Translational failure of anti-inflammatory compounds for myocardial infarction: a meta-analysis of large animal models

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Aims
Numerous anti-inflammatory drugs have been tested in large animal studies of myocardial infarction (MI). Despite positive results, translation of anti-inflammatory strategies into clinical practice has proved to be difficult. Critical disparities between preclinical and clinical study design that influence efficacy may partly be responsible for this translational failure. The aim of the present systematic review was to better understand which factors underlie the failure of transition towards the clinic.

Methods and results
Meta-analysis and regression of large animal studies were performed to identify sources that influenced effect size of anti-inflammatory compounds in large animal models of MI. We included 183 studies, containing 3331 large animals. Infarct size (IS) as a ratio of the area at risk (12.7%; 95% confidence interval, CI 11.1–14.4%, \( P = 0.001 \)) and IS as a ratio of the left ventricle (3.9%; 95% CI 3.1–4.7%, \( P = 0.001 \)) were reduced in treatment compared with control groups. Effect size was higher when outcome was assessed early after MI (\( P = 0.013 \)) and where studies included only male animals (\( P = 0.001 \)). Mortality in treated animals was higher in studies that blinded the investigator during the experiment (\( P = 0.041 \)) and depended on the type of drug used (\( P < 0.001 \)).

Conclusions
As expected, treatment with anti-inflammatory drugs leads to smaller infarct size in large animal MI models. Timing of outcome assessment, sex, and study quality are significantly associated with outcome and may explain part of the translational failure in clinical settings. Effect size depends on the type of drug used, enabling identification of compounds for future clinical testing.

Keywords
Myocardial infarction • Large animal model • Anti-inflammatory • Infarct size reduction • Translational failure

1. Introduction
Myocardial infarction (MI) and its consequences remain one of the greatest burdens of disease worldwide.¹,² Optimized medical care has resulted in reduced acute mortality, but patients surviving MI often develop diminished cardiac function and heart failure (HF).³

The progression from acute MI to chronic HF occurs through adverse remodelling, a complex mechanism involving infarct expansion, myocardial scar thinning, and left ventricular geometrical adaptation.⁴ Accumulating evidence indicates that adverse remodelling arises from an exaggerated inflammatory response that is initiated during ischaemia and early reperfusion.⁵,⁶

The damaged myocardium attracts inflammatory cells towards the site of injury where these cells secrete cytokines, proteases, and oxygen free radicals. This results in degradation of the extracellular matrix and clearing of necrotic cardiac resident cells, enabling scar formation in a later phase of cardiac wound healing.⁷ This inflammatory effect is essential to stabilize the scar tissue.⁸ However, it is outweighed by

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the acute effects of inflammatory cells on infarct expansion and cardiac function worsening early after ischaemia in the reperfusion phase.9–11 Indeed, multiple clinical studies have shown that elevated numbers of circulating neutrophils, monocytes, and cytokines in the acute setting are associated with adverse remodelling, the development of HF, and a worse overall prognosis.12–17 Attenuation of this inflammatory response is therefore a promising strategy to limit infarct size and preserve cardiac function post-MI.

Development of such anti-inflammatory strategies and transition from bench to bedside requires their testing in clinically relevant animal models.18 Both temporal and spatial development of tissue damage post-MI, along with inflammation-related signalling pathways, differ in small animals compared with larger mammals, including humans.19–21 Large animal models more closely resemble human anatomy, haemodynamics and pharmacodynamics and enable clinical treatment regimens, delivery routes, and functional read-outs.22–24 Since the early 1970s, many anti-inflammatory compounds have been shown to have efficacy in reducing reperfusion damage and post-MI remodelling in large animal models.25 However, none have proved successful in clinical trials and entered routine clinical practice.26,27 This lack of success may originate from discrepancies between large animal studies and clinical trials. These discrepancies could be attributed to an insufficient similarity between large animal models and MI patients.28 Patients often have co-morbidities, such as smoking, diabetes, and hypertension, while large animal models lack these characteristics due to increased costs and experimental complexity.19,24 This may lead to a difference in mechanisms between the post-MI inflammatory responses in humans compared with large mammals.

Perhaps of equal importance are the possible presence publication bias and the experimental study design of these large animal studies. Previous meta-analyses in different fields of preclinical research show that publication bias and methodological flaws are often responsible for systematic effect size overestimation and thereby incorrect conclusions about efficacy.28–30 Regarding the translational failure of anti-inflammatory compounds for MI, it is however, unclear which factors underlie this phenomenon.

