Supplementary information

Novel antisense therapy targeting microRNA-132 in patients with heart failure: results of a first-in-human phase 1b randomised, double-blind, placebo-controlled study.

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METHODS

Randomization

Patients in this study were assigned to a treatment regimen according to a randomization schedule generated by a statistician using PROC Plan, a SAS software procedure.

Methods for ensuring blinding

This clinical phase 1b study was conducted in a double-blind fashion whereby patients and clinical study site staff were blinded to CDR132L or placebo assignment. The clinical study site and patients were aware of the dose level being used. To maintain the double-blind, CDR132L and placebo were identical in appearance. The pharmacy staff preparing CDR132L or placebo were not blinded to study drug assignment.

During the study, the individual randomization codes were kept in the site's clinical trials pharmacy, accessible to the pharmacy personnel only. Upon completion of the study, after the database lock and after the blind was revealed, the randomization list was filed in the Trial Master File. Cardior staff involved in clinical decision-making were blinded to study drug assignment.

Inclusion and exclusion criteria for the clinical phase 1 study

Inclusion criteria

Subjects must meet all of the following criteria to be eligible for enrolment in this study:

1. Male or female, aged ≥ 30 to ≤ 80 years at the date of signing informed consent which is defined as the beginning of the Screening Period. This inclusion criterion will only be assessed at the Screening Visit.
2. Clinically stable patients with heart failure (NYHA 1-3 LVEF: ≥30%) of ischaemic origin.
3. A LVEF ≥30% and <50% at screening as measured by echocardiography or amino terminal fragment of pro-brain natriuretic peptide (NT-proBNP) level > 125 ng/l (though both assessments will be completed at the time-point indicated, only one is required for inclusion).
4. Female subjects of non-childbearing potential, e.g. post-menopausal (as defined as amenorrhoea for at least 12 months with no alternative medical cause) or permanently sterile (permanent sterilisation methods include hysterectomy, bilateral salpingectomy and bilateral oophorectomy).
5. Female subjects of childbearing potential (WOCBP) who anticipate being sexually active with a male during the study (from first IMP administration until the follow-up visit):

Contraception required: Highly effective contraception must start one complete menstrual cycle prior to the first day of dosing and continue until the end of the systemic exposure of the study

drug (for this study this is until the follow-up visit on Day 112). Highly effective contraception methods are defined as:

- Hormonal contraception:
  - Combined i.e. oestrogen- and progestogen-containing (oral, intravaginal or transdermal) associated with inhibition of ovulation
  - Progestogen-only (oral, injectable or implantable) associated with inhibition of ovulation
  - Intrauterine hormone-releasing system (IUS)
- Intrauterine device (IUD) or
- Bilateral tubal occlusion
- Male partner vasectomised (with documented evidence of azoospermia if possible)

6. Female subjects of childbearing potential (WOCBP) who agree to remain abstinent. Abstinence is defined as refraining from any sexual intercourse with a male for the duration of the study (in this trial this is from one week prior to first IMP administration - to be able to identify early pregnancy before dosing until the follow-up visit):

Contraception required: None. If the situation changes post-dose during the study, subjects must use an appropriate form of highly effective contraception as stated in (5) above from one complete menstrual cycle prior to the first intercourse with a male and continue until the follow-up visit.

For the avoidance of doubt, calendar, ovulation, symptothermal, post-ovulation methods and withdrawal do NOT meet the definition of abstinence. Although actively discouraged, females who wish to become sexually active with a male during the window of restriction must implement safe, highly effective methods of contraception one menstrual cycle before engaging in sexual activity.

7. To prevent transfer of IMP to a male or female partner or fetus/baby, all male subjects, including those who have had a vasectomy (with documented evidence of azoospermia if possible) must agree to use a barrier method (male condom) during intercourse with a male or female partner from the time of first IMP administration to follow-up for the stated time period.

To prevent a pregnancy, male subjects, if heterosexually active and with a female partner of childbearing potential must agree to use barrier contraception (male condom) for the treatment period until the follow-up. Female partners of male subjects who are of childbearing potential must have agreed to using or confirm having used one or more forms of highly effective contraception as defined above, starting at least one menstrual cycle before the first sexual intercourse and continue until the follow-up visit of the male partner.

8. Subjects must agree not to donate sperm or ova from the time of the first administration of study medication until 3 months after the end of the systemic exposure of the study drug.
9. Subjects must have a body mass index (BMI) between 18-28.0 kg/m^2 inclusive at screening, with maximum weight limit of 100 kg. Satisfactory medical assessment with no clinically significant medical conditions as judged by the PI or delegate, which are not well controlled and stable (e.g. Diabetes is permitted if good control is established

(stable HBA1c), other comorbidities permitted if well controlled and no active investigations or planned interventions)

10. Ability to provide written, personally signed, and dated informed consent to participate in the study, in accordance with the ICH Good Clinical Practice (GCP) Guideline E6 (R2) (2016) and applicable regulations, before completing any study-related procedures.
11. An understanding, ability, and willingness to fully comply with study procedures and restrictions.

