OSPEDALE
SAN RAFFAELE

PATENTED TECHNOLOGIES FOR LICENSING

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Via Olgettina 60

Milan, Italy

www.research.hsr.it/en/business-development

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Sector
Gene Therapy

Key Publication
Marinozzi M. *et al.*, J. Med. Chem 2017

Development stage
Preclinical Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights

The Technology
Portfolio of 16 novel steroidal stigmasterol and ergosterol derivatives that function as Liver X Receptor (LXR) agonists.

Side-chain modified ergosterol and stigmasterol derivatives as Liver X Receptor modulators (LXR)

I.R.C.C.S. Ospedale San Raffaele
Gruppo San Donato

CLINICAL NEED

The past two decades have seen an exponential increase in the number of studies on the physiological roles of mammalian oxysterols, as well as on their contribution to the pathogenesis of different diseases. The breakthrough was the identification of a specific subset of oxysterols as endogenous ligands of Liver X Receptor α and β isoforms (LXRs). Given the action of LXRs as whole-body cholesterol sensors and key regulators of lipogenesis, oxysterols have the potential to assume a key role in the modulation of diverse pathways in lipid metabolism, glucose homeostasis, reproduction, development, inflammation, and immunity. Accordingly, LXRs and their ligands are being intensely studied as potential therapeutic targets for diverse diseases such as lipid disorders, chronic inflammation, autoimmunity, neurodegenerative disease, and cancer. Up to now, development of LXR modulators has been mainly focused on non-steroidal compounds. Although some non-steroidal, high potency compounds have been discovered (e.g., T0901317, GW3965), they demonstrate a low or null gene-selectivity and induce lipogenic effects by increasing liver and circulating triglyceride levels. Consequently, these compounds have limited clinical use, thus creating an opportunity in the market for steroid based LXR modulators endowed with higher specificity and less toxicity.

PRODUCT & TECHNOLOGY

The present invention relates to a portfolio of 16 novel steroidal stigmasterol and ergosterol derivatives that function as potential Liver X Receptor (LXR) agonists for the treatment of diseases associated with LXR, such as cancer, inflammation, metabolic and autoimmune diseases. LXR agonists induce the expression of target genes (i.e. ABCA1), which are involved in cholesterol homeostasis. In the liver, LXR activation promotes the biosynthesis of fatty acids by inducing the expression of SREBP-1c, as well as several downstream genes in the SREBP-1c pathway, including FASN and SCD1. Importantly, no compound up-regulated the mRNA levels of FASN and SCD1.

SOLUTION & BENEFITS

The novel steroidal stigmasterol and ergosterol derivatives here described could be employed as potential Liver X Receptor (LXR) agonist for the treatment of diseases associated with LXR, such as cancer, inflammation, metabolic, and autoimmune diseases. In particular, these compounds show (i) high LXR selectivity, (ii) high gene selectivity for cholesterol homeostasis rather than for lipogenesis, and (iii) less toxic effect than non-steroidal compounds. Indeed, substantial efforts have been dedicated to the identification of LXR ligands able to turn on ABC transporter genes, without affecting lipogenic genes levels. In this context, the inventors demonstrated that all sixteen different compounds produced and tested were able to induce ABCA1 expression, with a particularly strong induction by four specific compounds when compared to a positive control (T0901317). According to pre-clinical experimental evidence, the derivatives 13, 19, 20, and 25, being strong inducers of ABCA1 and poor activators of SREBP1c and SCD1 in the U937 cell line, proved to be very promising derivatives for further clinical development.

MARKET & COMPETITION

We are currently seeking potential commercial partners with a strong pipeline in LXR modulators and small molecule therapeutics to develop new therapeutic agents for the treatment of LXR-related diseases. Currently, Lead Pharma, Innovimmune, and Inspirna are developing LXR agonists for diverse applications; from dermatology to cancer, with the furthest in a Phase II clinical trial for SCLC (NCT02922764).

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Side-Chain Modified Ergosterol and Stigmasterol Derivatives as Liver X Receptor modulators	WO2019/021122 US20200157135	Pending	EP USA

Blood-brain-barrier permeabilizing agent S-NGR-TNF to promote chemotherapy penetration in primary and secondary tumors of the CNS

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Sector
Oncology

Key Publication
Ferreri et al. Blood 2019;
Ferreri et al. Blood Adv 2020;
Corti et al. Mol Pharm. 2020.

Development stage
Clinical Development

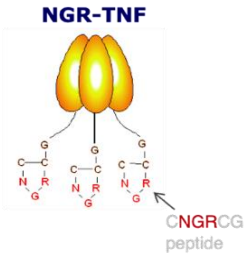
Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

CLINICAL NEED

Primary diffuse large B-cell lymphoma of the central nervous system (PCNSL) is a rare and highly aggressive neoplasm. It accounts for about 2% to 5% of all primary brain tumours and is associated with a poor prognosis, with half of patients dying in the first three years of follow-up. This is mostly due to the presence of the blood-brain tumor barrier (BBTB), a major obstacle for anti-cancer drug penetration to the tumor tissue. Thus, treatment of PCNSL consists of combinations of a few drugs able to penetrate the CNS tissues, which must be delivered at high dose, requiring hospitalization, and a consequently higher risk of toxicity. R-CHOP (rituximab, cyclophosphamide, doxorubicin, vincristine and prednisone) is the standard treatment of diffuse large B-cell lymphomas diagnosed in other organs, resulting in high cure rates. This is a worldwide used combination, delivered in out-patient setting and associated with an excellent safety profile. However, R-CHOP drugs exhibit a low capability to cross the BBTB, which results in an insignificant therapeutic effect on PCNSL. A strategy aimed to increase vascular permeability in the CNS tissues could allow a diffuse use of R-CHOP to treat PCNSL, with relevant advantage in favor of patients and cancer centers.

PRODUCT & TECHNOLOGY

NGR-TNF consists of human TNF fused to the CNGRCG (NGR), a peptide ligand of a CD13 isoform overexpressed in tumor vasculature. Low-dose NGR-TNF interacts, via NGR and TNF domains, with CD13 and TNF receptors, respectively, on tumor vessels, but not with normal vessels (lacking CD13), thus inducing the permeabilization of the BBTB and promoting the penetration of anticancer drugs. Clinical grade NGR-TNF has been produced and used in 18 clinical studies involving >1000 patients with different indications. Recent research advances at HSR have led to an improvement of NGR-TNF through a novel composition of matter patent.

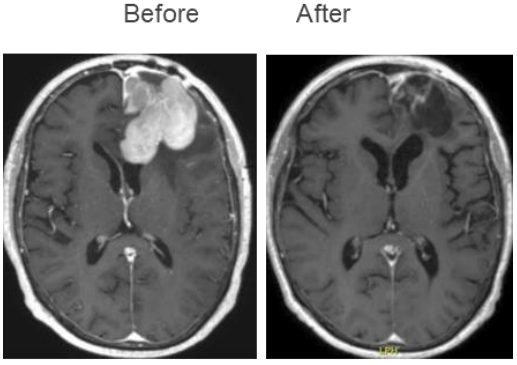


PRECLINICAL RESULTS:

NGR-TNF significantly potentiated chemotherapy in murine models of lymphoma, melanoma and glioblastoma (S-NGR-TNF). In these models, NGR-TNF enhanced chemotherapeutic drug penetration and lymphocyte infiltration and exerted synergistic effects with active and adoptive immunotherapy.

CLINICAL RESULTS (PHASE I/II TRIALS):

A Phase I/II trial in 28 patients with relapsed/refractory PCNSL revealed that the NGR-TNF/RCHOP treatment is active, with confirmed tumor responses in 21 (75%) patients (complete response in 11 patients; photo, right), indicating that the primary endpoint has been largely reached. NGR-TNF did not interfere with drug pharmacokinetics, and, importantly, showed an excellent safety profile, with no cases of unexpected toxicity. These encouraging results are in line with increased vascular permeabilization demonstrated with modern neuroimaging techniques in the enrolled patients.



MARKET & COMPETITIVE CONTEXT

The medical need in PCNSL is supported by over 100 past and ongoing deals for an overall value of billions of dollars. Various therapeutic approaches in PCNSL are being assessed at the preclinical stage: Protein Cereblon (CRBN) Activator, Tyrosine Protein Kinase BTK Inhibitor, Cyclin Dependent Kinase 4/6 Inhibitor, Phosphatidylinositol 3 Kinase Inhibitor, Phosphatidylinositol 4,5 Bisphosphate 3 Kinase Catalytic Subunit Alpha Isoform Inhibitor; Serine/Threonine Protein Kinase mTOR Inhibitor, and Osteopontin Inhibitor.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Combined treatment for primary central nervous system lymphoma	WO2020/109625	Pending	EP USA
NGR conjugates and uses thereof	WO2021/186071	Pending	

Peptide-based diagnostic tool for avb6 avb8-positive cancer imaging

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Sector
Molecular Diagnostics

Key Publication
Nardelli et al. Chem Comm 2019;

Development stage
Preclinical Development

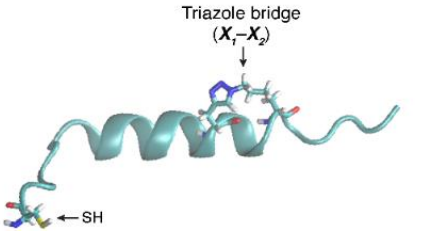
Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

DIAGNOSTIC NEED

Integrins αvβ6 and αvβ8 are upregulated in many cancers, including pancreatic adenocarcinomas, oral mucosal and bladder cancers and melanomas, thus representing potential targets for diagnostic/theranostic purposes.

