with the recommendations of ICH S9, were conducted to support an application for a Clinical Trial Authorisation (CTA) from the UK Medicines and Healthcare Products Regulatory Agency (MHRA) for the first clinical trial of MTL-005 in head and neck cancer. All animal studies used the clinical formulation, in which MTL-005 is first dissolved in a vehicle of 20% (w/v) Solutol HS15 (Macrogol 15-hydroxystearate), 10% (w/v) polyethylene glycol (PEG) 300 (Macrogol 300) in N,N dimethylacetamide (DMA) giving a solution which is then diluted with an appropriate amount of sterile saline. Each millilitre of the dose solution contains 12.5 mg MTL-005 with excipients Solutol (0.1 mL), Macrogol (0.05 mL) and DMA (0.35 mL).

Definitive GLP-Compliant Studies

GLP-Compliant Rat Toxicity Study

The rat study involved two IV bolus injections over 2 days followed by a 3-day observation period and a 14-day recovery period to assess reversibility.

	NUMBER OF ANIMALS						
	MAIN STUDY		RECOVERY GROUPS		SATELLITE TK ANIMALS		DOSE (MG/KG/DAY FOR 2 DAYS)
GROUP	MALE	FEMALE	MALE	FEMALE	MALE	FEMALE	
1	10	10	5	5	6	6	0 (VEHICLE)
2	10	10			6	6	10
3	10	10			6	6	20
4	10	10	5	5	6	6	30

Following administration of 10, 20, and 30 mg/kg/day MTL-005, salivation was noted in a few animals up to 10 minutes post-dosing and was no longer present after 30 minutes. Dark tails and darkened extremities were noted for a few animals given 20 and 30 mg/kg/day immediately following dosing and the effects were generally no longer present after 24 hours. In all animals, including controls, there was reduced body weight gain, but by Day 5 the majority of the animals were gaining weight. Food consumption data were variable and showed no consistent trend. There were no treatment-related changes in organ weights, ophthalmology, haematology, coagulation, clinical chemistry, or urinalysis. Following macroscopic examination of animals dosed at 20 and 30 mg/kg/day, dark discolouration was apparent in the liver, several lymph nodes, and the ovaries. Animals dosed at 10 mg/kg/day had dark discolouration in a few lymph nodes. Minimal to mild epithelial hyperplasia with or without mild gastritis was observed microscopically in the non-glandular stomach in a few males dosed at 30 mg/kg/day. After the recovery period, dark discolouration was still present at necropsy in the liver and several lymph nodes in animals given 30 mg/kg/day, and in the ovaries of females that received 30 mg/kg/day. There was complete recovery in relation to stomach histology findings.

In TK animals, systemic exposure to MTL 005 increased in a supra-proportional manner in relation to dose. Plasma concentrations of MTL-005 were generally greater in males than in females, which was most evident at the low dose on Day 1.

Although a No Observed Adverse Effect Level (NOAEL) could not be established, owing mainly to the body weight losses observed in animals that received MTL-005 and those that were treated with the vehicle only, doses up to 20 mg/kg/day for 2 consecutive days appeared to be well tolerated.

GLP-Compliant Dog Toxicity Study Cont.

IV administration of MTL-005 and of the vehicle was associated with inflammatory changes at the infusion site and also resulted in the premature sacrifice of one high-dose female animal on Day 2 3.5 hours after administration of MTL-005. The volume of vehicle administered as well as the vehicle components contributed to the effects seen in dogs treated with the vehicle alone or with MTL 005 at 10 mg/kg. The low and mid doses were well tolerated. Liver histopathology findings (centrilobular accumulation of mononuclear inflammatory cells, indicative of previous hepatocyte loss) and plasma chemistry changes (increased AP and ALT) were observed after the treatment period in all animals, MTL-005-dosed as well as in vehicle controls, and liver histopathology findings were still present after the 28-day recovery period. Injection-site effects (which were not seen in vehicle-treated animals) were partially reversed after the 28-day recovery period.

The principal target organ was identified as the liver and the pattern of responses suggests that centrilobular hepatocyte damage was most probably induced by the vehicle rather than MTL-005. Although a NOAEL could not be established under the conditions of the study, doses of 1 and 5 mg/kg on days 1, 2, 14 and 15 seemed to be reasonably well tolerated.

Discussion and Conclusions

Results from both rat and dog toxicity studies have identified the liver as the principal target organ (especially in the dog), with partial reversibility over 28 days. The pattern of effects in the dog strongly suggests that most of the observed hepatotoxicity was related to the vehicle rather than to MTL 005. N, N-Dimethylacetamide has been identified as hepatotoxic in dogs, albeit with a higher NOAEL (94 mg/kg/day) than encountered in these studies.

The non-clinical toxicity data presented here are considered to be consistent with the recommendations in ICH S9 and to support the first-in-man clinical trial of MTL-005, an open, Phase 1, exploratory study of radiosensitization in patients with advanced carcinoma of the head and neck: Part 1 to evaluate safety in a dose-escalation design in patients indicated for palliative radiotherapy, Part 2 to evaluate safety and efficacy in a dose-escalation design in patients indicated for cis-platin chemoradiotherapy. In addition, the doses for this clinical trial have been selected to ensure that the starting dose is below the equivalent doses that were tolerated in animal studies. This protocol received a CTA from the UK MHRA in 2012 and the first patients were enrolled from September 2013 onwards.