ASX Announcement

DEP theranostic presented at international oncology meeting

Melbourne, Australia; 16 October 2023: Starpharma (ASX: SPL, OTCQX: SPHRY) today provides a copy of the poster that showcases the results from a study of DEP® HER2-zirconium, Starpharma's HER2-targeted radiodiagnostic candidate, which demonstrated a favourable biodistribution profile, with excellent imaging contrast between tumour and normal tissues, as well as rapid uptake and high levels of tumour accumulation in a HER2+ breast cancer model. These results were announced by Starpharma in July 2023[1].

The poster was presented over the weekend in Boston, US, at the International Conference on Molecular Targets and Cancer Therapeutics, co-hosted by the American Association of Cancer Research (AACR), the National Cancer Institute (NCI) and the European Organisation for Research and Treatment of Cancer (EORTC) from 11 to 15 October 2023.

Starpharma's DEP® HER2-zirconium is a radiodiagnostic product that belongs to the rapidly growing radiotheranostic market. DEP® HER2-zirconium is designed to specifically diagnose, stage, and monitor HER2+ cancers with greater sensitivity, meaning that patients suffering from these cancers could be diagnosed earlier, more accurately, and monitored more closely during cancer treatment.

Starpharma CEO, Dr Jackie Fairley, said: "We are pleased to be presenting results on DEP® HER2-zirconium, our HER2-targeted radiodiagnostic candidate at this AACR international conference and to see the data on this product generate such positive interest. Radiotheranostics are an exciting new area for the application of our DEP® platform, which represents a significant global market opportunity that is expected to be US$4.2 billion by 2030[2]. This poster is one of three which Starpharma presented in Boston."

The poster is appended.

[1] ASX Announcement dated 21 July 2023: DEP® HER2-radiodiagnostic shows imaging benefits
[2] https://www.giiresearch.com/report/dmin1316304-global-theranostics-market.html

About Starpharma

Starpharma Holdings Limited (ASX: SPL, OTCQX: SPHRY) is a world leader in dendrimer technology for medical applications. As an innovative Australian biopharmaceutical company, Starpharma is focused on developing and commercialising novel therapeutic products that address significant global healthcare needs. Starpharma boasts a strong portfolio of products, partnerships, and intellectual property.

Starpharma's innovative technology is based on proprietary polymers called dendrimers, which are precise, synthetically manufactured, nanoscale molecules. The unique properties of dendrimers – including their size, structure, high degree of branching, polyvalency, and water solubility – are advantageous in medical and pharmaceutical applications.

Starpharma uses its dendrimer technology to develop novel therapeutics and to improve the performance of existing pharmaceuticals. Starpharma's portfolio includes multiple clinical-stage oncology products, which utilise its Dendrimer Enhanced Product ('DEP®') drug delivery technology, and marketed products, including VIRALEZE™ and VivaGel® BV, which utilise SPL7013, a proprietary dendrimer with antimicrobial properties.

Starpharma's DEP® drug delivery platform is being used to enhance the effectiveness of existing and novel therapies and to reduce drug-related toxicities through controlled and specified drug delivery.

In addition to Starpharma's internal DEP® programs, Starpharma has multiple DEP® partnerships with international biopharmaceutical companies, including AstraZeneca (oncology), MSD (Antibody-Drug Conjugates), Chase Sun (anti-infectives), and other world-leading pharmaceutical companies. Due to the broad applicability and optionality of Starpharma's DEP® platform, partnered DEP® programs have the potential to generate significant future milestones and royalties.

Starpharma's topical antiviral nasal spray, VIRALEZE™, is now registered in more than 35 countries*, including Europe, the UK, and Asia. Starpharma's novel non-antibiotic vaginal gel, VivaGel® BV, for the treatment of bacterial vaginosis (BV) and prevention of recurrent BV, is registered in more than 50 countries, including in the UK, Europe, Southeast Asia, South Africa, Australia and New Zealand.

For more information about Starpharma, visit www.starpharma.com or connect with Starpharma on LinkedIn.

