90

Begeleidingsformulier aanvraag dierproef DEC- UM

Versie 2006

Herziene versie

DECNR: 2011-123
Ontvangen: 20-10-2011

DEC datum goedkeuring#	Type aanvraag [2]
26-10-2011	Nieuw / Herz.versie / Pilot

VROM	LNV/CBDNR[4]

Hoofdproject	CARIM
Deelproject	2.

Financieel beheerder		Budgetnummer	30982250B

Titel van het onderzoek:

A new treatment for the prevention of HF following cardiac hypertrophy: evaluation of three different drugs

The project has been approved by the PI of CARIM of this project

startdatum 1-11-2011 einddatum [9] 1-11-2014 *Duur van de proef [10]:* *9 weken*

	Naam	Tel (+ Tel privé enkel VO, VVO en VM)	E-mailadres	Bevoegd-heid[5]	Cap. groep /afdeling
1.Verantwoordelijk onderzoeker (VO)				Art.9	Farmacol o-gie
2. Vervanger VO (VVO)				Art.9	
				Art.9	
				Art.9	
4. overige uitvoerenden			nl	Art.12	
4. overige uitvoerenden				Art.12	
4. overige uitvoerenden				Art.12	

Diergroep	1	2	3	4	5	6	7	8
ctrl/exp/sham	sham	Sham + drug A	Sham + drug B	Sham + drug C	TAC + vehicle A, B, C	TAC + drug A	TAC + drug B	TAC + drug C
Diersoort	01	01	01	01	01	01	01	01
Stam	C57BL/6J	C57BL/6J	C57BL/6J	C57BL/6J	C57BL/6J	C57BL/6J	C57BL/6J	C57BL/6J
Construct / mutatie	-	-	-	-	-	-	-	-
Herkomst (leverancier) *	02	02	02	02	02	02	02	02
Aantal	42	17	17	17	21	34	34	34
Geslacht	m	m	m	m	m	m	m	m
immuuncompetent	yes	yes	yes	yes	yes	yes	yes	yes
Leeftijd/gewicht	>10 weeks	>10 weeks	>10 weeks	>10 weeks	>10 weeks	>10 weeks	>10 weeks	>10 weeks
Doel van de proef *	31	31	31	31	31	31	31	31
Belang van de proef	01	01	01	01	01	01	01	01
Tox. onderzoek *	01	01	01	01	01	01	01	01
Bijz. technieken *	01	01	01	01	01	01	01	01
Anesthesie *	04	04	04	04	04	04	04	04
Pijnbestrijding *	04	04	04	04	04	04	04	04
Mate ongerief *	04	04	04	04	05	05	05	05
Toestand dier einde	01	01	01	01	01	01	01	01

1 Verantwoording

Aanvraag dierproef DEC-UM **(kaders zijn licht flexibel, maar het geheel is max. 5 pag. versie 2006)**

Titel: A new treatment for the prevention of HF following cardiac hypertrophy: evaluation of three different drugs

1. Doel van de proef.

Traditional approaches to classify heart failure (HF) in patients are not robust enough and for that reason many people with cardiac disease are diagnosed too late with HF. To improve the early identification of these patients routine clinical diagnosis techniques are moving towards a biomarker profile evaluation. Currently, the most reliable biomarkers for HF are "late markers" like pro-BNP, ANP and C-reactive protein. Unfortunately there is not yet an early marker that is able to predict HF development. However, there is consensus that there is much room for improvement in this field, especially by using inflammatory markers since HF is associated with inflammation in an early stage.
In our previous study (DEC2009-107) we investigated whether circulating inflammatory markers that are released due to hypertrophic remodeling of the heart can predict the development of HF before physiological symptoms of HF arise. By studying a large number of pro- and anti-inflammatory components (both the good and bad ones) in rodent (both rat and mice) models of HF we detected 5 inflammatory markers which are expressed during the early progression of HF. Interestingly, 2 out of 5 of these markers appear to be ligands . This receptor has been investigated in several inflammatory diseases but not yet in relation to cardiovascular diseases. It has been shown that s responsible for migration of a specific cellular-mediated response and we hypothesize tha is important for the migration of lymphocytes into the myocardial tissue. Our current aim is therefore to block the ignaling in a pharmacological way in order to examine if this treatment may prevent the pathophysiological remodeling of the heart and to have a better understanding of the molecular mechanisms involved. The proposed experiment is conducted according to the plan described within the TRIUMPH project of the Dutch CTMM (Center for Translational Molecular Medicine) initiative (sponsored by the government) in which the CARIM theme II research group of the department takes part. TRIUMPH is an acronym for a conglomerate of several academic and commercial partners in the NL that work together on a broad aspect of technologies that are developed to identify patients at risk for HF as early as possible.