PRODUCT & TECHNOLOGY

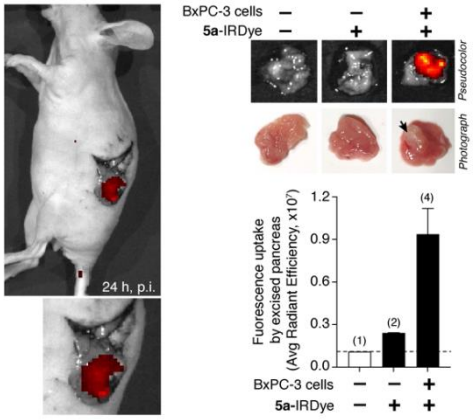
Peptide **5a**: CFETLRGDLRILSILRX_1QNLX_2KELQD$_{CONH_2}$



We have developed a 25-mer peptide, derived from the 39-63 region of human chromogranin A, which contains an RGDL motif followed by an alpha-helix chemically stapled with a bridge. Peptide 5a represents the strongest bi-selective ligand for αvβ6/αvβ8 described to date (K_d: αvβ6: 0.63 ± 0.13 nM, K_d αvβ8: 3.19 ± 1.20 nM). NMR and computational/biochemical studies showed that peptide 5a binds the RGD binding site of αvβ6/αvβ8 with receptor–ligand interactions similar to those observed for the pro-TGFβ1/αvβ6 complex. Peptide 5a can be efficiently coupled via maleimide chemistry to the thiol group of the unique N-terminal cysteine to various compounds, including imaging tracers, nanoparticles, and proteins, without losing affinity for its targets.

PRECLINICAL RESULTS

Peptide 5a was labeled with optical- and radio-imaging compounds currently used in the clinical setting, such as IRDye® 800CW, a near-infrared (NIR) fluorescent dye, and with ^{18}F-NOTA, a tracer for positron emission tomography (PET). *In vivo* dynamic and static optical NIR and PET/CT imaging studies, performed in mice with subcutaneous and orthotopic αvβ6-positive carcinomas of the pancreas, showed high target-specific uptake of the fluorescence- and radio-labeled peptide by tumors (Fig. 1 A and B, below). Significant target-specific uptake of the fluorescence-labeled peptide was also observed in mice bearing subcutaneous αvβ8-positive prostate tumors (unpublished preliminary data). These results indicate that peptide 5a specifically targets to αvβ6- and/or αvβ8-positive tumors and can be exploited for imaging αvβ6 and/or αvβ8 positive tumors.



*Representative fluorescence image of the exposed pancreas of mice bearing BxPC3 orthotopic pancreatic adenocarcinoma injected with **5a**-IRDye.*

COMPETITIVE ADVANTAGES

- Human-derived peptide, lower risk of immunogenicity compared to peptides containing viral sequences.
- Binding affinity for αvβ6 and αvβ8 in the range of sub-low nM.
- Suitable for surgeon platforms in the context of image-guided surgery.
- Versatile ligand for diagnostic purposes: fluorescent dyes and radiotracers.
- Delivery of imaging or therapeutic agents to αvβ6/αvβ8 single- or double-positive tumors.

MARKET & COMPETITION

The global surgical imaging market is projected to reach USD 2.4 billion by 2025 from an estimated USD 1.8 billion in 2020, at a CAGR of 6.3% during the forecast period*. The major factors driving the growth of this market include the technological advancements, reimbursement cuts for analog radiography systems, and the increasing demand for minimally invasive procedures

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Chromogranin A-derived peptides and uses thereof	WO2021094608	Pending	PCT contracting states

Prevotella melaninogenica impacts multiple myeloma

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Sector
Therapeutics

Key Publications
Calcinotto, Brevi et al. *Nat Commun* 2018; Bellone et al. *Microbiol Mol Biol Rev* 2020; Brevi et al. *Front Immunol* 2020.

Development stage
Preclinical Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

CLINICAL NEED

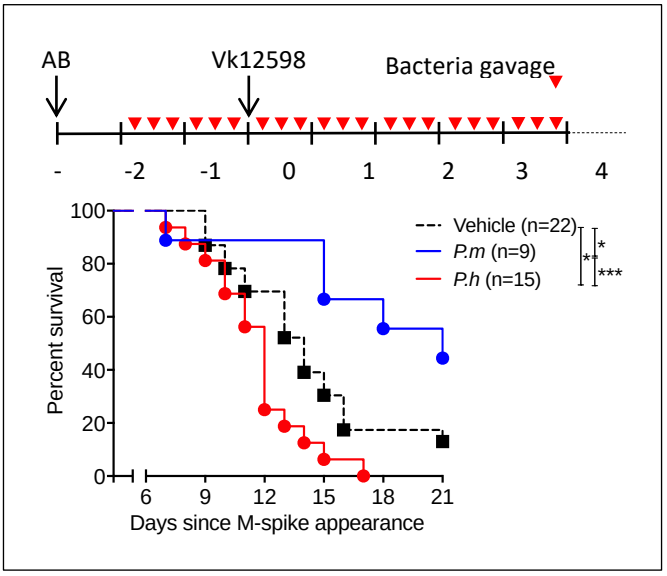
Multiple myeloma (MM) is a treatable, yet incurable plasma cell neoplasia. MM is often preceded by monoclonal gammopathy of undetermined significance (MGUS) or by smoldering MM (SMM). MGUS and SMM are asymptomatic and potentially curable. The following are unmet clinical needs for MM:

- MM has no cure.
- MM is preceded by MGUS or SMM.
- Probability of progression to MM in the first five years:
- MGUS: 1%/year
- SMM: 10%/year
- No validated pathological or molecular prognostic features.
- No preventive treatment available.

PRODUCT & TECHNOLOGY

Prevotella melaninogenica is a species of human commensal bacteria that can be administered orally as gut microbiota modulators (i.e. probiotic). *P. melaninogenica* has a proven utility as an agent capable of redirecting the local and systemic immune response toward a non-inflammatory type.

PRECLINICAL RESULTS



In a primary mouse model of MM, we showed that modulation of the gut microbiota by oral administration of the human commmensal *P. melaninogenica* delayed disease progression and associated with a reduced representation of Th17 lymphocytes both in the gut and in the bone marrow. Conversely, treatment with *P. heparinolytica* induced expansion of gut-born Th17 cells that migrated to the bone marrow and eventually propelled MM progression. Mechanistically, dendritic cells enriched from the bone marrow of mice treated with *P. heparinolytca* produced more pro-Th17 cytokines than dendritic cells from mice gavaged with *P. melaninogenica*. Similar results were obtained when human monocyte-derived dendritc cells were challenged with the *Prevotella* strains. The picture shows treatment schedule and overall survival (Kaplan-Meier plot) of t-Vk*MYC MM gavaged with vehicle (Vehicle), *P. heparinolytica* (P.h) or *P melaninogenica* (P.m). (Calcinotto, Brevi et al. Nat Commun 2018). In ongoing experiments, treatment with *P. melaninogenica* increased the therapeutic index of anti-PD-L1 antibodies and prevented appearance of symptomatic MM.

MARKET & COMPETITION

The MM market is expected to grow from $14.5bn in 2017 to $27.8bn by 2027 across the eight major markets at a compound annual growth rate (CAGR) of 6.7%*.
Regarding competition, MGUS and SMM patients aren't currently subject to any treatment since they are under an observational state. Considering IL-17 as key player involved in the progression of MM from MGUS/SMM, it is currently under development CJM-112 (Novartis AG) which acts by inhibiting interleukin-17, a cytokine that controls cells and activates inflammation. IL-17 is a key product of Th17 cells.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Bacterial strains for medical use	WO2020109620 EP3886881	Pending	EP US

(*) GlobalData(2019),EpiCastReport: Multiple Myeloma Epidemiology

The anti-tumoral potential of a saporin-based uPAR-targeting chimera

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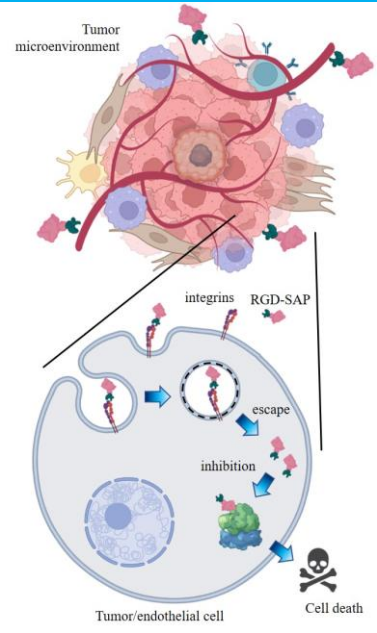
Key Publications
S. Zuppone et al. Frontiers in Oncology, 2022

Development stage
Preclinical Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