Exclusion Criteria

Subjects will be excluded from enrolment in this study if they meet any of the following criteria:

1. Heart failure of non-ischemic origin: hypertensive heart disease, myocarditis, alcoholic cardiomyopathy and cardiac dysfunction due to rapid atrial fibrillation.
2. History of decompensated heart failure or a history of LVEF < 30 % within 3 months prior to screening period.
3. NYHA class IV at randomization.
4. Any planned cardiac intervention (angiogram without angioplasty is acceptable) or any other planned operations after screening period.
5. An estimated Glomerular Filtration Rate (eGFR) < 45 ml/min/1.73 m².
6. Clinically relevant hepatic dysfunction.
7. Medical history of diseases affecting the blood-brain-barrier, e.g. multiple sclerosis or stroke.
8. Medical history of bleeding disorders.
9. Thrombocytopenia < 100.000/ uL
10. HbA1c levels of ≥ 9. %
11. Current or recurrent disease; not including stable heart failure (e.g. haematological, neurological, endocrine, immunological, renal, hepatic or gastrointestinal or other conditions) that could affect the action, absorption, or disposition of CDR132L, or could affect clinical assessments or clinical laboratory evaluations.
12. Any history of seizures or epilepsy.
13. Current or relevant history of physical or psychiatric illness that are not stable or may require a change in treatment, use of prohibited therapies during the study or make the subject unlikely to fully comply with the requirements of the study or complete the study, or any condition that presents undue risk from the investigational product or study procedures.
14. Any other significant disease or disorder which, in the opinion of the Investigator, may either put the subject at risk because of participation in the study may influence the result of the study, or the subject's ability to participate in the study.
15. The history or presence of any of the following cardiac conditions: known structural cardiac abnormalities beyond what would be in keeping with heart failure); family history of long QT syndrome; cardiac syncope or recurrent, idiopathic syncope, in the opinion of the Investigator.
16. Any clinically significant abnormalities in rhythm, conduction or morphology of resting ECG that pose an additional safety risk to volunteers. This includes subjects with any of the following (at screening or Day -1):
    - Clinically significant PR (PQ) interval prolongation.
    - Intermittent second or third degree AV block.
    - Complete bundle branch block.

- Sustained cardiac arrhythmia's including (but not limited to) supraventricular tachycardias; any symptomatic arrhythmia with the exception of isolated extra systoles.

Subjects with abnormalities may be included if the deviations do not pose a safety risk (e.g. Atrial fibrillation is okay to include, but atrial flutter may not be depending on if sustained and impact on morphology) and if agreed between the appointed Cardiologist and the PI.

17. Has vital signs outside of the following normal range at screening or Day -1:
    a. Blood pressure (BP):

Supine BP (after at least 5 minutes of supine rest):

- Systolic blood pressure: 100 - 180 mmHg.
- Diastolic blood pressure: 50 - 110 mmHg.
- Heart rate: 50 – 100 bpm

Subjects with borderline abnormalities may be included if the deviations do not pose a safety risk, at the discretion of the PI.

18. Positive test for Hepatitis B surface antigen (HBsAg), Hepatitis C antibody (HCV Ab), or human immunodeficiency virus antibody (HIV Ab) at screening.
19. Any other abnormal findings on vital signs, ECG, physical examination or laboratory evaluation of blood and urine samples that the Investigator judges as likely to interfere with the study or pose an additional risk in participating.
20. Positive test results for alcohol or drugs of abuse at screening or Day -1.
21. Female subjects who are pregnant (including a positive serum pregnancy test at screening and on Day-1) or breastfeeding.
22. History or clinical evidence of substance and/or alcohol abuse within the 2 years before screening. Alcohol abuse is defined as regular weekly intake of more than 14 units (for both males and females), using the following NHS alcohol tracker (http://www.nhs.uk/Tools/Pages/drinks-tracker.aspx).
23. Has used any other prescription medication, over the counter medication or homeopathic preparations (excluding paracetamol, hormonal contraception, hormone replacement therapy, and the medications used to manage heart failure and diabetes as listed in the concomitant medications) within 14 days or 10 half-lives (whichever is longer) prior to Day 1 of the dosing period that the Investigator judges is likely to interfere with the study or pose an additional risk in participating.
24. Consumption of herbal remedies or dietary supplements containing St. John's Wort in the 3 weeks before the planned Day 1 of the dosing period.
25. Has received an investigational product or been treated with an investigational device within 90 days prior to first drug administration and will not start any other investigational product or device study within 90 days after last study drug administration.
26. Known or suspected intolerance or hypersensitivity to the investigational product, any closely related compound, or any of the stated ingredients.
27. History of significant allergic reaction (anaphylaxis, angioedema) to any product (food, pharmaceutical, etc).
28. Has donated or lost 400 mL blood or more within the last 16 weeks preceding the first day of dosing.
29. Has a mental incapacity or language barriers precluding adequate understanding, co-operation, and compliance with the study requirements
30. An inability to follow a standardised diet and meal schedule or inability to fast, as required during the study.

31. Prior screen failure (where the cause of the screen failure is not deemed to be temporary), randomisation, participation, or enrolment in this study. Subjects who initially failed due to temporary non-medically significant issues are eligible for re-screening once the cause has resolved.

Echocardiography and electrocardiographic variables

Echocardiographic and electrocardiographic variables were assessed in the study using standard clinical practices.[1] The parameters included left ventricular volume and left ventricular ejection fraction (LVEF), heart rate, QRS interval, and QT interval. QRS narrowing index was defined as the difference between post treatment and pre-treatment QRS interval normalized to the pre-treatment QRS interval, multiplied by 100 ((QRS interval [at day 112] – QRS interval [at day -1] / QRS interval [at day -1]) × 100).