2. Maatschappelijke relevantie en/of wetenschappelijk belang

This study will provide more insight into the potential beneficial effects of treating cardiac inflammation during the development of heart failure. HF is a growing worldwide health problem and is one of the most important causes of mortality. In this study we try to establish if new drugs in this area can prevent the development of HF. If so these compounds might also then be considered for improving the clinical treatment.

3. Alternatieven

The inflammatory processes within the myocardium are complex and many different inflammatory proteins and cell types are involved. Due to the complex in vivo interaction between cardiac proteins, neurohormones, inflammatory cytokines and different inflammatory cell types it will be impossible to study these processes in cell cultures.

4. Ethische afweging

Cardiovascular disease is one of the most important causes of death. In the Netherlands, 45.000 people die each year because of cardiovascular diseases (±120 per day). Cardiac inflammation plays an important role in the development and progression of several types of cardiovascular diseases and early identification of the patients at risk will improve the success of treatment and survival of these patients. In our previous investigation we identified 5 inflammatory molecules that are up-regulated in the early stage of heart failure in TAC model. Two of these molecules are interestingly the same and only ligands) known to be the trigger of a specific bunch of inflammatory cells In this consecutive experiment we will block t in order to deny the response and to investigate whether such treatment could stop the development of cardiac hypertrophy and therefore the development of heart failure. The scientific profit and potential clinical application justify the use of these **severe intervention in rodents** to achieve the above mentioned study goals especially because human and cellular alternatives are not an option.

5. Wetenschappelijke onderbouwing

Nowadays the hypothesis that inflammation plays a key role in the development and progression of cardiovascular diseases and especially heart failure (HF) is quite consolidated. However, the clinical application of anti-inflammatory therapy in preventing HF is limited because of controversial results that have been achieved in early clinical trials with such therapeutic approaches (i.e. the potential application of TNF-alpha scavenging antibodies in HF is therefore still under investigation and discussed). Among the reasons that may explain failure for getting clear cut results in these intervention trials (in contrast to results obtained in selected animal studies) is the view that the inflammatory response in HF is rather complex and involves both pro- and anti-inflammatory components. The interventions that have been tried can be regarded as being rather non-selective. The interventions may have affected both the pro-inflammatory components (the bad guys) as well as the anti-inflammatory components (the good guys), whereas it is believed that a more balanced approach may be more successful. Before such an approach can be carried out the inflammatory characteristics of HF should be examined in detail. Thanks to a new screening approach, named Proteome Profiler™ we have been able to identify the up-regulation of 5 new inflammatory markers for heart failure. Within these 5 markers, two are ligands . The significance of this receptor has not been fully uncovered, but it has been proven that the receptor is expressed on esponsive cells. Given our first data we hypothesize that the blockade of during the development of induced cardiac hypertrophy in TAC mice will give us more insight in the role of this receptor in the remodeling of the heart. Moreover we would have the opportunity to show the potential effectiveness of a new treatment for cardiac hypertrophy.

In order to investigate signaling in HF we will use the same severe cardiac hypertrophic mouse model as used in DEC2009-107. The TAC model was chosen above the infarct model (MI) because in the first model all tissue of the whole heart can be used for all biochemical assays, whereas after an infarct cardiac tissue is inhomogeneous. Moreover MI animals develop a different type of cardiac remodeling than TAC animals. If we would choose a transgenic model the intrinsic phenotype is probably different because of the transgene. This is not logical because the present study is a follow up of DEC2009-107.