WE Communications
Hannah Howlett
+61 450 848 064
WE-AUStarPharma@we-worldwide.com

Starpharma Holdings Limited
Dr Jackie Fairley, Chief Executive Officer
Justin Cahill, CFO and Company Secretary
+61 3 8532 2704
investor.relations@starpharma.com
4-6 Southampton Crescent
Abbotsford Vic 3067

Disclosure
This ASX Announcement was authorised for release by the Chair, Mr Rob Thomas.

Forward-Looking Statements

This document contains certain forward-looking statements, relating to Starpharma's business, which can be identified by the use of forward-looking terminology such as "promising", "plans", "anticipated", "will", "project", "believe", "forecast", "expected", "estimated", "targeting", "aiming", "set to", "potential", "seeking to", "goal", "could provide", "intends", "is being developed", "could be", "on track", "outlook" or similar expressions, or by express or implied discussions regarding potential filings or marketing approvals, or potential future sales of product candidates. Such forward-looking statements involve known and unknown risks, uncertainties and other factors that may cause actual results to be materially different from any future results, performance or achievements expressed or implied by such statements. There can be no assurance that any existing or future regulatory filings will satisfy the FDA's and other authorities' requirements regarding any one or more product candidates nor can there be any assurance that such product candidates will be approved by any authorities for sale in any market or that they will reach any particular level of sales. In particular, management's expectations regarding the approval and commercialization of the product candidates could be affected by, among other things, unexpected trial results, including additional analysis of existing data, and new data; unexpected regulatory actions or delays, or government regulation generally; our ability to obtain or maintain patent or other proprietary intellectual property protection; competition in general; government, industry, and general public pricing pressures; and additional factors that involve significant risks and uncertainties about our products, product candidates, financial results and business prospects. Should one or more of these risks or uncertainties materialize, or should underlying assumptions prove incorrect, actual results may vary materially from those described herein as anticipated, believed, estimated or expected. Starpharma is providing this information as of the date of this document and does not assume any obligation to update any forward-looking statements contained in this document as a result of new information, future events or developments or otherwise. Clinical case studies and other clinical information given in this document are given for illustrative purposes only and are not necessarily a guide to product performance and no representation or warranty is made by any person as to the likelihood of achievement or reasonableness of future results. Nothing contained in this document nor any information made available to you is, or shall be relied upon as, a promise, representation, warranty or guarantee as to the past, present or the future performance of any Starpharma product.

A HER2 targeted polylysine dendrimer nanoparticle radiotheranostic demonstrates excellent tumor accumulation, rapid clearance from circulation, and promising performance in PET-CT imaging

Cameron N. Johnstone,[1] Graham P. Heery,[1] Brian D. Kelly, [1] James Humphries,[2,3] Dewan T. Akhter, [2,3] Kristofer J. Thurecht, [2,3] Daniel Yuen, [4] Michael Giannis, [1] Ken Yong, [1] Gareth Chadwick, [1] Helena Barker, [1] Anne Cargill, [1] Anthony Eglezos, [1] Alex Castellarnau[1], Jeremy R.A. Paull [1], Richard Hufton[1]

[1] Starpharma Pty Ltd, Abbotsford, Victoria 3067, Australia; [2] Centre for Advanced Imaging, [3]ARC Research Hub for Advanced Manufacture of Targeted Radiopharmaceuticals, University of Queensland, St Lucia, Queensland 4072, Australia; [4] Monash Institute of Pharmaceutical Sciences, Monash University, Parkville, Victoria 3052, Australia.

A147

Background

- Development of targeted radiotheranostics for human epidermal growth factor receptor type 2 positive (HER2⁺) tumors has lagged advances made in other receptor systems such as prostate specific membrane antigen (PSMA) in prostate cancer.
- Myelosuppression is a dose limiting toxicity in clinical studies of Trastuzumab monoclonal antibody-based radioconjugates due to slow clearance of this high molecular weight radiotherapeutic from the circulation.[1]
- Conjugation of cancer therapeutics such as taxanes, irinotecan/SN38, or Bcl2-family inhibitors to highly optimized dendrimer (DEP®) nanoparticles improves pharmacokinetics (PK), tumor uptake and antitumor efficacy *in vivo*.[2]
- DEP® dendrimer nanoparticles can be used as targeted radiotheranostics with potential to achieve superior PK, enhanced tumor killing, and reduced toxicity (improved therapeutic index) compared to traditional antibody-based radiotheranostics.[3]
- A radio-imaging and biodistribution study of the HER2-targeted dendrimer-based radiodiagnostic, DEP® HER2-zirconium, was conducted in mice implanted with HER2⁺ tumors.
- The study compared performance of DEP® HER2-zirconium in positron emission tomography-computed tomography (PET-CT) imaging, and in the kinetics of tumor and normal tissue uptake, to radiolabeled Trastuzumab control.