The Technology



CLINICAL NEED

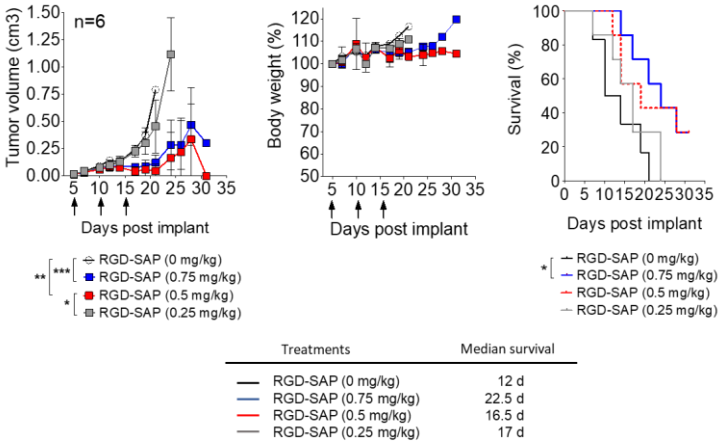
Current clinical protocols for the treatment of tumors are mostly based on surgical debulking, followed by radiation and chemotherapy. These types of therapies suffer from a lack of specificity, killing cells in a cell cycle dependent manner and causing an increased toxicity. Therapies based on toxins have gained great attention in the last decades, as they opened up the possibility to specifically deliver drugs to, and kill, cancer cells without, or minimally, affecting healthy organs

PRODUCT & TECHNOLOGY

Our technology is based on a chimeric recombinant protein formed by a tumor-targeting RGD sequence that enables specific and selective for αv-integrins, and a ribosome inactivating protein saporin (SAP) with a potent and efficient cytotoxic effect and an unusual resistance to high temperature, denaturation and proteolysis. The resulting protein (called RGD-SAP) can kill cells expressing αv-integrins. The αvb3, αvb5 and αvb6 significantly over-expressed in tumor context in a stage and grade-dependent manner lend support to the hypothesis that this class of receptors may represent important molecular targets for toxin delivery to cancer. Our recombinant protein can be easily produced in E.coli.

PRECLINICAL RESULTS

The results of in vivo studies of bladder cancer in two different mouse models show that RGD-SAP can reduce tumor growth and significantly prolong animal survival without inducing detectable side effects. The inventors used a subcutaneous cancer model to determine the optimal dosage and found that RGD-SAP can inhibit tumor growth in a dose-dependent manner.



Treatments	Median survival
RGD-SAP (0 mg/kg)	12 d
RGD-SAP (0.75 mg/kg)	22.5 d
RGD-SAP (0.5 mg/kg)	16.5 d
RGD-SAP (0.25 mg/kg)	17 d

Due to its potential effects on tumor cells and microenvironment, RGD-SAP may represent a good therapeutic tool for cancer. In addition, by inhibiting protein synthesis, SAP acts in a cell cycle independent manner, thus targeting both quiescent and rapidly dividing tumor cells. This feature makes it suitable to contrast both aggressively growing cancers and tumors with slower progression. RGD-SAP can also be employed in combination with other therapeutic options based on different mechanisms of action, e.g. inhibition of DNA synthesis, cell division, and signal transduction. Dose dependent effects of RGD-SAP on tumor growth in a subcutaneous syngeneic bladder cancer mouse model are shown in the following figures.

MARKET & COMPETITION

The production of recombinant proteins has gained great interest in the biopharmaceutical sector as an increasing amount of protein drugs are currently undergoing preclinical and clinical studies or have already been approved to be on the market. The global recombinant proteins market is projected to reach USD 1.7 billion by 2026, at a CAGR of 9.8% during the forecast period.(*)

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Fusion proteins and uses thereof	Not yet published	Pending	IT

Inhibition of glycosylation to potentiate CAR-T cell therapy

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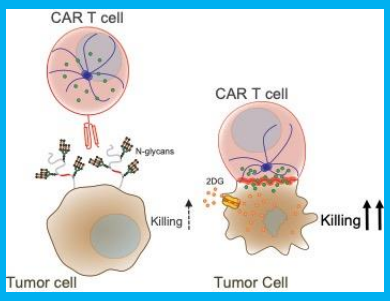
Sector
CAR-T Therapeutics

Key Publication
Greco et al., Sci. Transl. Med. 14, eabg3072 (2022)

Development stage
Preclinical Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

The Technology



CLINICAL NEED

Chimeric antigen receptors (CARs) are artificial molecules combining antigen-binding and T-cell activating functions into a single receptor, to provide T cells with the ability to target a desired antigen. Most commonly, the antigen-binding moiety is represented by the single-chain fragment variable of a monoclonal antibody. T cells engineered to express a tumor-specific CAR (CAR T cells) represent a great promise in the context of cancer immunotherapy. Different institutions have recently reported impressive clinical responses by infusing CD19 CAR T cells in patients with refractory B-cell malignancies. However, first-in-man studies revealed unique hurdles contributing to the lack of a sharp demonstration of efficacy of CAR T cells in the context of solid malignancies. The first requirement for CAR T cells to work properly is efficient antigen engagement, which leads to the formation of an immunological synapse able to activate effector functions and drive tumor cell elimination. Glycosylation is the enzymatic process linking sugars to other sugars, proteins or lipids. It has been shown that glycosylation alterations are very frequent in cancer, where they promote tumor growth and metastasis. These changes comprise increased branching of N-glycans that form huge sugar structures on the surface of malignant cells. Interestingly, it has been reported that these glycans can mask peptidic epitopes from antibody recognition and are required for the proper interaction of co-inhibitory ligand/receptor pairs that drive T-cell exhaustion.

PRODUCT & TECHNOLOGY

We have shown that an excess of branched N-glycans is present on multiple carcinomas and that these sugar structures inhibit the killing of malignant cells by CAR T cells. The invention regards the exploitation of glycosylation inhibition to improve the efficacy of CAR T cells in solid tumors. In this context, we report that tumor treatment with 2-Deoxy-D-glucose (2DG), a de-glycosylating agent, causes membrane exposure of de-glycosylated antigens, sensitizing tumor cells to be recognized and killed by CAR T cells. Moreover, we have shown that glycosylation inhibition ameliorates the exhaustion profile of CAR T cells and preserve their fitness in vivo. The efficacy of combining CAR T cell therapy with 2DG administration has been demonstrated with different CAR specificities and multiple types of solid malignancies.

SOLUTION & BENEFITS

2-DG improves CAR T-cell efficacy through several mechanisms: (i) unmasks tumor antigens; (ii) potentiates immunological synapse; (iii) reduces inhibitory receptors' engagement; (iv) improves CAR T cells' fitness, (v) can kill hypoxic tumors, (vi) exerts a beneficial effect on the tumor microenvironment (TME). 2-DG is safe due to its preferential accumulation in tumor cells. It has been extensively used in clinical trials (>200 patients), where it showed good tolerability with only minimal and reversible side effects (hypoglycemic symptoms).

MARKET & COMPETITION

The global market regarding CAR T-cell therapy applied to only solid tumors is made up of 6.227 past and ongoing deals for an overall value of billions of dollars. There are many competitors on the market considering all combinatorial treatments with the aim to potentiate CAR T cells efficacy, such as small molecules, gene therapy, and monoclonal antibodies. However, to the best of our knowledge, this project is the only inhibiting N-glycosylation to improve CAR-T efficacy against cancers. The added value of this technology is that it can be applied to multiple tumors and CARs, does not require new CAR design, and allows overcoming several barriers at once as such defective antigen engagement and tumor cell killing, T-cell exhaustion, T-cell differentiation, Immunosuppressive TME.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Combination of a glycosylation inhibitor with one CAR cell therapy for treating cancer	WO2020020841	Pending	Europe, USA, Canada, China, Japan, Australia and Israel

Adult skeletal muscle progenitors for the therapy of Duchenne Muscular Distrophy

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Sector
Cell Therapy

Key Publication
Cossu et al., EMBO Mol. Med. 2015

Development stage
Phase I/II

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

CLINICAL NEED

Several genetic diseases affect mesoderm tissues such as skeletal and smooth muscle, cartilage, bone, joints, and the wall of blood vessels. Many of them are rare, not well studied, and are devastating conditions that dramatically affect numerous organs, and compromise life quality and expectancy. All lack an efficacious therapy.
Among genetic diseases affecting skeletal muscle, Duchenne Muscular Dystrophy (DMD) is the most common, affecting 1/4,000 newborn males. It is caused by mutations in the dystrophin gene, located on X chromosome, and characterized by progressive deterioration of skeletal and cardiac muscle. This quickly leads to a variable but progressive limitation of the patient's mobility, including confinement to a wheelchair and heart and/or respiratory failure typically by 30-40 years of age. Despite numerous attempts with oligonucleotides aimed at correcting the mutated mRNA and AAV delivering micro-dystrophins (shorter and partially functioning proteins), DMD is still incurable and does not yet have a therapy with long lasting/curative benefits

PRODUCT & TECHNOLOGY

The current invention aims at developing a range of treatments for DMD patients with various genotype backgrounds by combining cell therapy with exon skipping to exploit the advantages of both and compensate their respective drawbacks. We have compelling in vitro and in vivo evidence of dystrophin production above 30% of healthy muscle, considered a therapeutic level. While the commercial potential of cell therapies has always been limited due to the risk of immune rejection, the current invention overcomes this limitation by using engineered multipotent cells, called mesoangioblasts (immune privileged universal donor cells), which were developed as an allogenic product for DMD patients affected with the same genetic mutation.
With this approach, we aim to create GMP grade, off the shelf, immune privileged, universal donor mesoangioblasts, vessel associated myogenic progenitors. Thanks to a newly discovered method to indefinitely expand human mesoangioblasts without immortalizing agents, these will be genome-edited to delete HLA and create a bank of cells available to be engineered to treat many monogenic diseases affecting the mesoderm, thus cutting cost. As proof of principle, cells will be engineered to treat Duchenne and Congenital muscular dystrophies, and a similar approach has been envisioned for merosin-deficient congenital muscular dystrophy type-1A (MDC1A) and Limb Girdle Muscular Dystrophy 2C (LGMD2C) for which pre-clinical models have already been validated.