The aim of this study was therefore to identify factors that influence the efficacy of anti-inflammatory compounds in large animal studies of MI and thereby hamper the transition of anti-inflammatory compounds towards clinical application. To this extent, we performed a meta-analysis in which we assessed the overall effect of anti-inflammatory compounds in large animal MI studies. Through meta-regression, we then aimed to identify methodological factors that were of influence on outcome.31 Additionally, meta-regression allowed us to identify differences in the efficacy of different anti-inflammatory drug classes in these large animal studies.

2. Methods

The protocol for this systematic review and meta-analysis has been published.32 We searched PubMed and Embase on 1 May 2014 for studies using anti-inflammatory compounds in large animal models of MI. Our inclusion criteria were controlled study design and the use of large animal models (defined as pigs, dogs, sheep, or goats) of myocardial infarction (coronary obstruction >30 min). In the current study, we decided not to include rabbits given their average weight (<5.0 kg) and the inability to perform transluminal coronary interventions in these animal models. Anti-inflammatory compounds were defined as either having FDA approval for anti-inflammatory effects or as directly targeting pivotal inflammatory mechanisms.32 To exclusively investigate anti-inflammatory compounds, we chose to exclude interventions that have evident pleiotropic effects. Also, if the mechanism of action was unclear regardless of the effect on inflammatory parameters, these interventions were excluded. Finally, the treatment had to be solely pharmacological. Abstract publications were excluded, as were case reports and studies lacking a control group. There were no language restrictions. Studies were screened in two phases (title/abstract followed by full text) by two independent researchers (G.v.H. and S.J.). In case of disagreement, consensus was achieved by discussion in all cases. A summary of the research protocol has been added to the Supplementary material online, Method.

The primary outcome for meta-analysis was infarct size as percentage of the area at risk (IS/AAR). Secondary outcomes were infarct size as percentage of the left ventricle (IS/LV), mortality, left ventricular ejection fraction (EF), left ventricular end-diastolic volume (LVEDV), left ventricular systolic volume (LVESV), and wall thickness (WT) as ratio of the opposing wall. Data were extracted from included studies and added to the ‘Collaborative Approach to Meta-Analysis and Review of Animal Data from Experimental Studies’ (CAMARADES)—online international database.

2.1 Statistics

Due to the anticipated heterogeneity, we performed a random-effects meta-analysis. We used a raw difference in mean (RMD) analysis for the outcome parameters IS/AAR, IS/LV, EF, and WT. For mortality, we used odds ratios.33 Where individual studies reported outcome for several treatment groups (i.e. dose–response studies), the size of the control group was adjusted to reflect this, as previously described.31,33 To determine whether the observed results were confounded by publication bias, we used three approaches: visual inspection of funnel plots, Egger’s regression analysis for small study effects, and Tweedie and Duval’s trim and fill.34,35 Funnel plotting can be used to visually identify studies with small precision that overstate the effect of an intervention and are consistent with the presence of small study publication bias. Egger’s regression statistically assesses the presence of publication bias of small studies in a funnel plot by determining whether the regression line and its 95% CIs for precision vs. standardized effect size intersect at the origin of the graph. Trim and fill analysis non-parametrically attempts to correct for funnel plot asymmetry by identifying and imputing theoretical missing studies. This enables recalculation of the overall treatment effect in the absence of publication bias.32–35

We used meta-regression to explore heterogeneity and to test which parameters were significantly associated with outcome. We included 15 parameters beforehand as potential sources of heterogeneity that are relevant in either a clinical or translational perspective, provided that we had at least 10 times that number of experimental comparisons.33 We tested parameters in three separate categories: therapeutic characteristics (timing of therapy, timing of administration, and drug group), model characteristics [species, sex, surgical approach (open vs. closed chest), permanent occlusion vs. ischaemia/reperfusion (I/R), duration of ischaemia, location of coronary occlusion, and method infarct size quantification], and risk of bias (reporting of blinding of the operator, blinded outcome assessment, randomization, and language). Compounds were pooled in drug groups, based on working mechanism. We only performed meta-regression for parameter values reported in at least five independent groups for comparison.

2.1.1 Sensitivity analysis

Post hoc sensitivity analysis was performed on infarction model. Since inflammation is particularly an issue when the blood flow is re-established and since permanent occlusion models have low clinical relevance, we performed a sensitivity analysis by excluding all permanent occlusion models. Meta-analysis and meta-regression for all predetermined parameters were performed for the primary outcome IS/AAR and the secondary outcome IS/LV.
Meta-regression is more conservative than stratification of heterogeneity. We set the statistical threshold for our predefined primary outcome measure at \( P < 0.05 \), and for secondary outcome measures, we applied a Bonferroni correction to give a critical \( P \)-value of 0.017. All analyses were performed using Stata version 11 (StataCorp LP, TX, USA).