Blood biomarker determinations

Besides standard haematology, coagulation, and other laboratory determinations (made at the certified in-house laboratory of Richmond Pharmacology, Ltd., which performed the clinical study at their research unit), an array of state-of-the-art blood biomarkers reflecting cardiac, renal, and systemic health were measured in this study as safety markers, and in some cases, also as exploratory pharmacodynamic endpoints. Additionally, CDR132L and microRNA-132 (miR-132), i.e., the active study drug and its molecular target, were measured in plasma for pharmacokinetic and exploratory pharmacodynamic analyses, respectively. All measurements of biomarkers were made at Prolytic GmbH, a certified laboratory in Frankurt/Main, Germany, except that Richmond Pharmacology performed (NT-proBNP) determinations. Axolabs GmbH, a certified laboratory in Kulmbach, Germany, measured plasma CDR132L, and the study sponsor, Cardior Pharmaceuticals, GmbH, measured plasma miR-132 at their in-house laboratory in Hannover, Germany. Blood samples for circulating safety, pharmacokinetic, or pharmacodynamic analyte determinations were immediately stored at -65°C, and, for analytes tested at Prolytic, Axolabs, or Cardior, shipped on dry ice via overnight delivery. Once at Prolytic, Axolabs, or Cardior, samples were immediately stored at -80°C until assays were performed. All assays were run by laboratory staff masked as to study treatment assignment (except for dosage).

Analysis of plasma miR-132 levels

For all patients, plasma miR-132 concentration was measured 10 times: at screening, immediately before and h after study treatment administrations on days 1 and 28, and on days 3, 10-14, 56, 84, and 112 (end-of-study). Additionally, plasma miR-132 concentration was assessed once in 30 self-reportedly healthy volunteers (samples were purchased from Central Biohub, Hennigsdorf, Germany). 10 out of these 30 samples were specifically selected to be gender- and age-matched (males, mean age 66±1 year). Since plasma miR-132 levels were comparable to plasma miR-132 levels detected in 20 additional neither age-, nor gender-matched samples (women, mean age 55±4 years) data sets were merged.

Quantitative real-time polymerase chain reaction (qRT-PCR) was used; this quantitation technique has been validated in large-animal studies[2,3] and in a 953-patient observational study [4]. Total RNA was isolated from plasma samples (150 µl each) with the miRNeasy Mini Serum/Plasma Kit (Qiagen, Hilden, Germany), according to manufacturer's instructions, including spike-in with 5.6 x 10^8 copies of synthetic cel-miR-39 as exogenous reference gene. Samples were eluted with 14 µl nuclease-free water, of which 2.5 µl were used for cDNA synthesis with the TaqMan MicroRNA Reverse Transcription (RT) Kit (Thermo Fisher Scientific, Waltham, MA, USA) and corresponding TaqMan MicroRNA RT Assays (Thermo Fisher Scientific).

qRT-PCR then was performed using the ABsolute Blue QPCR Mix (Thermo Fisher Scientific) and TaqMan MicroRNA Assays (Thermo Fisher Scientific; miR-132-3p: 000457 and cel-miR-39-3p: 00200) on a ViiA 7 Real-Time PCR System (Thermo Fisher Scientific). Data were collected and analysed using QuantStudio Real-Time PCR versions 1.1 and 1.3 (Thermo Fisher Scientific). Changes in plasma miRNA level were quantified using the $2^{-\Delta Ct}$ method, using the formula, plasma miR-132 = $2^{-(Ct\ miR\text{-}132\ -\ Ct\ cel\text{-}miR\text{-}39}$ Data points not fulfilling qRT-PCR quality criteria (either or both of standard deviation $[SD]_{duplicates}$>0.5 or no amplification) were considered inevaluable.

Pharmacokinetic/pharmacodynamic modelling of projected monthly and quarterly therapeutic dose ranges

For ethical and practical (i.e., study recruitment-related) reasons, cardiac tissue samples, and hence, measurements of CDR132L and miR-132 concentrations in heart tissue, were unavailable in this phase 1b study of patients with stable heart failure. Lack of these data potentially complicated calculation of estimated therapeutic dose ranges for phase 2 studies of monthly or quarterly CDR132L therapy of patients with heart failure. To address this issue, we used a novel approach of modelling exposure of CDR132L in human cardiac tissue through translation of pig pharmacokinetic/pharmacodynamic data to patients. This evaluation aimed to extrapolate pig pharmacokinetic/pharmacodynamic data to humans for variables where actual human data were unobtainable.

The administration schedule (day 1 and day 28) in the present study matched the schedule in a pharmacodynamic study in a pig model of early post-myocardial infarction heart failure[2], and certain doses (1 and 10 mg/kg) overlapped between the two trials (the pig study examined a 5 mg/kg dose, while we also examined 0.32 and 3 mg/kg doses). Therefore, data from this pig study were regarded as the most relevant for projection of human day 56 cardiac tissue data. For projection of human day 112 cardiac tissue data, data from a pharmacodynamic study[3] in a pig model of chronic post-myocardial infarction were included. In this latter study, CDR132L, 5 mg/kg, was given in 3 or 5 monthly infusions.