We plan to treat severe cardiac hypertrophy in mice induced by surgical transverse aortic constriction (TAC) with three different antagonists; their respective vehicle solutions will serve as controls. To assess the validity of the 5 biomarkers in these studies it is important to examine the presence of expression of these biomarkers at different time points after induction of TAC and the start of the treatment regimen. We plan to do this by taking regularly (every 2 weeks) small blood samples in which the whole array of cytokines can be measured. Urine samples and tissue samples and hemodynamic cardiac function measurements will be examined at 8 and 4 weeks after TAC. We will correlate all parameters to the well established molecules that characterize the severity of HF such as BNP, ANP and CRP. If urine samples can be used then it will be possible to shift from invasive diagnosis to non-invasive diagnosis.

In this study we want to compare the efficacy of 3 different drugs. The three agents have a different chemical entity and it is important to measure the potential effect of both drugs on the heart; this test has not been investigated before. Firstly we will test the drugs in the "established HF groups" (8wks TAC operated animals have uncompensated HF) and only in case of the drugs show a beneficial effect we will proceed with the "developing HF groups" (4wks TAC operated animals have compensated HF).

6. Wetenschappelijke beoordeling

This study is part of Triumph, a CTMM (Center for Translational Molecular Medicine) project, work package 1: "Biomarker discovery" and has been approved by the PI of this project (zie voorblad)

4 Proefdier

7. Proefdier keuze
7a. Soort, stam / herkomst / eindbestemming

Mice: C57BL/6J from Charles River.

After the study mice are sacrificed in order to obtain the organs and blood for further analysis. Re-use is therefore impossible.

7b. Sexe
Mice: males.
Aorta constriction results in pressure overload leading to left ventricular hypertrophy and depressed contractile reserves that is more severe in males than in females. In response to pressure overload, female animals may develop HF in a lesser degree and death occurs at later time points than in males. Therefore we plan to include only males to limit the variation.

7.c. Aantallen
An important parameter in this study is the cytokine concentration in plasma or serum before, during and after treatment. Therefore this parameter will be used in the power analysis with the formula:
$n=2(Z_{\alpha/2}+Z_{\Pi})^2 *(\sigma/\delta)^2$ (L. Sachs).

With a coefficient of variation of 25% (σ), Power of 80%, and α of 0.05, we want to detect a difference of 30% between the sham group and the experimental group. The value of the variation coefficient and 30% difference are based upon the previous study (DEC_2009-107)

In total we need $n=15.7*(0.25/0.30)^2= 11$ animals per group.

As about 35% of the animals (samples) may be lost due to different reasons, like

- the development of acute HF during the TAC surgery,
- failure of obtaining good cardiac function measurements,
- technical failure of the (Proteome Profiler™ assay),

more animals are needed.

Therefore we need n= 11/(1-0.35=0.65) = **17 animals per group**

5 Dierproef

8. Experiment

Species and total amount of animals:
Experiment: Mice C57BL/6j (
1. (n=21) Sham-operated mice + vehicle of drug A,B,C for 8wks treatment **(*A)**
2. (n=21) Sham-operated mice + vehicle of drug A,B,C for 4wks treatment **(*B)**
3. (n=17) Sham-operated + drug A for 8 wks (only)
4. (n=17) Sham-operated + drug B for 8 wks (only)
5. (n=17) Sham-operated + drug C for 8 wks (only)
6. (n=21) TAC operated + vehicle of drug A,B,C for 8wks treatment (uncompensated HF) (*A)
7. (n=17) TAC operated + drug A for 8wks treatment (uncompensated HF)
8. (n=17) TAC operated + drug A for 4wks treatment (compensated HF) (*C)
9. (n=17) TAC operated + drug B for 8wks treatment (uncompensated HF)
10. (n=17) TAC operated + drug B for 4wks treatment (compensated HF) (*C)
11. (n=17) TAC operated + drug C for 8wks treatment (uncompensated HF)
12. (n=17) TAC operated + drug C for 4wks treatment (compensated HF) (*C)