Materials and Methods

- A HER2 targeted VHH (single domain antibody [sdAb], or antigen binding fragment of heavy chain only camelid antibody) was covalently linked to a generation 4 dendrimer with 16 deferoxamine (DFO) chelation groups on its surface (DEP® HER2_DFO(16)).
- Trastuzumab was covalently linked to DFO (Trastuzumab_DFO(2)).
- Human and dog HER2 receptor extracellular domains (ECDs) were immobilised (3 µg/mL) onto a CM5 chip and surface plasmon resonance (SPR) analysis of binding conducted using a Biacore 8K+ instrument. Biacore Insight Evaluation software was used to evaluate the binding kinetics, using the in-built single cycle kinetics analysis. Data were fit to a 1:1 binding model.
- DEP® HER2_DFO(16) and Trastuzumab_DFO(2) were successfully radiolabeled with ^{89}Zr to high specific activity.
- Female BALB/c nude mice were inoculated subcutaneously with BT474 HER2⁺ breast cancer cells. When tumors reached 150mm , a single dose (3MBq) of DEP® HER2_^{89}Zr (DEP® HER2-zirconium) or Trastuzumab_^{89}Zr was administered intravenously. Uptake was evaluated in tissues by *ex vivo* gamma scintillation counting (n=3/group) from 4h-12d. *In vivo* biodistribution (n=3/group) was determined using quantified PET-CT data (4h-5d). PET-CT images were obtained on day 2 and 4.

Figure 1A: Schematic representation of the monoclonal antibody control bioconjugate. The anti-HER2 humanized monoclonal antibody, Trastuzumab, was covalently linked (through a DBCO linker) to approximately two chelators (one on each of the heavy chains). DFO was used for the PET isotope, ^{89}Zr, and DOTA for therapeutic isotope, ^{177}Lu.

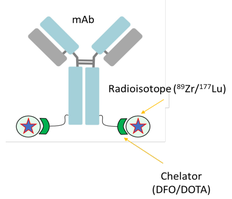
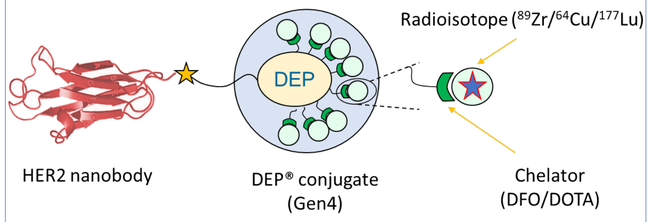


Figure 1B: Schematic representation of the DEP® HER2 radiotheranostics platform. The lead bioconjugate consists of HER2 targeting nanobody clone 2D3 linked via an engineered C terminal cysteine residue to a generation 4 polylysine dendrimer. The generation 4 dendrimer is covalently linked, through the epsilon amino group of lysine, to 16 chelator molecules (16x DFO for PET isotope ^{89}Zr, 16x DOTA for PET isotope ^{64}Cu, or therapeutic isotope ^{177}Lu).



Results

Table 1: SPR analysis of interaction between free anti-HER2 nanobody (clone 2D3), unlabelled DEP® HER2 radiotheranostic bioconjugates, or unlabelled Trastuzumab bioconjugate and the ECDs of either human (left) or dog (right) HER2 receptor. The equilibrium dissociation constants (K_D) are expressed in nM.