Measurement methods underwent minor adjustments from study to study, in response to changes in reagents and due to a continuous effort to improve quality and ease of measurement. Therefore, percentage adjustment of data from the pig studies was performed whenever appropriate. The calculation and modelling scheme itself were stepwise, consisting of:

1. Modelling of functional changes (delta left ventricular ejection fraction [LVEF]) versus miR-132 concentration in cardiac tissue in the pig study using a model of

early heart failure[2] to determine a therapeutically-relevant window of cardiac functional improvement and corresponding miR-132 level reduction in the heart.
2. Modelling of cardiac tissue and plasma miR-132 levels (derived from preclinical pig study of early heart failure) in active treatment relative to placebo groups to generate a graph equation to use human plasma concentrations to project human cardiac tissue concentrations as percentages of the baseline cardiac tissue miR-132 concentration (in the range of 100% to 0% of baseline).
3. Plotting of extrapolated miR-132 levels in human cardiac tissue against CDR132L dose in the relevant miR-132 tissue concentration range (human projection against dose).
4. Calculation of CDR132L pharmacokinetic variables (area under the plasma concentration curve from time zero to infinity [$AUC_{0\text{-}inf}$], maximum concentration [C_{max}]) and calculation of the CDR132L dose required in humans for efficacious monthly (every 28 days) therapy.
5. Determination of dosing frequency for efficacious long-term dosing in patients.

In general, non-parametric methods were preferred for our modelling, due to the small sample size. Also due to the limited number of patients, it was not possible to split the sample into a test set and a validation set and to validate results with appropriate re-sampling methods. Therefore, the approach presented here must be regarded as exploratory.

To model the relationship between delta LVEF and miR-132 in heart tissue, different functions were executed in order to achieve the best fit of the underlying curve to the data. The function achieving the best R^2 with optimised coefficients was ultimately chosen. Translational modelling was generated following the same criteria.

Limitations of the study

There are important limitations as this was a phase 1b trial not designed and powered to demonstrate clinical efficacy. However, the developed PK/PD modelling approach and the observed PD data will be used for designing further Phase 2 proof of concept studies. Therefore, CDR132L's target reduction and all other pharmacodynamic variables were examined in an exploratory fashion. However, the sample size (N=28) was sufficient to set up a safety profile up to 3 months follow up after the second dose and a PK profile for CDR132L in those doses tested. Another limitation was that no PK or PD data could be collected from cardiac tissue due to ethical consideration. However, we were able to combine pharmacokinetic and pharmacodynamic data from plasma measurements in the present study and such data from cardiac tissue as well as plasma measurements from large-animal studies with large sample sizes, to project dosage ranges for future clinical investigation. This new methodology offers a strategy to mitigate the lack of access to cardiac tissue common in phase 1 studies in cardiovascular disease settings.