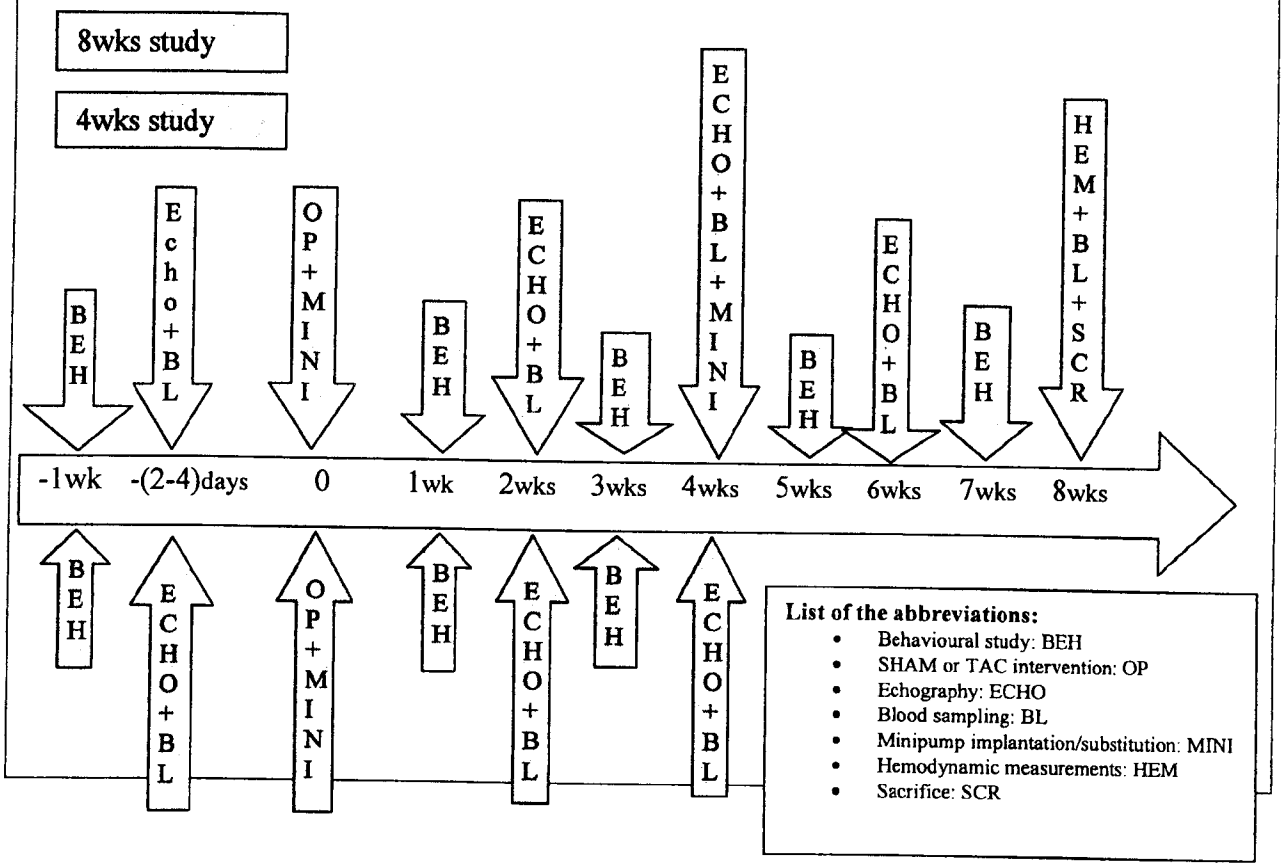
Total amount of animals: **216**
The motivation for these numbers is given in the paragraph below.

Experimental set up:
The entire experiment is divided in two stages. The first stage is necessary to check if the drugs do have beneficial effects on the pressure-overload model. We reasoned that if we do not see an effect in 8 weeks treated animals, i.e. the animals with the most overt uncompensated HF, then we would not conduct the 4 weeks treatment study (compensated HF). The latter groups are marked by (*C). Only in case that the drugs give positive results we will run the second stage with the other 72 mice (see the table below).
Animals will be treated with three different drugs (A, B and C). These are 3 compounds from different companies and unrelated from each other. These compounds have NOT been tested in any pathological cardiology model so we do not know anything about the potential efficacy. The compounds have been tested in other inflammatory diseases (see references). To rule out if the drugs do have a cardiac effect by themselves we need to test these drugs in sham-operated animals. The 3 drugs are dissolved in 3 different vehicles. We assume that the vehicles are without any effect. Therefore we propose to reduce the size of the n for these vehicles from 17 to 7 and pool the 3 groups to one (n=3*7=21) for the sham-operated as well as the TAC groups that need to be treated with the vehicle. This is indicated by (*A). In the worst case scenario that any of the vehicles does cause an effect we will ask permission for additional animals.
For drug A, we will use a dose of solubilized in vehicle A = *hydroxypropyl-beta-cyclodextrin (HPβCD)* and we plan to administer it s.c. via osmotic minipump (Alzet pump model 2004 s.c. implanted) to get a steady state concentration
study of Pradelli et al. (see ref. 12). After four weeks the minipump will be substituted with a new one (SOP:
The dose, route of administration and optimal vehicle to be used in case of drug B and C treatment are still under discussion with the respective pharmaceutical companies. We prefer and assume that the mice can be treated using the same osmotic minipump as used for drug A. If there will be a deviation in this regard we will notify the DEC and art. 14 responsible and adjust this protocol.
Treatment starts at the same time the TAC is implemented. Cardiac function and tissue concentrations of markers will be analyzed at 8 TAC. The details of the experimental protocol are listed below. It is similar as the previous protocol (DEC 2009-107) but now treatment is added.