SOLUTION & BENEFITS

This is the first characterization of a human cell population that fulfils all the criteria of a successful cell therapy protocol in Duchenne Muscular Dystrophy and has demonstrated several clear advantages over the current standard of care:

- Efficient engraftment of treated cells in muscles
- Amplification of treatment along the multinucleated muscle fibres: each engrafted nucleus produces a small nuclear RNA that diffuses to and induces exon skipping also in neighboring nuclei
- Development of a single cell therapy product for the treatment of whole populations with the same genetic defect (unlike autologous approaches)
- Pre-clinical evidence of a long lasting effect
- Previous experience of phase I/IIa clinical trials and ongoing proof of concept trial.

The nature of the approach is flexible and modular in addressing gene disruption in many mesoderm genetic diseases. It may be extended to other muscular dystrophies and to rare diseases of the connective tissue that are due to mutations in genes encoding for proteins of the extra-cellular matrix.

MARKET & COMPETITION

Currently, of the 16 products marketed for muscular dystrophies, all are small molecules and none are classified as gene or cell therapy. Sarepta Therapeutics leads the way in DMD with several gene therapies being developed. The forecasted global sales is expected to grow from USD 117M in 2023 to USD 4.1B in 2028.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Skeletal muscle periangioblasts and cardiac mesoangioblasts, method for isolation and uses thereof	WO2007093412	Active	EP US

Use of neural stem cells to induce neuro protection in inflammatory CNS disorders

I.R.C.C.S. Ospedale San Raffaele
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Sector
Cell Therapy

Key Publications
Pluchino et al., Nature 2005; Martino G, Pluchino S. Nat Rev Neurosci. 2006

Development stage
Phase I

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

CLINICAL NEED

Transplantation of neural stem precursor cells in patients affected by central nervous system (CNS) disorders characterized by chronic inflammation (e.g., multiple sclerosis, brain tumors, ischemic stroke) has limited therapeutic impact due to recurrent or persisting inflammation that targets and kills both CNS-resident and transplanted cells. Consequently, there is a strong clinical need for immunomodulatory therapies that avoid inflammatory self-reactivity and boost neuronal protection.

PRODUCT & TECHNOLOGY

We have developed a novel immunomodulatory mechanism that boosts such limited therapeutic effect by transplanting undifferentiated adult neural stem/progenitor cells (aNPC), which promote direct neural cells replacement by acquiring *in vivo* terminally differentiated phenotype. As a proof of concept, subventricular zone (SVZ)-derived syngenic adult NPCs (aNPC) were transplanted in a mouse model of chronic-recurrent autoimmune CNS inflammation, namely relapsing-remitting experimental autoimmune encephalomyelitis (R-EAE). While assessing their therapeutic potential, they demonstrated that during R-EAE inflamed CNS, perivascular areas function as ideal, although atypical, niche-like microenvironments where transplanted cells can survive for long-term (up to 3 months post-transplantation) as bona fide aNPCs. Furthermore, upon systemic injection, aNPC can exert a neuroprotective effect by inducing *in situ* programmed cell death of blood-borne CNS-infiltrating pro-inflammatory Th1, without affecting anti- inflammatory Th2 cells in the inflamed CNS perivascular area. Thus, the CNS inflammatory microenvironment dictates aNPCs cell fate, and therefore their therapeutic efficacy is as follows:

- when neurodegeneration prevails, transplanted aNPCs acquire a mature phenotype and thus replace damaged neural cells, while
- when neuroinflammation predominates, transplanted aNPCs survive to recurrent inflammatory episodes by retaining both an undifferentiated phenotype and notable proliferating capacities.

SOLUTION & BENEFITS

Undifferentiated aNPCs have relevant therapeutic potential in chronic inflammatory CNS disorders because they display immune-like functions that promote long-lasting neuroprotection in inflamed CNS perivascular area on the one hand, and brain repair on the other. Among their competitive advantages, they have:

- aNPC-mediated apoptosis of blood-borne CNS-infiltrating encephalitogenic T cells, promoting long-lasting neuroprotection in chronic inflammatory CNS disorders;
- Selective accumulation of intravenously-injected aNPCs within CNS inflamed areas using constitutively functional homing molecules (e.g., a4 integrins and GPCRs) canonically used by pathogenic CNS- infiltrating blood-borne lympho- and monocytes;
- Preferential maintenance of an undifferentiated phenotype upon aNPCs transplantation, thus potentially escaping the chronic CNS-reactive autoimmunity;
- In vivo maintenance of their proliferation capacity for up to 100 days after aNPCs transplantation, thus potentially modulating their fate in vivo (proliferation vs quiescence vs migration and differentiation) in response to specific environmental signals (e.g., cytokines, chemokines, stem cell regulators).

MARKET & COMPETITION

The global market for CNS disorders in general is quite large, with 2,393 past and present deals for a total of over USD 70B in sales for 2021, and a forecasted market over USD 170B (representing a 14% CAGR). Regarding the subset of Multiple Sclerosis, a narrower market of around 300 past and ongoing deals with sales of around USD 21B for 2021 and an estimated USD 28B in 2028 (representing around 4% CAGR). In both cases, the vast majority of marketed products are small molecules, making this strategy and attractive alternative to currently available therapies.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Inflammation	WO2007015173	Active	EP JP

IL2-based gene therapy for HBV treatment

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Key Publication
A.P. Bénéchet et al. Nature 2019

Development stage
Lead Optimization

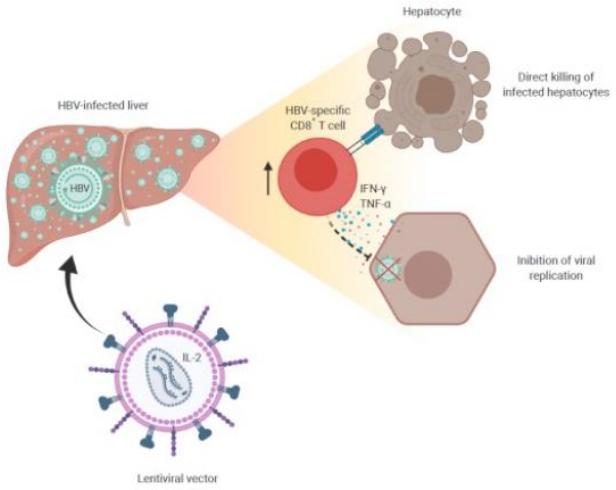
Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

CLINICAL NEED

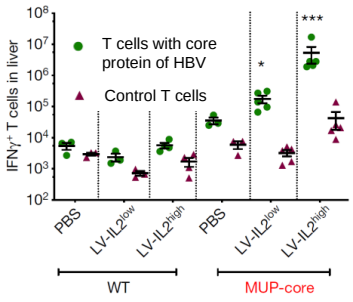
Hepatitis B virus (HBV) infection remain a major public health issue worldwide. Current treatment for HBV mainly relies on direct acting antiviral drugs, which suppress virus production, but do not eradicate HBV from the liver. The World Health Organization estimates that in 2015, 257 million people were chronically infected with the HBV virus globally (World Health Organization, 2020). Accordingly, this leads to a requirement for lifelong treatment.
In general, CD8+ T cells have a critical role in eliminating intracellular pathogens. The liver, for its specific features, is thought to be biased towards inducing a state of T cell unresponsiveness or dysfunction. This phenomenon underpins the unresponsiveness toward antigens specifically expressed in hepatocytes, and the propensity of some hepatotropic viruses, such as HBV, to establish persistent infections.

PRODUCT & TECHNOLOGY

We have found that administration of interleukin-2 (IL-2) enables reinvigoration and restoration of effector responses in dysfunctional CD8+ T cells, such as against antigens specifically expressed in hepatocytes. Moreover, the studies have revealed that local administration of IL-2 to the liver is able to increase the effector responses while avoiding the toxicity that is associated with systemic administration of IL-2.



PRECLINICAL RESULTS



To test the clinical potential of IL-2 in a system that would limit its systemic toxicity, we generated proprietary third-generation, self-inactivating lentiviral vectors (LVs) that selective hepatocellular expression of IL-2. We observed that the targeted delivery of IL-2 to the liver promotes the differentiation of HBV-specific dysfunctional CD8+ T cells into effector, IFNγ-producing cells endowed with antiviral potential. (Figure, left).