Supplementary references

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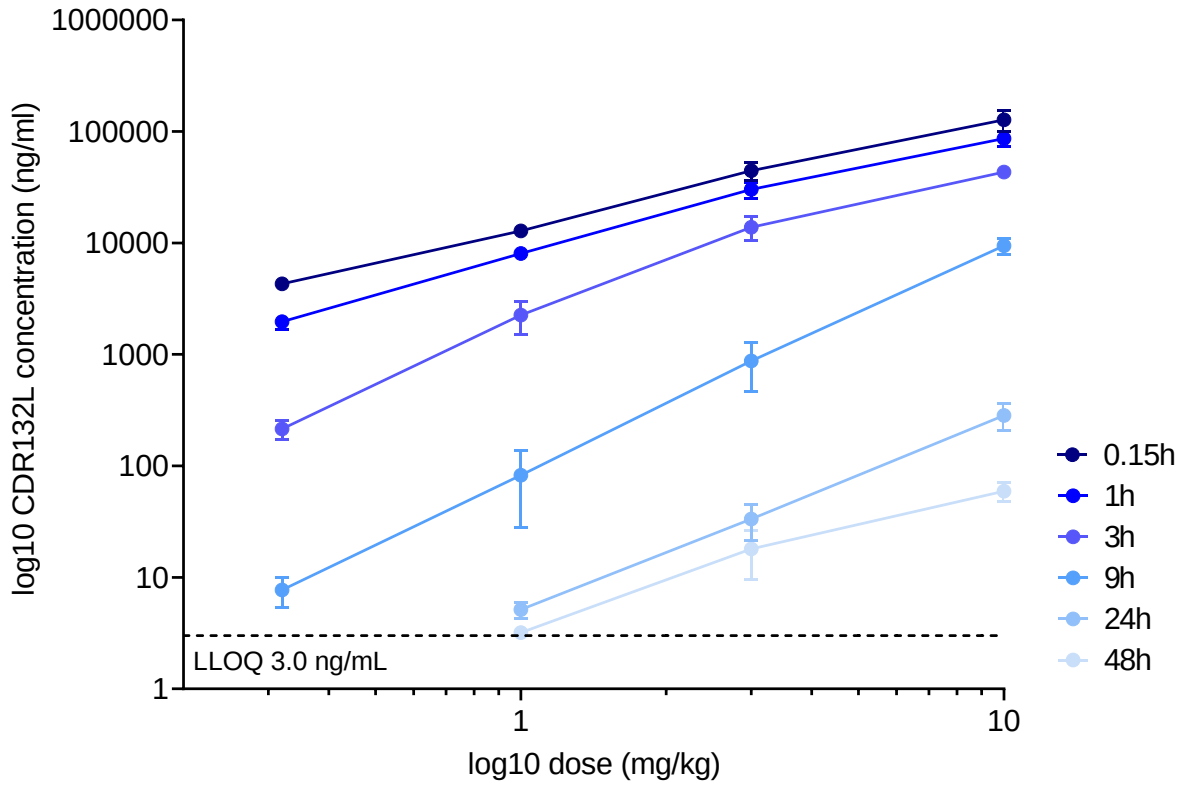
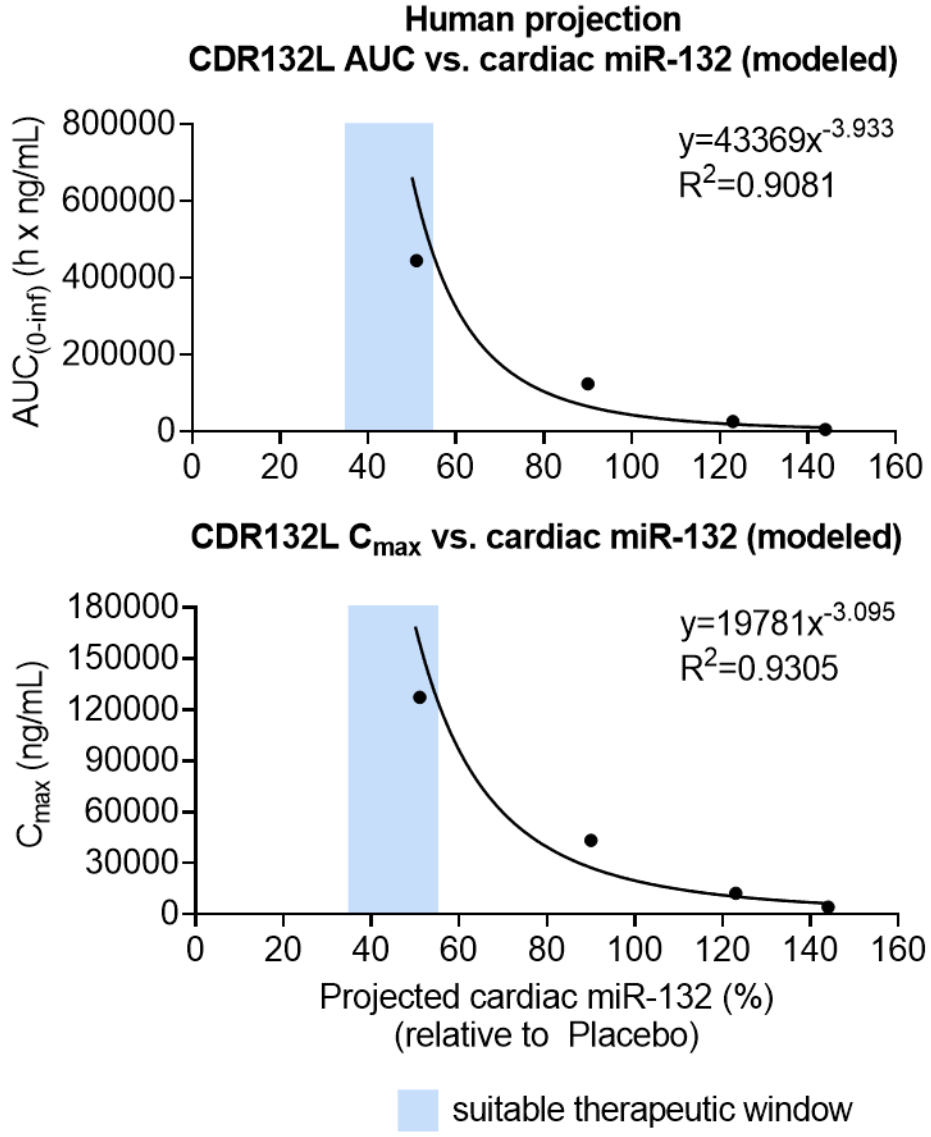
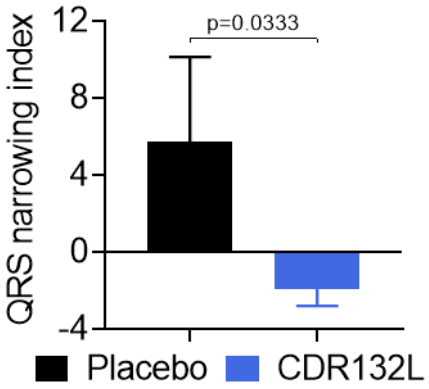


Supplementary Figure 1: **Plasma CDR132L concentrations over treatment dose through 48h after CDR132L administrations (first and second dosing combined).** Data are mean (95% CI). Measurements were taken 9 minutes, and 1, 3, 9, 24, and 48 h after CDR132L administration. CDR132L concentration fell below the lower limit of quantification (LLOQ) at 9h and subsequent timepoints for the treatment group receiving 0.32 mg/kg CDR132L.