As can be seen in the timeline we will measure baseline cardiac function by echo at 2-4 days before the TAC or sham operation (t=-3). In each animal additional ultrasound recordings will be performed after about 2, 4, 6, 8 weeks after the initial operation. As specified in the previous DEC we will, directly after the echo has been made, take a blood sample (by orbita punction) when the animal is still under isoflurane anesthesia. In these we will measure the inflammatory cytokines (see below). It is important to notice that we take blood samples 14 days after implantation of the minipumps and surgery. The study is not focused at detecting inflammatory parameters directly after surgical interventions, therefore we will not take blood at earlier timepoints. Moreover we included controls that undergo the same procedure so we can account for the potential disturbing surgical factor. At the day of sacrifice (after 8wks, or when animals display signs of severe HF), cardiac function will be determined by invasive hemodynamic measurements just before the animals are sacrificed and tissues will be then harvested. During the course of the experiments, all mice will be screened frequently for signs of HF, such as a reduced body weight of >20% (cachexia), increased respiratory rate, obvious lack of physical activity, edema in the legs and a poor fur. To detect if the therapeutic intervention influences physical activity, all mice will be placed in an open-field system for 30 min. similarly as performed in the previous study DEC2009-107. During these 30 min, locomotion will be detected by a camera and analyzed by specific software. The idea is that HF influences physical and mental health and that animals with HF show a decreased locomotion. This test will be performed every 2 weeks.

To characterize the expression and release pattern of pro- and anti-inflammatory cytokines during the development of HF (before other symptoms of HF are present), myocardial and plasma levels of pro- and anti-inflammatory cytokines will be measured at various time points (2, 3, 4, 6, 8 weeks after sham or TAC operation). In each blood sample the cytokines will be assayed by Proteome Profiler™ and verified by ELISA assays.

The circulating and tissue amounts of drug A, B and C will be determined by high-performance liquid chromatography (HPLC) in plasma and heart at the end of the study.



	Group	Animal amount	Drug treatment	Intervening echo + blood sampling	Hemodynamic measurement + blood sampling + sacrifice	Minipump
Experiment: *(C57BL/6J)*	***CONTROLS***					
	Sham mice					
	Group 1:	21	Vehicle A, B, C	2,4,6 weeks	8 weeks	Every 4 wks
	Group 2:	21	Vehicle A, B, C	2,3 weeks	4 weeks	The day of surgery
	SHAM mice (only drug)					
	Group 3:	17	A	2,4,6 weeks	8 weeks	Every 4 wks
	Group 4:	17	B	2,4,6 weeks	8 weeks	Not yet known
	Group 5:	17	C	2,4,6 weeks	8 weeks	Not yet known
	INQUIRY					
	TAC + drugs vehicle					
	Group 6:	21	Vehicle A, B, C	2,4,6 weeks	8 weeks	Every 4wks for drug A
	TAC + drug A					
	Group 7:	17	Drug A	2,4,6 weeks	8 weeks	Every 4 wks
	Group 8:	17	Drug A	2,3 weeks	4 weeks	The day of surgery
	TAC + drug B					
	Group 9:	17	Drug B	2,4,6 weeks	8 weeks	Not yet known
	Group 10:	17	Drug B	2,3 weeks	4 weeks	Not yet known
	TAC + drug C					
	Group 11:	17	Drug C	2,4,6 weeks	8 weeks	Not yet known
	Group 12:	17	Drug C	2,3 weeks	4 weeks	Not yet known
Animals requested	Stage 1	144				
	Stage 2	72				
	Total (1+2)	**216**				