MARKET & COMPETITION

Chronic Hepatitis B is still considered a great global health burden even though there is an effective vaccine. A definitive treatment for HBV eradication is not yet available on the market. Conventional treatments are based on antiretroviral such as tenofovir, lamivudine, adefovir, entecavir or telbivudine. In this regard, the prevalent view in the scientific community is that, similarly to HIV infection, combination therapy (a “cocktail” of drugs) will be required to cure chronic HBV infection. The global chronic hepatitis B therapeutics market is expected to reach a value of $3 billion by 2024.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Agents and methods for treating viral infections	WO2020239964	Pending	EP

Gene therapy for Rett syndrome

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Key Publication
Luoni et al., eLife, 2020

Sector
Gene Therapy

Development stage
Preclinical Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights

The Technology
MECP2 minigene that expresses physiological levels of MECP2 for the treatment of Rett syndrome

CLINICAL NEED

Rett syndrome (RTT) is a severe neurological disorder and second cause of intellectual disabilities in girls. RTT is distinguished by 6-12 months of overtly normal development followed, then, by a rapid regression with the loss of purposeful motor skills and the onset of repetitive and autistic behaviors. Prevalence in the general population is 1:30.000. In the vast majority of cases, RTT is caused by loss-of-function mutations in the MECP2 gene, which encodes for the methyl-CpG binding protein 2 (MeCP2), a global chromatin regulator highly expressed in neurons. MeCP2 is a global determinant of the neural chromatin structure and is a pervasive regulator of gene expression in brain cells and, thus, it remains challenging to target a single MeCP2 downstream pathway to obtain a substantial therapeutic benefit. RTT affected girls, indeed, are a mosaic of healthy and affected cells depending whether the MeCP2 wild-type or mutant gene copy is expressed, respectively. Therefore, it is important to silence the endogenous gene in order to homogenize the total expression levels of meCP2 driven by the viral transgene.
The inherent monogenic nature of RTT makes gene therapy a strong translational option for this disease; however, MECP2 gene duplication in humans is responsible for a serious and clinically distinguished neurodevelopmental disorder. Thus, a successful gene therapy for RTT must deliver the correct MeCP2 dosage in a tight range that overlaps with endogenous levels.

PRODUCT & TECHNOLOGY

The technology is a transgene cassette with the Mecp2_e1 isoform including the coding sequence and a short proximal 3'UTR (3'pUTR, ~200bp). This Mecp2 transcript occurs naturally in embryonic stem cells and various tissues, but during development of the neural system this form is progressively overcome by transcripts with longer 3'-UTR (8,6 Kb). Given the limited efficiency in transcript stability and translation efficacy, only the use of the CBA strong promoter sustains Mecp2 protein levels comparable to those found in WT neurons. As all the viral cassettes caused a similar instability of Mecp2 transcripts, the CBA-iMecp2-pUTR-pA (M2a) construct was chosen for further in vivo studies, and referred to it as iMecp2, for instability-prone Mecp2.

PRECLINICAL RESULTS:

An optimized gene therapy MeCP2 viral vector ensures physiological levels of Mecp2 in neurons and glial cells;

- Mecp2 gene transfer is beneficial even when applied to Mecp2 symptomatic mutant mice;
- Mecp2 gene transfer is safe and well tolerated in wild-type animals;
- Mecp2 gene therapy in patients can be delivered by multiple intracerebral injections;
- M2a cassette sustains physiological levels of MeCP2 expression in neurons combining a strong promoter with an enhanced instability of the transgene RNA.

MARKET & COMPETITION

Global Rett syndrome market is growing with a CAGR of 75.1% in the forecasted period of 2021 to 2028 and is expected to reach USD 653K by 2028 from USD 446K in 2020*. Regarding competition, there are currently several small molecule drugs and gene therapies being tested, all of which have several downside effects:
Pharmacological approaches that are meant to treat some secondary symptoms have failed to provide any significant effect on the severe neurological manifestations of the syndrome (autisms, lack of speech, and minimal voluntary movements).
Most of the pharmacological approaches have severe side effects up to now (nausea, vomit, psychiatric changes).
AVXS-201 gene therapy is based on AAV9 lumbar intrathecal delivery. However, multiple reports have shown that AAV9 has a very poor capacity to cross the ependymal layer and therefore the viral transduction of the brain remains very low.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Gene Therapy for Rett Syndrome	WO2020212448 US 20220133912	Pending	EP USA

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Sector
Gene Therapy

Development stage
Preclinical

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

The Technology
AAV vector with enhanced brain penetration and transduction

Identification of new Adeno-Associated Virus 9 (AAV9) capsid variants targeting the BALB/c mouse brain through directed evolution

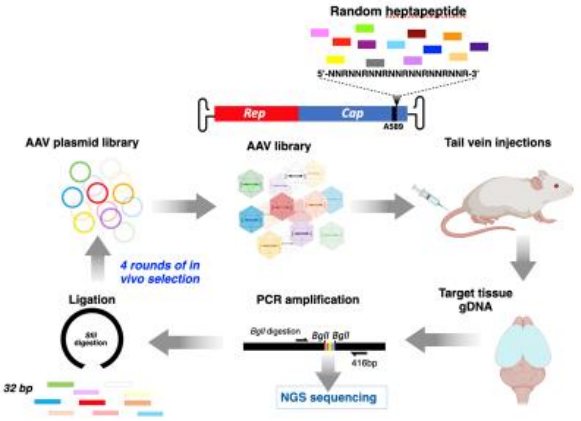
I.R.C.C.S. Ospedale San Raffaele
Gruppo San Donato

CLINICAL NEED

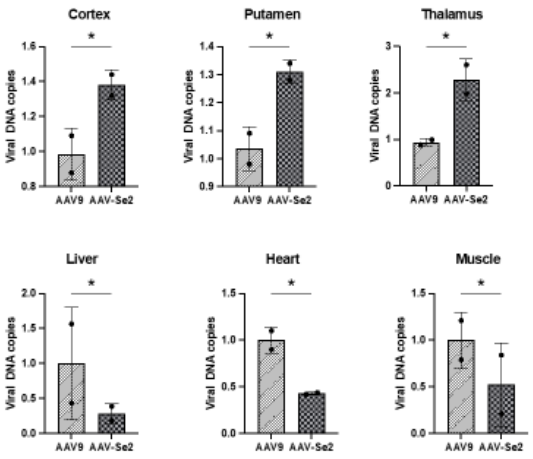
In order to achieve optimal delivery of the target tissue and avoid possible detrimental effects, gene therapy approaches require vectors endowed with both high transduction efficiency and specificity. Moreover, systemic delivery of the gene therapy formulation is preferable and more effective, but diffusion into the central nervous system (CNS) is limited by the blood-brain barrier (BBB) that restricts the passage of molecules, including AAV capsids. Since cerebral endothelial cells play a key role in BBB integrity and function, a recombinant AAV specifically targeting these cells could be helpful for the development of new therapeutic approaches for various CNS diseases. Accordingly, there is a significant need for vectors that not only target brain endothelial cells, but also cross them, thus reaching the cerebral parenchyma and infecting neurons and glia cells. The clinical relevance of such a tool would be invaluable and could bear innumerable applications in CNS pathologies.

PRODUCT & TECHNOLOGY

Starting from these premises, our researchers executed an in vivo screening approach using an AAV9 display peptide library to select novel brain targeting capsid variants. The peptide library was inserted in the AAV9 capsid with additional W503A mutation which is known to erase the AAV9 natural biding on galactose thus facilitating new interactions. In order to avoid Ly6a binding the selection was carried out using BALB/c mice that present mutations in the Ly6a locus that significantly decrement its expression. Three enriched capsid variants were identified after four consecutive rounds of selection. All of them displayed prevalent endothelial cell transduction that we identified and characterized on transduced mouse brains after tail vein injections. Brain cell transduction and neuronal targeting were also observed with interesting differences between brain regions with two capsid variants and equally conserved in BALB/c mice.



SOLUTION & BENEFITS



These targeted modifications in AAV9 capsids result in enhanced brain targeting and transduction capability. In particular, our researchers generated AAV9 capsid libraries and tested them for in vivo screening for new variants with enhanced transduction ability through consecutive rounds of selection for the desired tissue. They have discovered modifications of capsid proteins that enhance targeting to cells of the brain or central nervous system (e.g. increase the ability to cross the BBB), increase the transduction efficiency of these cells, and decrease tropism for non-brain tissue, when compared to AAV9. Moreover, the resulting capsid proteins have been validated in both mice and marmosets.

WHAT WE ARE LOOKING FOR

Industrial partners with experience in AAV technology interested in out-licensing this technology for commercialization and potentially further development with option rights.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Gene Therapy	Not yet available	Pending	Priority

HMGB1-based approach for Duchenne Muscular Dystrophy

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Key Publications

Venereau et al. J Exp Med. 2012;
Tirone et al. J Exp Med. 2018;
Careccia et al. Sci Transl Med. 2021.