Supplementary Figure 2: **Calculation of CDR132L AUC, C_{max} and dose required in human for efficacious monthly therapy.** Projected human cardiac miR-132 tissue level on day 56 post treatment, plotted against AUC and C_{max} in the relevant miR-132 % tissue concentration range.



Supplementary Figure 3: **Changes in QRS narrowing index, before first study treatment versus end-of-study (day 112).** Mean±SEM changes in QRS narrowing index. (Placebo n=3, CDR132L treatment groups n=7).

Supplementary Table 1: Adverse events

Category (n (%) patients; n events)	**Cohort 1 (CDR132L, 0.32 mg/kg) (n=5 patients, n=10 doses)***	**Cohort 2 (CDR132L, 1 mg/kg) (n=5 patients, n=10 doses)**	**Cohort 3 (CDR132L, 3 mg/kg) (n=5 patients, n=9 doses)†**	**Cohort 4 (CDR132L, 10 mg/kg) (n=5 patients, n=10 doses)**	**Placebo (0.9% saline) (n=8 patients, n=16 doses)**
Any adverse event	3 (60%); 6	4 (80%); 10	4 (80%); 8	4 (80%); 9	7 (88%); 20
Any serious adverse event	0	0	0	0	0
Any adverse event of special interest‡	0	0	0	0	0
Any adverse event leading to study withdrawal	0	0	0	0	0
Adverse event severity	..	..	..	..	..
Mild	3 (60%); 6	4 (80%); 9	4 (80%); 8	4 (80%); 8	7 (88%); 20
Moderate	0	1 (20%); 1	0	1 (20%); 1	0
Severe	0	0	0	0	0
Any study treatment-related adverse event	0	0	1 (20%); 2	1 (20%); 2	2 (25%); 2
Any study treatment-related adverse event of special interest‡	0	0	0	0	0
Any concomitant medication-related adverse event	0	0	1 (20%); 1	1 (20%); 1	0

* Includes one administration of CDR132L that inadvertently was given paravasally instead of intravenously. This patient was included in this analysis.

† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L, which is included in the safety/tolerability analysis. This patient was included in this analysis.

‡ In this study, thrombocytopenia was defined as an adverse event of special interest, due to reports of this toxicity being associated with administration of certain "first-generation" ASOs in clinical studies.

Supplementary Table 2: Study treatment-related adverse events

Adverse event (n patients (%); n events)	**Cohort 1 (CDR132L, 0.32 mg/kg) (n=5 patients, n=10 doses)***	**Cohort 2 (CDR132L, 1 mg/kg) (n=5 patients, n=10 doses)**	**Cohort 3 (CDR132L, 3 mg/kg) (n=5 patients, n=9 doses)†**	**Cohort 4 (CDR132L, 10 mg/kg) (n=5 patients, n=10 doses)**	**Placebo (0.9% saline) (n=8 patients, n=16 doses)**
Total	0	0	1 (20%); 2	1 (20%); 2	2 (25%); 2
Dizziness	0	0	1 (20%); 1	1 (20%); 1	1 (13%); 1
Euphoric mood	0	0	1 (20%); 1	0	1 (13%); 1
Diarrhoea	0	0	0	1 (20%); 1	0

* Includes one administration of CDR132L that inadvertently was given paravasally instead of intravenously. This patient was included in this analysis.

† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L, which is included in the safety/tolerability analysis. This patient was included in this analysis.

Supplementary Table 3: Pharmacokinetics of intravenous CDR132L by dose cohort

	Cohort 1 (CDR132L, 0.32 mg/kg) (n=5 patients, n=9 doses)*	**Cohort 2 (CDR132L, 1 mg/kg) (n=5 patients, n=10 doses)**	**Cohort 3 (CDR132L, 3 mg/kg) (n=4 patients, n=9 doses)†**	**Cohort 4 (CDR132L, 10 mg/kg) (n=5 patients, n=10 doses)**
Day 1				
C_{max} (ng/mL)	4357.075 (473.306)	13209.16 (1457.641)	45535.94 (7286.316)	126963.14 (46410.337)
t_{max} (h)	0.404 (0.008)	0.403 (0.007)	0.407 (0.015)	0.57 (0.38)
$t_{½}$ (h)	1.038 (0.069)	5.207 (4.474)	4.244 (0.239)	4.346 (0.229)
λ_z (1/h)	0.67 (0.044)	0.198 (0.107)	0.164 (0.009)	0.16 (0.008)
$AUC_{0\text{-}t}$ (h*ng/mL)	5392.035 (737.494)	25264.486 (5885.729)	116537.16 (32005.567)	424301.866 (71319.403)
$AUC_{0\text{-}inf}$ (h*ng/mL)	5403.536 (741.727)	25289.533 (5889.116)	116628.451 (32046.812)	424609.461 (71319.249)
V_z (L)	7.054 (1.267)	24.166 (21.972)	14.201 (2.979)	11.753 (1.316)
CL (L/h)	4.724 (0.925)	3.217 (0.725)	2.334 (0.558)	1.875 (0.2)
Day 28				
	Cohort 1 (CDR132L, 0.32 mg/kg) (n=5 patients, n=9 doses) †	**Cohort 2 (CDR132L, 1 mg/kg) (n=5 patients, n=10 doses)**	**Cohort 3 (CDR132L, 3 mg/kg) (n=4 patients, n=9 doses†)**	**Cohort 4 (CDR132L, 10 mg/kg) (n=5 patients, n=10 doses)**
Cmax (ng/mL)	4257.68 (726.876)	12386.86 (1942.225)	43398.65 (15517.698)	127426.36 (35483.565)
tmax (h)	0.4 (0)	0.403 (0.007)	0.508 (0.142)	0.4 (0)
t½ (h)	1.056 (0.056)	5.095 (5.15)	4.594 (0.344)	4.57 (0.178)
λz (1/h)	0.658 (0.037)	0.223 (0.116)	0.152 (0.011)	0.152 (0.006)
AUC0-t (h*ng/mL)	5268.442 (985.505)	24487.184 (5174.646)	109434.756 (32055.356)	407547.331 (58055.114)
AUC0-inf (h*ng/mL)	5280.611 (987.861)	24514.76 (5178.426)	109585.811 (32112.526)	408010.193 (58116.759)
Vz (L)	7.349 (1.301)	24.682 (26.064)	16.746 (5.13)	12.87 (1.49)
CL (L/h)	4.828 (0.81)	3.282 (0.412)	2.528 (0.726)	1.955 (0.238)