9. Experimentele condities
9a. Anesthesie

Isoflurane (3-4% for induction, 1.5-2.5% for maintenance) will be used for both aorta constriction, and echocardiography (SOP . To measure the hemodynamic parameters, urethane (2.5 mg/kg i.p.) will be used (SOP:] After the hemodynamic measurements, animals will be sacrificed by bleeding (still under anesthesia).
All mice will be anesthetized in order to measure baseline cardiac function with echo (at day -3), to induce TAC or sham operation (at day 0), to perform intervening echo's in combination with blood sampling (every 2 weeks) and finally to measure the hemodynamic parameters and sacrifice the animals. Group 2, 8, 10, 12 will receive 4x anesthesia (3x isoflurane, 1x urethane); group 1, 3, 4, 5, 6, 7, 9, 11 will receive 5x anesthesia (4x isoflurane, 1x urethane).

9b. Pijnbestrijding

The animals will be treated pre- and post-operatively with buprenorphine (Temgesic) 0.1 mg/kg s.c. (see SOPs). We'll give the analgesia to the animals also the next two days post operation.

9c. Euthanasie en Humane eindpunten

At the end of the experiment, cardiac function is assessed by terminal hemodynamic measurements (SOP . Animals are anesthetized by urethane and after the measurements a blood sample will be obtained. Then they will be sacrificed by bleeding.
If an animal shows any symptoms of severe HF like a reduced body weight of >20 % of body weight (cachexia), increased respiratory rate, obvious lack of physical activity, severe edema in the legs and a poor fur or skin complications due to the minipump, animals will be subjected to the terminal measurement protocol immediately. The animals will be inspected by the art 12 and art 9 person involved.

10a. Ongerief

- Echocardiography (including induction and recovery of anesthesia): 02
- Blood sampling (directly after echo, when the animal is still under anesthesia). We will try to collect 200 ul via the cheek of the mouse. If that does not work we will do the saphenous vene, and if that does not work we will do the retro-orbital approach. Our experience with the tail vein is that it is impossible to get 200 µl: 03
- Open-field test for physical activity (including handling and different environment for 30 min): 02
- SHAM (including induction and recovery of anesthesia, recovery after surgery at day 1): 04
- TAC (including induction and recovery of anesthesia, recovery after surgery at day 1, and the development of hypertension in week 2-12): 05
- Osmotic mini-pump implantation: 02
- Terminal hemodynamic measurements: 02

Nature of intervention	**Duration (of anesthesia)**	**Frequency**	**Discomfort**	
TAC	30-45 minutes	1 time	Code 05	
SHAM	30-45 minutes	1 time	Code 04	
Echo + blood sampling	15-20 minutes	3-5 times	Code 03	
Minipump implantation	Consecutive to TAC and after echo	Every 4 weeks. Not yet known for drug B and C	Code 02	
Terminal Measurement	45-60 minutes	1 time	Code 02	
Activity measurement	1 hour	2-4 times	Code 02	
Living with heart failure	Depending on the effect of TAC	Permanent after decompensation	Code 04	

In total this is classified as 05 for the TAC operated mice and 04 for the SHAM operated mice.

10b. Welzijnsevaluatie

Most of the animals will completely recover after the aorta constriction. Usually the first 24 hours are critical and the majority of animals die due to complications of acute heart failure. After 4 weeks the animals may develop cardiac hypertrophy and in the next 4 weeks they may proceed to HF. Possible symptoms of HF are: reduced body weight (cachexia), increased respiratory rate, obvious lack of physical activity, severe edema in the legs and a poor fur (raised hairs). If there are signs of severe HF, the animal will be sacrificed to prevent suffering. We expect that the discomfort is less in sham-operated TAC animals and therefore we have scored it with 04, hence the TAC animals are scored with 05. This difference is indicated for the various groups on the front page.

11. Verzorging en huisvesting

The CPV will house and care the animals. The surgery and echocardiography in combination with blood sampling will be performed at the department of , at room ... Animals will be kept in roon · 24 hours after the surgical interventions at about 28 degrees Celsius to improve the success of recovery. In case of emergency / questions on the condition of the animals we will contact the art. 14 functionaries.