Development stage
Preclinical Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

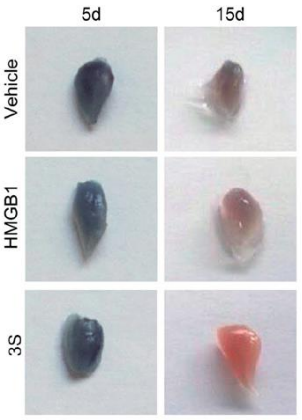
CLINICAL NEED

Duchenne Muscular Dystrophy (DMD) symptom onset is in early childhood, usually between ages 2 and 3. The disease primarily affects boys, but in rare cases it can affect girls. In Europe and North America, the prevalence is 6 per 100,000 individuals, incidence 1 per 3500/5000 live male birth. Limb-Girdle Muscular Dystrophy (LGMD) affects males and females in equal numbers; its prevalence is unknown, but estimates range from one in 14,500 to one in 123,000. The age of onset can vary greatly even among individuals of the same family. In all cases, muscular dystrophy is characterized by progressive muscle wasting associated to chronic local inflammation and oxidative stress. High Mobility Group Box 1 (HMGB1) is a nuclear protein that signals tissue damage when released into the extracellular medium We demonstrated that the oxidation of HMGB1 cysteines switches its extracellular activities from the orchestration of tissue regeneration to the exacerbation of inflammation. Specifically, oxidized HMGB1 acts as a proinflammatory mediator by interacting with Toll-Like Receptor 4 (TLR4) and the Receptor for Advanced Glycation Endproducts (RAGE) while reduced HMGB1 supports tissue regeneration through CXCR4 by acting on both the stem cells and their microenvironment. Currently, there is no cure for DMD and a very limited efficacy of current treatments (Steroids, ASO for Exon skipping, small molecule for read-through premature nonsense stop codon).

PRODUCT & TECHNOLOGY

We designed a non-oxidizable form of HMGB1 where all cysteines are replaced by serines, called 3S, to create a regenerative and non-inflammatory form of HMGB1. Following acute injury, treatment with 3S promotes muscle regeneration without exacerbating inflammation, by inducing the expansion of satellite cells and by mobilizing tissue-healing macrophages. Similarly, 3S administration accelerates liver regeneration after drug intoxication, bone healing after fracture and hematopoietic recovery following chemotherapy. Hence, the common regeneration responses to 3S indicate that the HMGB1/CXCR4 axis may be involved in the repair and regeneration of most tissues.

PRECLINICAL RESULTS:



Extracellular HMGB1 is present at high levels and undergoes oxidation in dystrophic patients and in mouse models of Duchenne Muscular Dystrophy (DMD) and Limb-Girdle Muscular Dystrophy type 2d (LGMD2D). Pharmacological treatment with the 3S variant improves functional performance, muscle regeneration and satellite cell engraftment in dystrophic mice, while reducing inflammation and fibrosis. Overall, the balance between HMGB1 redox isoforms dictates whether skeletal muscle is in an inflamed or regenerating state, and the non-oxidizable form of HMGB1 is a promising therapeutic approach to counteract the progression of the dystrophic phenotype and to potentiate combined treatments. **KEY FINDINGS:**

No genetic mutation restriction: preclinical data demonstrates the therapeutic properties of this drug candidate in different mouse models of muscular dystrophies; Drug with potent regenerative properties in multiple muscles and other tissues: treatment with this drug in preclinical models has shown potent regenerative properties in multiple tissues/organs (skeletal muscles, bone, liver); Synergic/complementary effects of the drug with current/future treatments: treatment with the 3S-HMGB1 promotes stem cell engraftment and expansion in dystrophic mice; High efficiency and ease of administration: a single systemic administration of this molecule per week for only 3 weeks is sufficient to observe functional improvement in muscles of dystrophic mice; No apparent toxicity: both in acute (a single administration) and chronic treatments (6 weeks treatment).

MARKET & COMPETITIVE CONTEXT

In 2020, the global Duchenne Muscular Dystrophy market size was USD 935 million, and it is expected to reach USD 9904.4 million by the end of 2027, with a CAGR of 42.1% during 2021-2027.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
HMGB1 variants and uses thereof	WO2014016417 US10626153	Granted	EP US

Silencing inhibition of Asparagine Synthase (ASNS) for the treatment of Polycystic Kidney Disease

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Sector
Gene Therapy

Key Publication
Podrini et al., Communication Bio 2018, 1:194

Development stage
Preclinical

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

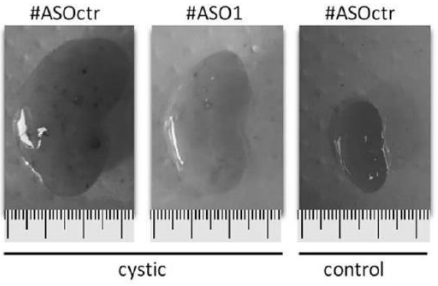
CLINICAL NEED

Autosomal Dominant Polycystic Kidney Disease (ADPKD) is a chronic progressive disease, characterized by bilateral renal cyst formation, that is the most common monogenic disorder affecting the kidney, with an estimated prevalence of individuals affected at birth of 1:500 to 1:1000. ADPKD Cysts grow progressively over time, and the gradual expansion of cysts compresses and eventually replaces the normal tissue, causing end-stage renal disease in most affected individuals. The disease is due to loss-of function mutations in two genes: PKD1, which is mutated in 85% of cases, and PKD2, in the remaining 15%. The crucial goal of correcting the gene defect still seems far away. The ideal therapeutic compound in ADPKD should effectively block both abnormal cysts growth and fluid secretion into the cysts. Thus, therapeutic interventions targeting cyst expansion are currently being tested in multiple clinical trials to delay renal disease progression. Among these opportunities for therapy, the use of a vasopressin receptor antagonist, Tolvaptan, is, as of today, the only one that results in an effective amelioration of disease progression in patients and the only molecule that has been accepted as a therapy for ADPKD (in Japan, Canada and U.K.)

PRODUCT & TECHNOLOGY

We have carried out a comprehensive metabolomics characterization of cells and renal tissues from a mouse model carrying the kidney-specific inactivation of the Pkd1 gene and discovered that glutamine metabolism is interlinked with asparagine synthesis in ADPKD, and that the Asparagine Synthase (ASNS) gene is an essential component of the process. ASNS is a transaminase that converts aspartate into asparagine while deaminating glutamine to form glutamate. We have found that silencing ASNS with anti-sense oligonucleotides impacts the growth of *Pkd1* cells and is more prominent when cells are also deprived of glucose. Overall, this work demonstrated that the reduction of ASNS levels is associated to a decrease of the expansion of renal cysts in ADPKD. Accordingly, interfering with both glucose and glutamine uptake (via ASNS) in PKD seems to be an ideal combinatorial strategy.

SOLUTION & BENEFITS



The current invention represents a new druggable target for the treatment of ADPKD. The major limit of Tolvaptan, the current standard of care for ADPKD, is that it causes adverse effects like increased thirst and aquaretic effects affecting patient's quality of life. The therapy in ADPKD might be required for years and so it is essential that the side effects are limited. ASNS inhibition causes apoptosis specifically in mutant cells in comparison to wild-type cells therefore it should induce apoptosis only in cells which had acquired a second mutation in Pkd1 gene, and which give rise to cysts. Therefore, ASNS inhibition slows down cyst expansion and the results show that treatment with the ASNS ASOs results in a complete absence of side effects in the animals, in line with the normally very low expression levels of this enzyme in adult tissues. In addition, combination therapy with 2DG would allow targeting multiple pathways altered in ADPKD reducing the dosage of one single drug limiting the side effects.

MARKET & COMPETITION

The global market for PKD is growing with a CAGR of 10.4% for the forecasted period of 2021-2028 and is expected to reach USD 1.78B in 2028 from USD 894M in 2021. With respect to the competition, there is only one approved therapy (the small molecule Tolvaptan), which saw global sales of USD 894M in 2021. Other therapies in preclinical development include: 1) an antisense oligonucleotide targeting miRNA-17, in development by Regulus Therapeutics and under IND/CTA review; 2) a Ceramide Glucosyltransferase inhibitor development by Sanofi, and; 3) a CFTR activator development by Eloxx Pharmaceuticals.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Asparagine synthetase inhibitors and uses thereof	WO2020049069	PCT published	EP

Compounds for use in Polycystic Kidney Disease

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Key Publications
Rowe et al, Nat Med, 2013
Chiaravalli et al, JASN, 2016;
Podrini et al, Comms. Bio., 2018.