		Human HER2 ECD			Dog HER2 ECD		
Construct	**MW (kDa)**	**K_a ($M^{-1}s^{-1}$)**	**k_d (s^{-1})**	**k_D (nM)**	**K_a ($M^{-1}s^{-1}$)**	**k_d (s^{-1})**	**k_D (nM)**
Free HER2 VHH	13.7	7.12E+05	1.13E-03	**1.59**	6.99E+05	1.95E-03	**2.79**
DEP® HER2_DFO(16)	50.8	2.13E+05	5.35E-04	**2.51**	1.15E+05	7.59E-04	**6.62**
DEP® HER2_DOTA(16)	47.6	4.11E+05	6.42E-04	**1.56**	3.88E+05	1.16E-03	**2.98**
Trastuzumab_DFO(2)	150	2.48E+05	2.84E-04	**1.14**	nd	nd	nd

Table 2: Radiochemistry parameters. Assessment of radiochemical purity (RCP), radiolysis, and serum stability of DEP® HER2 radioconjugates and Trastuzumab radioconjugate. RCP of Trastuzumab_^{89}Zr determined by radio-size exclusion chromatography (SEC)-HPLC (radio-SEC-HPLC) while RCP of the smaller DEP® HER2_^{89}Zr and DEP® HER2_^{64}Cu bioconjugates was determined by radio-reverse phase (RP)-HPLC (radio-RP-HPLC). All three bioconjugates showed excellent (> 95%) RCP at t=0. (Auto)radiolysis was measured by incubation in PBS for 5 days (for ^{89}Zr radioconjugates) and 1 day (for the ^{64}Cu radioconjugate). Serum stability was measured by incubation in 50% serum for 5 days (for ^{89}Zr radioconjugates) and 1 day (for the ^{64}Cu radioconjugate). Both DEP® HER2 radioconjugates were resistant to radiolysis and demonstrated excellent stability in serum (RCP of both > 90%).

Bioconjugate	**Specific Activity MBq per µg**	**Molar Activity MBq per mol**	**Percentage RCP (t=0)**	**Percentage RCP (t=5d) (PBS)**	**Percentage RCP (t=5d) (50% serum)**
DEP® HER2_^{89}Zr	0.1	2.03 x 10^{10}	100.0	94.2	93.6
Trastuzumab_^{89}Zr	0.034	2.03 x 10^{10}	95.0	75.2	48.5
Bioconjugate	**Specific Activity MBq per µg**	**Molar Activity MBq per mol**	**Percentage RCP (t=0)**	**Percentage RCP(t=1d) (PBS)**	**Percentage RCP (t=1d) (50% serum)**
DEP® HER2_^{64}Cu	3.85	5.48 x 10^{12}	98.2	92.8	93.1

Figure 2A: *Ex vivo* tissue biodistribution of DEP® HER2_^{89}Zr in mice bearing subcutaneous human BT474 HER2⁺ breast tumors. Radioconjugate was injected at t=0. Biodistribution was evaluated between 4h and 12d. Mean ± SEM (n=3). **Sustained high level tumor uptake (1d-5d) was observed, along with fast clearance from blood.**

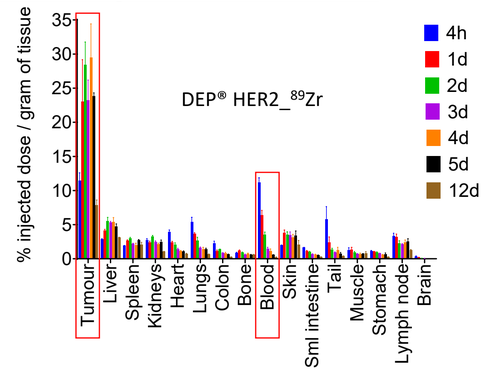


Figure 2B: *Ex vivo* tissue biodistribution of Trastuzumab_^{89}Zr in mice bearing subcutaneous human BT474 HER2⁺ breast tumors. Radioconjugate was injected at t=0. Biodistribution was evaluated between 4h and 12d. Mean ± SEM (n=3). **Sustained high level tumor uptake (1d-12d) was observed with slower clearance from blood.**

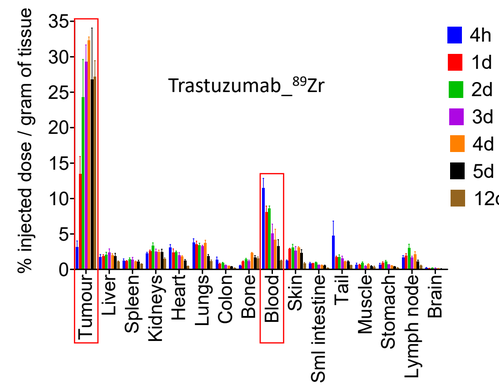


Figure 3: Comparison of blood levels and tumor:blood ratios between DEP® HER2_^{89}Zr and Trastuzumab_^{89}Zr radioconjugates. DEP® HER2_^{89}Zr demonstrated faster clearance from blood (left y axis) and sustained elevated tumor:blood ratios (right y axis) compared to Trastuzumab_^{89}Zr.