Data are mean (SD) and include both the day 1 and day 28 CDR132L administrations for all patients in the respective cohort, except where noted otherwise. SD=standard deviation. C_{max}= maximum concentration. T_{max}=time to C_{max}. $AUC_{0\text{-}t}$=area under the plasma concentration curve from time zero to the last quantifiable concentration. $AUC_{0\text{-}inf}$=area under the plasma concentration curve from time zero to infinity. λ_z=terminal rate constant. $t_{1/2}$=terminal elimination half-life. V_d=volume of distribution. CL=blood clearance.

* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was excluded from the analysis of the first dosing.

† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was included in this analysis.

Supplementary Table 4: **Plasma miR-132 concentrations by treatment group**

..	**Cohort 1 (CDR132L, 0.32 mg/kg) (n=5 patients, n=10 doses)***	**Cohort 2 (CDR132L, 1 mg/kg) (n=5 patients, n-10 doses)**	**Cohort 3 (CDR132L, 3 mg/kg) (n=4 patients, n=9 doses)†**	**Cohort 4 (CDR132L, 10 mg/kg) (n=5 patients, n=10 doses)**	**Placebo (0.9% saline) (n=8 patients, n=16 doses)**
Day 1 (pre-first administration)	0.0343	0.0068	0.0147	0.0055	0.0077
Day 28 (pre-second administration)	0.0021	0.0007	0.0006	0.0003	0.0089
Relative change, day 28 vs day 1, %	10.5	10.7	4.9	3.8	86.5
Day 112 (end-of-study)	0.0045	0.0015	0.0013	0.0014	0.007
Relative change, day 112 vs day 1, %	10	27.9	9.1	30.9	77.5

Data are median.

* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was included in this analysis.

† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was excluded from this analysis.

Supplementary Table 5: **Summary statistics for NT-proBNP (amino terminal fragment of pro-brain natriuretic peptide) pre-post difference**

	PD-active (n=14)*	**non PD-active (n=13)†**
Absolute change		
Mean (SD)	-93.86 (271.29)	17.46 (144.21)
Minimum	-736.0	-206.0
Median	-53.50	3.00
Maximum	463.0	398.0
Relative change (%)		
Mean (SD)	-17.44 (33.77)	-3.35 (28.44)
Minimum	-56.5	-41.8
Median	-23.34	0.88
Maximum	56.4	47.5
Responder (%)	71.43	46.15
Non-responder (%)	28.57	53.85
Statistics		
p-value Fisher's exact test	0.2519	
Phi Coefficient	0.2570	
Odds Ratio (95% confidence limits)	2.9167 (lower: 0.5938; upper: 14.3270)	

PD-active: cohort 2 (1 mg/kg), cohort 3 (3 mg/kg) and cohort 4 (10 mg/kg);
non PD-active: Placebo and cohort 1 (0.32 mg/kg)
* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was included in this analysis.
† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was excluded from this analysis.

Supplementary Table 6: **Summary statistics for NT-proBNP and LVEF improvement (delta LVEF) combined pre-post difference**

	PD-active (n=14)*	**non PD-active (n=13)†**
Responder (%)	78.57	53.85
Non-responder (%)	21.43	46.15
Statistics		
p-value Fisher's exact test	0.1201	
Phi Coefficient	0.3354	
Odds Ratio (95% confidence limits)	4.2778 (lower: 0.7981; upper: 22.9276)	

PD-active: cohort 2 (1 mg/kg), cohort 3 (3 mg/kg) and cohort 4 (10 mg/kg);
non PD-active: Placebo and cohort 1 (0.32 mg/kg)
* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was included in this analysis.
† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was excluded from this analysis.