Development stage
Preclinical Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

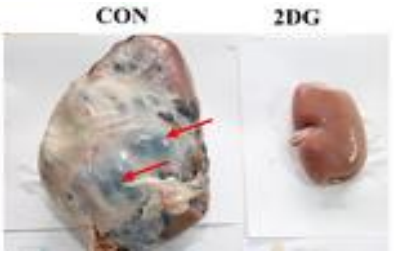
CLINICAL NEED

Autosomal Dominant Polycystic Kidney Disease (ADPKD) is the most frequent rare monogenic disorder affecting approximately 3.5 to 14 million people worldwide. ADPKD is characterized by relentless development and growth of fluid-filled renal cysts, developing from any segment of the renal tubule and eventually causing progressive kidney enlargement. Pyelonephritis and cyst infections usually occur. When the majority of nephrons has been destroyed, renal function declines, typically after the fourth decade of life. Kidney failure requiring renal-replacement therapy (RRT; ie dialysis or kidney transplant) occurs in approximately 50% of patients and typically develops in the fourth to sixth decade of life. Tolvaptan is the currently available therapy able to retard disease progression; it's a vasopressin receptor antagonist that retards disease progression by lowering the levels of cAMP in the distal tubule and collecting duct of the kidney. However, inhibition of the vasopressin receptor is not curative. Disease progression is slowed down and not halted, therefore procrastinating the need of RRT. Among the several limitations related to the use of Tolvaptan, the major are: liver toxicity; potent diuretic effect, thus causing a sensible worsening in the quality of life of the treated patients; not effective in cysts of proximal tubule origin, nor in the cysts of the liver, often associated with ADPKD.

PRODUCT & TECHNOLOGY

The present technology is the use of the glucose analog 2-deoxyglucose (2-DG) in ADPKD. Studies of PKD mouse models have shown that PKD1 inactivation in vivo in the renal tubule results in increased glucose uptake and lactate production. Using a metabolomic approach, we have identified, both in cells from a murine model of PKD and from patients-derived ADPKD kidney, a new pathogenic ADPKD pathway involving defective glucose metabolism. Specifically, we showed that mutations in PKD1 results in enhanced glycolysis. In addition, experimental data indicate that the cells lining the cysts are more prone to uptake 2-DG than other cells and they are less effective in reprogramming their metabolism to use other energy sources

SOLUTION & BENEFITS



Glucose deprivation by 2-DG reduced proliferation and sensitized PKD1 mutant cells to apoptosis, suggesting that interfering with this pathway may be effective in slowing down cyst expansion in ADPKD. Moreover, 2-DG treatment ameliorated the proliferation rate and cystic kidney volume in a PKD mouse model. From a safety standpoint, it is worth noting that 2-DG has been tested in clinical trials in >200 patients as an anticancer drug (alone or in combination with chemotherapy), where 2-DG proved safe in the clinical setting. Among the reported side effects, the most common ones were attributable to hypoglycemic symptoms (somnolence, hunger, sweating) and were rapidly reversible with glucose administration. Dose limiting cardiac toxicity (QTc 3-degree elongation) was observed for chronic administration of the drug at doses higher than 45 mg/Kg, which are higher than the ones expected to be used in ADPKD patients (8 - 30 mg/kg).

MARKET & COMPETITION

A very large number of different signaling pathways were reported to be implicated in the processes underlying ADPKD pathogenesis and shown to be defective in ADPKD cystic epithelia, in particular the cyclic adenosine monophosphate (cAMP) and mammalian target of rapamycin (mTOR) pathways. In recent years, several compounds have been tested in preclinical studies based on original observations of dysfunctional pathways or biological processes identified. Meanwhile, global market regarding PKD therapy is made up of 56.555 past and ongoing deals for an overall value of 9 billion of dollars.

- Average yearly healthcare expenditure for ADPKD patients in Italy varying from €3,913.89 for non-dialyzed patients to € 45,059.62 for dialyzed patients.
- Tolvaptan global sales in 2017 were 604M$, forecasted to increase to 1.892M$ in 2024 (Globaldata).
- Considering the disease is progressive, chronic treatment has to be envisaged.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Compounds for use in polycystic kidney disease	US10478446 WO2014006093	Granted Pending	US EP

DUX4 inhibitor for the treatment of FSHD

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Sector
Therapeutics

Development stage
Preclinical

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights.

The Technology

CLINICAL NEED

Facioscapulohumeral muscular dystrophy (FSHD) is one of the most prevalent neuromuscular disorders and leads to significant lifetime morbidity, with up to 25% of patients requiring a wheelchair. The disease is characterized by rostro-caudal progressive wasting in a specific subset of muscles. Extra-muscular manifestations can occur in severe cases, including retinal vasculopathy, hearing loss, respiratory defects, cardiac involvement, mental retardation, and epilepsy.
FSHD affects 1:7500 individuals of all sexes and ages, with about 870.000 patients estimated worldwide. Due to an unknown molecular mechanism, FSHD displays overlapping manifestations with amyotrophic lateral sclerosis (ALS). FSHD is caused by aberrant expression of the transcription factor double homeobox 4 (DUX4), which is toxic to skeletal muscle leading to disease.
Currently, there is no cure or therapeutic option available to FSHD patients. However, the consensus that ectopic DUX4 expression in skeletal muscle is the root cause of FSHD pathophysiology has opened the possibility of targeted therapies. Importantly, it has been shown that the ability of DUX4 to activate its direct transcriptional targets is required for DUX4-induced muscle toxicity. Accordingly, DUX4 targets account for the majority of gene expression alterations in FSHD skeletal muscle. Thus, blocking the ability of DUX4 to activate its transcriptional targets has strong therapeutic relevance.

PRODUCT & TECHNOLOGY

Matrin 3 (MATR3) is a natural, physiologic, endogenous inhibitor of DUX4. We showed that MATR3 binds directly to the DNA binding domain of DUX4 and blocks its activity. Moreover, MATR3 also indirectly inhibits DUX4 expression. In so doing, MATR3 rescues survival and myogenic differentiation of cellular models of FSHD.

SOLUTION & BENEFITS

MATR3-based approaches work through: (i) blocking of DUX4 activity and (ii) inhibition of DUX4 expression. The MATR3 fragment is safe due to lack of MATR3 functional domains known to be involved with toxicity.
By combining proteomics, expression profiling, cellular, and molecular biology approaches, validated in primary muscle cells obtained by several FSHD patients and healthy donors, we found that MATR3 levels are intimately connected to DUX4 activity. We demonstrated that MATR3 binds directly to the DNA-binding domain of DUX4 and blocks DUX4 expression and activity. Using gene therapy-like approaches, we found that MATR3 delivery rescues the muscle differentiation and cell death phenotypes displayed by FSHD muscle cells. Importantly, we found that a MATR3 region of just 20 amino acids is sufficient to bind to DUX4, opening the possibility to develop a drug-like molecule. Importantly, MATR3 targeting is safe for muscle cells from healthy donors.

MARKET & COMPETITION

The global market, in the last three years, regarding cell and gene therapy applied to FSHD has an overall value of 364 billion dollars. Competition, considering all combinatorial treatments with the aim to inhibit DUX4 activity and ameliorate FSHD symptoms, is mainly focused on small molecules, gene therapy, and antisense oligonucleotides. The added value of the endogenous MATR3 with respect to the previous approaches consists in the ability to block both DUX4 expression and activity with a single inhibitory molecule.

INTELLECTUAL PROPERTY

Patent Title	Publication #	Status	Coverage
Inhibitor of DUX4 and uses thereof	WO2020152367 US20220098252A1	Pending	EP USA

Single-Cell Genome and Epigenome by Transposases sequencing (scGET-seq)

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Key Publication
Tedesco et at., Nature Biotech, 2021

Sector
Genomics

Development stage
Technological Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights

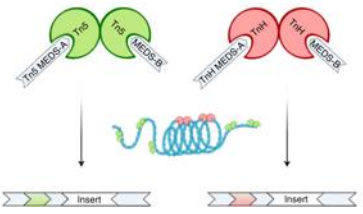
Intellectual Property
International patent application pending

RESEARCH NEED

Cancers are characterized by extensive inter-patient and intra-tumour heterogeneity. This fuels clonal evolution, leading to treatment resistance, the leading cause of death for cancer patients. Increasingly detailed analysis of cancer genomes, before and after treatment, have so far failed to identify genetic causes which could explain the ensuing refractoriness to therapy. Recently, epigenetic changes have emerged as key contributors of drug resistance in cancer, suggesting that only a comprehensive assessment of the genetic changes of the cancer genome, including somatic mutations and copy number changes, alongside a detailed description of the concomitant chromatin remodelling events ensuing after treatment, could finally provide the insights required to tackle this pressing unmet clinical need. Single cell analyses are needed to deconvolve genetic and epigenetic heterogeneity; however, currently available technologies are not able to probe both *omic* layers.

PRODUCT & TECHNOLOGY

We developed a novel single cell technology named "single-cell genome and epigenome by transposases sequencing" (scGET-seq) to comprehensively probe both open and closed chromatin and to concomitantly record the underlying genomic sequences.



scGET-seq exploits a hybrid transposase (TnH) including the chromodomain (CD) of the heterochromatin protein-1α (HP-1α), which is involved in heterochromatin assembly and maintenance through its binding to trimethylation of the lysine 9 on histone 3 (H3K9me3). scGET-seq data enables cell trajectory inference according to analysis of dynamic chromatin states. This approach also reduces the bias in sampling genomic sequences, increasing the resolution for CNV analysis.