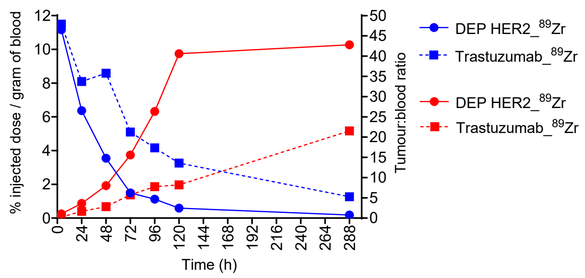


Figure 4: Maximum intensity projection (MIP) PET-CT images of BT474 HER2⁺ tumor-bearing mice dosed with either DEP® HER2_^{89}Zr or Trastuzumab_^{89}Zr. Radioconjugates administered at t=0. Representative mice are shown at day 2 (left side) or day 4 (right side) after injection. The scale bar (% ID/g) is shown to the right. **In addition to high level uptake in tumor, signals were observed in heart for Trastuzumab_^{89}Zr (which is likely to reflect higher levels in the circulation), and liver for DEP® HER2_^{89}Zr, as indicated. Deposition of ^{89}Zr in shoulder and hip joints was prominent for Trastuzumab_^{89}Zr but not for DEP® HER2_^{89}Zr (arrows).**

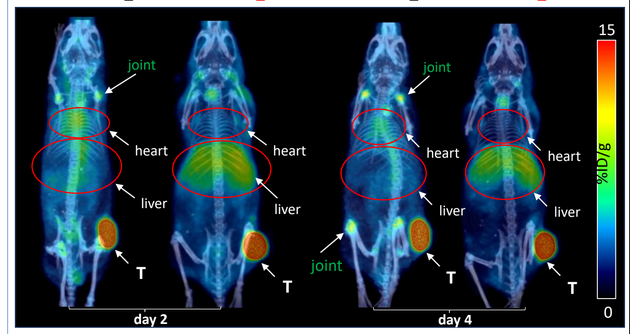
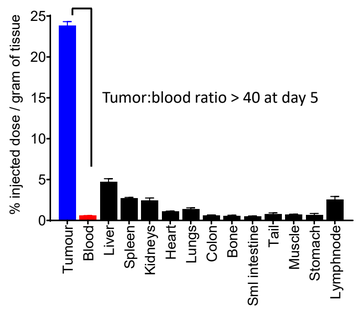


Figure 5: DEP® HER2_^{89}Zr resulted in high tumor:blood ratios.



Conclusions

DEP® HER2_^{89}Zr (DEP® HER2-zirconium) demonstrated imaging benefits in a HER2+ breast cancer model:

- More rapid tumor accumulation and superior PK vs. HER2 monoclonal antibody, labelled with zirconium, Trastuzumab_^{89}Zr ;
- A favourable biodistribution profile, with excellent imaging contrast between tumor and normal tissues
- Highly desirable "fast-in"/"fast-out" kinetics, with rapid accumulation in tumor and rapid clearance from the bloodstream;
- High tumor:organ ratios, delivering excellent specificity in imaging the tumor in HER2+ breast cancer
- High tumor:blood ratios, suggesting potential advantages of DEP® technology for radiotherapeutic applications.

References:

1. Bhusari P. *et al.,* (2017) Int. J. Cancer. 140(4):938-947
2. Patterson C.M., *et al.* (2021) Commun. Biol. 4(1):112
3. Akhtar, N., *et al.* (2023) J. Pharm. Sci. 112(3):844-858

Acknowledgements: We thank Dr. Martina Jones from the National Biologics Facility (NBF) at the University of Queensland for conducting SPR experiments (https://www.nationalbiologicsfacility.com/). We also thank the ARC Research Hub for Advanced Manufacture of Targeted Radiopharmaceuticals (https://www.amtarhub.com.au/).