12. Deskundigheid

The surgery and echocardiography will be performed by art. 12 technicians of the department and The behavioral tests, collection of blood, heart tissue, and other organs will be performed by the VO (art. 9).

13. Standard Operation Procedures (SOP)

SOPs

Relevante literatuur

1. Skavdahl M, Steenbergen C, Clark J, Myers P, Demianenko T, Mao L, Rockman HA, Korach KS, Murphy E: Estrogen receptor-beta mediates male-female differences in the development of pressure overload hypertrophy. Am J Physiol Heart Circ Physiol 2005;288:H469-476.
2. Weinberg EO, Thienelt CD, Katz SE, Bartunek J, Tajima M, Rohrbach S, Douglas PS, Lorell BH: Gender differences in molecular remodeling in pressure overload hypertrophy. J Am Coll Cardiol 1999;34:264-273.
3. Witt H, Schubert C, Jaekel J, Fliegner D, Penkalla A, Tiemann K, Stypmann J, Roepcke S, Brokat S, Mahmoodzadeh S, Brozova E, Davidson MM, Ruiz Noppinger P, Grohe C, Regitz-Zagrosek V: Sex-specific pathways in early cardiac response to pressure overload in mice. J Mol Med 2008;86:1013-1024.
4. Gerszten RE, Wang TJ: The search for new cardiovascular biomarkers. Nature 2008;451:949-952.
5. Barrick CJ, Rojas M, Schoonhoven R, Smyth SS, Threadgill DW: Cardiac response to pressure overload in 129S1/SvImJ and C57BL/6J mice: temporal- and background-dependent development of concentric left ventricular hypertrophy. Am J Physiol Heart Circ Physiol 2007;292:H2119-2130.
6. Bozkurt B, Mann DL, Deswal A: Biomarkers of inflammation in heart failure. Heart Fail Rev 2009.
7. Hancock, W. W., B. Lu, et al. (2000). "Requirement of the chemokine receptor CXCR3 for acute allograft rejection." J Exp Med **192**(10): 1515-20.
8. Yates, C. C., P. Krishna, et al. "Lack of CXC chemokine receptor 3 signaling leads to hypertrophic and hypercellular scarring." Am J Pathol **176**(4): 1743-55.
9. Walser, T. C., S. Rifat, et al. (2006). "Antagonism of CXCR3 inhibits lung metastasis in a murine model of metastatic breast cancer." Cancer Res **66**(15): 7701-7.
10. Tonn, G. R., S. G. Wong, et al. (2009). "An inhibitory metabolite leads to dose- and time-dependent pharmacokinetics of (R)-N-{1-[3-(4-ethoxy-phenyl)-4-oxo-3,4-dihydro-pyrido[2,3-d]pyrimidin-2-y l]-ethyl}-N-pyridin-3-yl-methyl-2-(4-trifluoromethoxy-phenyl)-acetamide (AMG 487) in human subjects after multiple dosing." Drug Metab Dispos **37**(3): 502-13.
11. Thoma, G., R. Baenteli, et al. "Special ergolines efficiently inhibit the chemokine receptor CXCR3 in blood." Bioorg Med Chem Lett 21(16): 4745-9.
12. Pradelli, E., B. Karimdjee-Soilihi, et al. (2009). "Antagonism of chemokine receptor CXCR3 inhibits osteosarcoma metastasis to lungs." Int J Cancer 125(11): 2586-94.

University Maastricht

Faculty of Health, Medicine
and Life Sciences

Dierexperimenten Commissie

DEC

Aan:

voorzitter
p/a Secretariaat DEC-UM
Postbus 616
NL-6200 MD Maastricht
Telefoon:

Uw referentie: Onze referentie : Maastricht, 28-09-2011

Geachte Onderzoeker,

Uw projectaanvraag: "*A new treatment for the prevention of HF following cardiac hypertrophy: evaluation of two different drugs*", is op de DEC vergadering van 23 september 2011 besproken.