3. Results

We identified 2530 results in PubMed and 2778 in Embase. After merging and removal of duplicates, 4105 unique publications remained. We excluded 3570 studies in the first phase (title/abstract) of screening. In the next phase (full text screening), we included 183 publications (see Supplementary material online, Figure S1). These 183 publications reported 219 experimental comparisons for the primary outcome IS/AAR, reporting outcome from 3331 animals (1839 treated and 1492 control). All included studies reported the animal species, infarct location, ischaemia duration, timing of therapy, time of outcome assessment, and drug type. Of the 219 included study groups, permanent occlusion vs. I/R was unknown in 3, delivery route in 5, and surgical approach in 1, and sex of the animals was unknown in 45 groups. For sex, we included ‘not reported’ as a separate group in the meta-regression. Most experiments used both male and female animals (\( n = 101 \), male \( n = 60 \), not reported \( n = 45 \), female \( n = 13 \), and used dogs (\( n = 163 \); pig, \( n = 54 \); sheep, \( n = 2 \)). Included studies predominantly used occlusion of the left anterior descending coronary artery (140 experiments) rather than the circumflex coronary artery (75 experiments); 4 used both. All studies except for one (using cardiac MRI) used ex vivo methods for determining infarct size and area at risk. The majority of studies used tetrodotoxin staining (nito blue or triphenyl). All included studies reporting the primary outcome IS/LV and their main characteristics are provided in Supplementary material online, Table S1. References of all included 183 studies are included in the Supplementary material online.

Random allocation of animals, blinded outcome assessment, and blinding of the operator were reported in 143 (65%), 64 (29%), and 21 (10%) of the 219 comparisons, respectively. For the secondary outcomes, 88 studies reported IS/LV (138 experiments) and 97 reported mortality (143 experiments). LVEF was reported in 21 studies (31 experiments), and the analysis for WT included 11 studies (16 comparisons).

3.1 Meta-analysis

For the primary outcome, treatment led to an absolute reduction in the infarct size as a percentage of the area at risk of 12.7% (95% CI 11.1–14.4%, \( P < 0.001 \)) (Figure 1A). There was substantial heterogeneity between studies (\( I^2 = 80.9\% \), \( \tau^2 = 104.5 \)). Visual inspection of the funnel plot and Egger regression suggests publication bias of small studies (Figure 1C and D). This was confirmed by Tweedie and Duval’s trim and fill, with 12 imputed missing studies and a corrected efficacy of 11.4% (95% CI 9.6–13.1%) (Figure 1C).

Treatment with anti-inflammatory compounds was also associated with improved secondary outcome measurements. There was an absolute difference in mean IS/LV of 3.9% (95% CI 3.1–4.7%, \( P < 0.001 \)) in favour of treated animals (Figure 1B). EF was higher in treated animals compared with control animals (RMD 3.4, 95% CI 0.8–6.1, \( P < 0.001 \)). No difference in mortality was observed (OR 0.96, 95% CI 0.79–1.17, \( P = 1.0 \)). WT as a percentage of the opposing wall was lower in treated animals, but of note, this outcome was predominantly assessed in studies testing the effect of non-steroid, anti-inflammatory drugs (NSAIDs) (11 out of 16) (WT RMD 16.3% 95% CI 8.5–24.0%, \( P < 0.001 \)) (see Supplementary material online, Figure S2). Only a small subset of studies reported LVEDV and LVESV, and the extent of the data set was too small to enable meta-analysis.

3.2 Meta-regression

We sought possible sources of heterogeneity using meta-regressions with effect on IS/AAR, IS/LV, and mortality as the dependent variable. The number of comparisons reporting EF and WT was insufficient for meta-regression and comparison of individual study characteristics. Classification of drug groups is provided in Supplementary material online, Table S2. Drug class was a predictor of efficacy for IS/AAR and IS/LV endpoints (\( P < 0.001 \), \( P = 0.002 \)) but not for mortality, although the point estimates for different drug classes were similar (Figure 2). For all three outcome measures, leukotriene inhibitors and cell adhesion molecule (CAM) inhibitors performed best, showing greater reduction in infarct size and lower mortality. The effect of anti-inflammatory compounds in improving IS/AAR and IS/LV was less apparent at later times of outcome assessment, both when measured in a categorized fashion (\( P = 0.013 \) IS/AAR, \( P = 0.001 \) IS/LV (Figure 3A and B) and on a continuous scale (\( P = 0.016 \) IS/AAR, \( P = 0.007 \) IS/LV). Furthermore, the sex of the animals appeared to be important; for both IS/AAR and IS/LV, greater effects were seen in experiments using male animals than in those using female animals or a mixed population (IS/AAR \( P = 0.002 \); IS/LV \( P = 0.002 \)) (Figure 4). Additionally, the effect on IS/LV was larger in temporary occlusion models compared with permanent occlusion models (\( P = 0.014 \)) (Figure 5A).