Supplementary Table 7: Summary statistics for GAL-3 (Galectin 3) pre-post difference

	PD-active (n=14)*	**non PD-active (n=13)†**
Absolute change		
Mean (SD)	-2.40 (5.94)	-0.39 (3.47)
Minimum	-20.6	-7.4
Median	-0.65	-0.72
Maximum	3.2	4.3
Relative change (%)		
Mean (SD)	-8.57 (20.15)	0.18 (18.07)
Minimum	-51.7	-26.3
Median	-4.98	-4.06
Maximum	22.0	27.9
Responder (%)	64.29	53.85
Non-responder (%)	35.71	46.15
Statistics		
p-value Fisher's exact test	0.7036	
Phi Coefficient	0.1062	
Odds Ratio (95% confidence limits)	1.5429 (lower: 0.3294; upper: 7.2261)	

PD-active: cohort 2 (1 mg/kg), cohort 3 (3 mg/kg) and cohort 4 (10 mg/kg);
non PD-active: Placebo and cohort 1 (0.32 mg/kg)
* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was included in this analysis.
† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was excluded from this analysis.

Supplementary Table 8: **Summary statistics for ST-2 (suppression of tumourgenicity-2) pre-post difference**

	PD-active (n=14)*	**non PD-active (n=13)†**
Absolute change		
Mean (SD)	-0.05 (3.49)	1.49 (3.01)
Minimum	-4.4	-1.5
Median	-1.02	1.59
Maximum	7	9.5
Relative change (%)		
Mean (SD)	1.48 (13.63)	6.65 (13.24)
Minimum	-18.4	-6.4
Median	-1.87	8.06
Maximum	31.7	44.5
Responder (%)	50.00	38.46
Non-responder (%)	50.00	61.54
Statistics		
p-value Fisher's exact test	0.7036	
Phi Coefficient	0.1160	
Odds Ratio (95% confidence limits)	1.6000 (lower: 0.3459; upper: 7.4015)	

PD-active: cohort 2 (1 mg/kg), cohort 3 (3 mg/kg) and cohort 4 (10 mg/kg);
non PD-active: Placebo and cohort 1 (0.32 mg/kg)
* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was included in this analysis.
† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was excluded from this analysis.

Supplementary Table 9: Summary statistics for NGAL (lipocalin-2) pre-post difference

	PD-active (n=14)*	**non PD-active (n=13)†**
Absolute change		
Mean (SD)	-0.11 (0.4)	0.03 (0.3)
Minimum	-0.9	-0.6
Median	-0.05	0.02
Maximum	0.5	0.7
Relative change (%)		
Mean (SD)	-4.48 (14.81)	2.8 (14.52)
Minimum	-26.7	-24.5
Median	-1.92	0.98
Maximum	23.4	35.5
Responder (%)	64.29	46.15
Non-responder (%)	35.71	53.85
Statistics		
p-value Fisher's exact test	0.4495	
Phi Coefficient	-0.1823	
Odds Ratio (95% confidence limits)	2.1000 (lower: 0.4484; upper: 9.8356)	

PD-active: cohort 2 (1 mg/kg), cohort 3 (3 mg/kg) and cohort 4 (10 mg/kg);
non PD-active: Placebo and cohort 1 (0.32 mg/kg)
* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was included in this analysis.
† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was excluded from this analysis.

Supplementary Table 10: **Summary statistics for MMP-1 (Matrix Metallopeptidase 1) pre-post difference**

	PD-active (n=6)*	**non PD-active (n=8)†**
Absolute change		
Mean (SD)	-1754 (9549.91)	-213.2 (5310.25)
Minimum	-11511	-6252.4
Median	-3303	-866.1
Maximum	15646.5	10048.9
Relative change (%)		
Mean (SD)	-13.49 (45.42)	4.49 (44.21)
Minimum	-81.4	-45.4
Median	-16.79	-1.43
Maximum	57.8	97.9
Responder (%)	64.29	53.85
Non-responder (%)	35.71	46.15
Statistics		
p-value Fisher's exact test	0.6270	
Phi Coefficient	0.1667	
Odds Ratio (95% confidence limits)	2.0000 (lower: 0.2235; upper: 17.8938)	

PD-active: cohort 2 (1 mg/kg), cohort 3 (3 mg/kg) and cohort 4 (10 mg/kg);
non PD-active: Placebo and cohort 1 (0.32 mg/kg)
* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was included in this analysis.
† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was excluded from this analysis.

Supplementary Table 11: **Summary statistics for QRS complex narrowing pre-post difference**

	PD-active (n=4)*	**non PD-active (n=6)†**
Absolute change		
Mean (SD)	-3.95 (2.61)	3.33 (7.84)
Minimum	-7.3	-4.6
Median	-3.33	1.68
Maximum	-1.8	17.2
Relative change (%)		
Mean (SD)	-2.68 (1.62)	2.44 (6.35)
Minimum	-4.8	-3.8
Median	-2.28	0.96
Maximum	-1.4	14.1
Responder (%)	100.00	50.00
Non-responder (%)	0.00	50.00
Statistics		
p-value Fisher's exact test	0.2000	
Phi Coefficient	0.5345	
Odds Ratio (95% confidence limits)	not calculable	

PD-active: cohort 2 (1 mg/kg), cohort 3 (3 mg/kg) and cohort 4 (10 mg/kg);
non PD-active: Placebo and cohort 1 (0.32 mg/kg)
* In one patient in the CDR132L, 0.32 mg/kg cohort, the first administration drug was inadvertently given paravasally instead of intravenously. This patient was included in this analysis.
† One patient in the CDR132L, 3 mg/kg cohort withdrew from study for personal reasons on day 4 and therefore received only the first dose of CDR132L. This patient was excluded from this analysis.