De DEC heeft een aantal vragen en opmerkingen:

- De duur van de proef is niet ingevuld op het voorblad (dit is de langste periode binnen één project dat één dier in proef is). De DEC verzoekt dit aan te passen.
- De DEC verzoekt de GGO medewerker van het voorblad te verwijderen. GGO is niet van toepassing op dit protocol (er wordt niet gewerkt met genetisch gemodificeerde dieren).
- Het ongerief op het voorblad en bij punt 10a stemmen niet overeen. De DEC verzoekt dit in overeenstemming te brengen.
- De DEC verzoekt bij punt 4 het woord "ernstig ongerief" toe te voegen.
- De DEC verzoekt de orbitapunctie te vervangen door een andere wijze van bloedafname.
- De DEC verzoekt de shamgroepen apart te vermelden bij punt 10a en het leven met hartfalen ook in de tabel te vermelden en tevens de bolletjes te verwerken in de tabel en het totale ongerief **per** groep te sommeren.

Gelieve eventuele vragen te beantwoorden in **een brief en indien noodzakelijk Uw project aan te passen en duidelijk de aanpassingen grijs te markeren.**

Uw project staat bij de DEC geregistreerd onder nummer **2011-123**, gelieve dit nummer in verdere correspondentie te vermelden.

Hoogachtend,

Voorzitter DEC-UM

University Maastricht

Faculty of Health, Medicine and Life Sciences

Dierexperimenten Commissie

DEC

Aan:

, voorzitter
p/a Secretariaat DEC-UM
Postbus 616
NL-6200 MD Maastricht
Telefoon:

Uw referentie: Onze referentie : Maastricht, 28-09-2011

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- De DEC verzoekt de GGO medewerker van het voorblad te verwijderen. GGO is niet van toepassing op dit protocol (er wordt niet gewerkt met genetisch gemodificeerde dieren). Indeed, I removed it.
- Het ongerief op het voorblad en bij punt 10a stemmen niet overeen. De DEC verzoekt dit in overeenstemming te brengen. The SHAM operation has been addressed as Code 04. As you pointed out, the animals having a SHAM operation have a less severe discomfort.
- De DEC verzoekt bij punt 4 het woord "ernstig ongerief" toe te voegen. It has been addressed that the animals receive a "severe intervention"
- De DEC verzoekt de orbitapunctie te vervangen door een andere wijze van bloedafname. We will then try to collect blood via the cheek of the mouse. If that does not work we will do the saphenous vene, and if that does not work we will do the retro-orbital approach. Our experience with the tail vein is that it is impossible to get 200 µl (as needed)
- De DEC verzoekt de shamgroepen apart te vermelden bij punt 10a en het leven met hartfalen ook in de tabel te vermelden en tevens de bolletjes te verwerken in de tabel en het totale ongerief per groep te sommeren. The table as the bullets have been up-dated according to your request.

Gelieve eventuele vragen te beantwoorden in **een brief en indien noodzakelijk Uw project aan te passen en duidelijk de aanpassingen grijs te markeren.**

Uw project staat bij de DEC geregistreerd onder nummer **2011-123**, gelieve dit nummer in verdere correspondentie te vermelden.

Hoogachtend,

Voorzitter DEC-UM

Maastricht University

Faculty of Health, Medicine and Life Sciences

Aan:

Ons kenmerk *Doorkiesnummer* *Maastricht*
27-10-2011

Project: *A new treatment for the prevention of HF following cardiac hypertrophy: evaluation of two different drugs.*

Verantwoordelijk onderzoeker (VO).

Namens de Vergunninghouder van de DEC-UM, delen wij u mede dat voornoemd project aan de ethische toetsingscriteria voor proefdiergebruik voldoet.
De DEC maakt geen bezwaar tegen uitvoering van dit project zoals aangevraagd en geeft een **positief advies met de opmerking dat de DEC u verzoekt de meest verfijnde methode voor bloedafname te gebruiken. Indien u daar niet voldoende in getraind bent, dient u zich hierin te bekwamen. Ref.** http://www.nc3rs.org.uk

Projectnummer:	**2011-123**
Diersoort:	**muis**
Aantal dieren:	**216**
Einddatum:	**26-10-2015**

Uw project staat bij de DEC en CPV geregistreerd onder bovenstaand nummer. Gelieve dieren, die voor dit project bestemd zijn, ook onder dit nummer aan te vragen.

Voorzitter DEC-UM

Vicevoorzitter DEC-UM

DEC-UM
Voorzitter DEC-UM

p/a secretariaat DEC-UM

Secretariaat DEC-UM

Bezoekadres

Postadres
Postbus 616
6200 MD Maastricht