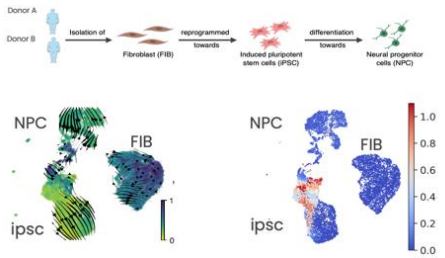
PRECLINICAL VALIDATION

In colorectal cancer derived organoids, scGETseq proved superior to scATAC-seq in identifying clonal structure In a PDX model of colon carcinoma, scGET-seq defines genomic plasticity driven by epigenetic remodeling and identifies resistance patterns to anti-EGFR treatment In a neuronal differentiation model, scGET-seq was able to discriminate the different subpopulations and, through the assessment of accessibility likelihood, identify cell trajectories and genomic regions involved in the transition. Such regions are associated with neural development and morphogenesis. From scGET-seq data it was also possible to compute a global transcription factor (TF) dynamic score, identifying cell type specific TFs.

SOLUTION/BENEFITS

There are several advantages introduced by the scGET-seq technology:

- comprehensively profiles hetero and euchromatin;
- captures cell-identity and CNVs at higher resolution respect to standard scATAC-seq;
- outperforms scATAC-seq to identify clonality;
- data can be used to characterize dynamic chromatin states by measuring the accessibility likelihood
- can provide important insights in fields as diverse as development, regenerative medicine and the study of human diseases, including cancer.



MARKET & COMPETITIVE CONTEXT

The global genomics market is projected to reach USD 54.4 billion by 2025 from USD 22.7 billion in 2020, at a CAGR of 19.0% during the forecast period. The major factors driving the growth of this market include the increasing government funding to support genomics projects, the growing incidence of cancer and increasing applications of NGS in cancer research, the entry of new players and start-ups in the genomics market, and the growing application areas of genomics (Genomics Market, MarketsandMarkets).

Adaptive cell therapy program

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Sector
Gene Therapy

Key Publication
Marinozzi M. *et al.,* J. Med. Chem 2017

Development stage
Preclinical Development

Business Model
Out-licensing for commercialization and/or sponsored research agreement with option rights

The Technology
Portfolio of 16 novel steroidal stigmasterol and ergosterol derivatives that function as Liver X Receptor (LXR) agonists.

CLINICAL NEED

Chimeric antigen receptors (CARs) and T cell receptors (TCRs) are artificial molecules combining antigen-binding and T-cell activating functions into a single receptor, to provide T cells with the ability to target a desired antigen. Most commonly, the antigen-binding moiety is represented by the single-chain fragment variable of a monoclonal antibody. T cells engineered to express a tumor-specific CAR/TCR (TCR/CAR T cells) represent a great promise in the context of cancer immunotherapy. Different institutions have recently reported impressive clinical responses by infusing CD19 CAR T cells in patients with B-cell acute lymphoblastic leukemia (ALL) or B-cell Lymphoma. However, there are increasing reports of subjects with ALL who relapsed following CD19 CAR therapy due to the loss of CD19 expression on leukemic cells (30-60%¥). In addition, first-in-man studies revealed unique hurdles contributing to the lack of a sharp demonstration of efficacy of CAR T cells in the context of solid malignancies.

SOLUTION & BENEFITS

To overcome current hurdles faced by adoptive cell therapy approaches, with respect to both hematological and solid tumors, we developed the below pipeline with the aim to:

- increase efficacy in solid tumors;
- improve fitness and performance of T cell products;
- overcome limitations of current CD19 therapies;
- allow future allogeneic T cell therapies.

Indeed, we identified new CAR and TCR specificities for both hematological and solid tumors and developed 'enabling' strategies to improve their antitumor efficacy through several mechanisms: (i) unmasking of tumor antigens and potentiating immunological synapse (de-glycosylation); (ii) reduction of inhibitory receptors' engagement (de-glycosylation and inhibitory receptor disruption); (iii) improvement of CAR T cells' fitness (de-glycosylation, IL7/15 and preselection methods), (iv) exerting a beneficial effect on the tumor microenvironment (TME) (de-glycosylation), (v) increasing T cell recruitment and infiltration at the tumor site (NGR-TNF), and (vi) allowing the production of allogeneic T cell products (ZINC finger nucleases-mediated approach).

PRODUCT & TECHNOLOGY

Categories	Technology	Field/Application	Development Stage
CAR T	CD44v6	Solid tumors (GI, Ovarian and Bladder)	In vivo PoC[1,2]
	Target undisclosed	Solid tumors (gastrointestinal tumors, Liver mets)	Lead opt./Animal test.*
	Target undisclosed		
TCR	Target undisclosed	CRC and PDAC (frequent HLA-elements)	Lead opt/Animal test*
		AML (frequent HLA-elements)	
Enablers	2-DeoxyGlucose	Combination with any CAR T	In vivo PoC[2]
	De-glycosylating CAR	Combination with any CAR T	Lead identification*
	NGR-TNF	Combination with any CAR T	In vivo PoC[3]
Methods	IL7/IL15	Manufacturing of engineered T cells highly enriched of stem cell memory T cells and characterized by improved fitness and safety	GMP-grade reagents[4]
	Preselection		In Vivo Poc[5]
	gRNA for IR disruption	Combination with any TCR/CAR in relevant tumor models	In vivo PoC*
	ZINC finger nucleases for TCR disruption.		In vivo PoC[6]

MARKET & COMPETITION

The global market for adoptive cell therapy applied to both hematological and solid tumors in the last three years is made up of 10.138 deals for an overall value of 1.1 trillion USD¥¥.

¥Xu et al. Front Immunol. 2019.
¥¥GlobalData, May 2022.

Duchenne Muscular Dystrophy (DMD) program

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Development stage
Preclinical Development

Key Publications
[1]Cossu et al. EMBO Mol Med 2015
[2]Careccia et al. Sci Transl Med. 2021
*Manuscript in preparation

Relevant Intellectual Property
[1]International patent application published as WO2007093412. Patent granted in US (US8071380). Patent pending in Europe

[2]International patent application published as WO2014016417. Patents granted in Europe (validated in 5 Countries) and in USA.

*Patent under filing.

CLINICAL NEED

Duchenne Muscular Dystrophy (DMD) is a devastating disease that occurs almost exclusively in males. Birth prevalence ranges from 1.6 to 3 per 10,000 live births (depending on different screenings). Loss of ambulation occurs at a median age of 12 and ventilation starts at about 20 years. There is international variation in use of corticosteroids, scoliosis surgery, ventilation and physiotherapy. The economic cost of DMD climbs dramatically with disease progression - rising as much as 5.7 fold from the early ambulatory phase to the non-ambulatory phase. DMD currently lacks an efficacious therapy and steroids are the only drugs that delay the progression of the disease but with serious side effects (Guiraud et al. 2015). On the other hand, new generation drugs (ASO for Exon skipping, small molecule for read-through premature nonsense stop codon), based upon correction of the mutated transcript, have not shown clear evidence of efficacy (McDonald et al. 2017; Lim et al. 2017), despite many trials and controversial market authorizations (Kesselheim & Avorn 2016).

SOLUTION & BENEFITS

To overcome the limitations that current approaches are experiencing within the treatment of DMD, and common to many other musculoskeletal disorders, we developed the below product pipeline enabling:

- A cell therapy approach, which proved to be safe in a Phase I clinical trial in 5 DMD patients. The approach is currently under optimization in order to develop an off the shelf, affordable product, for the creation of a universal donor mesoangioblast cell bank;
- A non-oxidizable form of HMGB1 (called 3S). Pharmacological treatment with the 3S variant improves functional performance, muscle regeneration and satellite cell engraftment in mouse models of DMD and Limb-Girdle Muscular Dystrophy type 2d (LGMD2D), while reducing inflammation and fibrosis;
- Matrin 3 (MATR3), a natural, physiologic, endogenous inhibitor of the transcription factor double homeobox 4 (DUX4). The aberrant expression of DUX4 is the main cause of FacioScapuloHumeral muscular Dystrophy (FSHD), for which there are no therapies. MATR3 binds directly to the DNA binding domain of DUX4 and blocks its activity

PRODUCT & TECHNOLOGY

Categories	Product	Field/Application	Development Stage
Cell therapy	Mesoangioblasts	Proved safety in 5 Duchenne Muscular Dystrophy (DMD) patients	PH I CT[1]
	Universal, Genetically Corrected Mesoangioblasts	- Human mesoangioblasts can be indefinitely expanded with the novel, proprietary, culture medium. - Off-the-shelf universal donor mesoangioblast cell bank. - Potential expansion to many recessive monogenic diseases of the mesoderm.	Lead opt.*
Biologics	Non-oxidable HMGB1	Targeting DMD regardless of the genetic mutation	In vivo PoC in DMD and limb-girdle MD[2]
	MATR3	- Safety of the approach (MATR3 fragment binds only to DUX4). - Versatile approach (MATR3 can be administered as biologics or gene therapy approach).	Lead opt/Animal test[3]
Enabler	HMGB1	Combination therapy with mesoangioblasts.	In vivo proof of concept[2]
Methods	Proprietary culture protocol for mesoangioblasts	Proprietary culture method	Research-grade reagents*

MARKET & COMPETITION

In 2020, the global Duchenne Muscular Dystrophy market size was USD 935 million, and it is expected to reach USD 9904.4 million by the end of 2027, with a CAGR of 42.1% during 2021-2027*. The propelling factors for the growth of the DMD treatment market include the rising disease burden, and increasing investments in biopharmaceuticals.