Measures taken to preserve the internal validity of studies influenced reported outcomes. When outcome assessment was performed in a blinded fashion, efficacy for IS/AAR tended to be higher compared with non-blinded studies (\( P = 0.075 \)). Blinding of the operator performing the intervention was associated with increased mortality in the treatment group (\( P = 0.041 \)) (Figure 5B and C).

To assess whether sources of heterogeneity could also be identified within specific drug groups, we performed meta-regression for sex and time of assessment for the primary outcome IS/AAR for each of the four largest drug groups [NSAIDs (\( n = 29 \)), CAM inhibitors (\( n = 27 \)), leukotriene inhibitors (\( n = 32 \)), and reactive oxygen species (ROS) scavengers (\( n = 98 \))] (see Supplementary material online, Table S2 for classification). For time of assessment, results similar to those found for the complete data set were observed, but only had sufficient power in the subgroup of ROS scavengers to reach statistical significance (\( P = 0.015 \)) (see Supplementary material online, Figure S3).

3.2.1 Sensitivity analysis

After excluding all permanent occlusion models, anti-inflammatory treatment was associated with a significantly decreased IS/AAR of 12.4% (10.8–14.2, \( P < 0.001 \), \( n = 197 \)) and IS/LV of 4.5% (3.4–5.6, \( P < 0.0001 \), \( n = 90 \)). Despite the decreased number of studies, the 95% CI of the reduction in IS/LV narrows, suggesting a more pronounced effect in I/R models compared with the complete data set (Figure 6).

Meta-regression revealed that the same parameters have significant influence on the effect size in I/R models alone compared with the complete data set (i.e. gender, drug group, and timing of outcome assessment). The only additional predictor for the effect on IS/AAR was timing of therapy (\( P = 0.01 \)), where the effect was most pronounced when therapy was administered within 1 h after occlusion (see Supplementary material online, Figure S3).
4. Discussion

4.1 Efficacy of anti-inflammatory compounds

Both clinical and preclinical studies suggest that the post-MI inflammatory response orchestrates cardiac wound healing and is accountable for infarct expansion.\textsuperscript{17,36} Our systematic review and meta-analysis of data from >180 large animal studies, including over 3300 animals subjected to MI, confirms that treatment with anti-inflammatory compounds reduces infarct size in these animals.

Our meta-regression suggests that inhibition of different inflammatory pathways has different effects on infarct size, which can be useful...
for the identification of candidate compounds. The current study implies that treatment with, for instance, leukotriene inhibitors is more effective than NSAIDS or immunosuppressive drugs. To our knowledge, this is the first study that allows for direct comparison of different anti-inflammatory drug groups. It shows that the acute anti-inflammatory response is detrimental to cardiac function, and inhibition of this response leads to a preservation of cardiac function in large animal MI studies. However, profound downstream immunosuppression by corticosteroid administration is not as effective as partial suppression by sole attenuation of oxygen free radical or leukotriene signalling. Likely, the influx of inflammatory cells is required to clear out damaged tissue and enable cardiac wound healing during the late-phase post-MI, and complete abolishment of the inflammatory response does therefore not lead to beneficial effects post-MI.37 One of the major differences between large animal models and patients could be that the presence of inflammatory cells in cardiac tissue after MI is of major importance in patients, while scar formation in large animals is less dependent on these inflammatory cells. The inflammatory response should hence be ‘fine-tuned’ rather than completely abolished in patients. To a similar extent, this could also apply to the effect of timing of therapy on effect size, since inflammatory cells are needed during the proliferation and maturation phase of cardiac wound healing. Attenuating inflammation during these phases could, depending on the inhibited mechanism, therefore also be less effective. Additionally, most of the included studies in our meta-analysis used ischaemia–reperfusion models. Since anti-inflammatory therapies given immediately after

Figure 2 Meta-regression of drug group. Effect size per drug group for the primary outcome IS/AAR (A), secondary outcomes IS/LV (B), and mortality (C). Dots and vertical lines represent the mean and 95% CI per drug group; horizontal grey bar represents the 95% CI of the mean effect size. Dotted line represents no effect. IS/AAR, infarct size as percentage of area at risk; IS/LV, infarct size as percentage of the left ventricle; n, number of groups for comparison.

Figure 3 Meta-regression of timing of therapy. Effect size per timing interval after occlusion of the coronary artery, for the primary outcome IS/AAR (A) and secondary outcome IS/LV (B). IS/AAR, infarct size as percentage of area at risk; IS/LV, infarct size as percentage of the left ventricle. Dots and vertical lines represent the mean and 95% CI per group; horizontal grey bar represents the 95% CI of the mean effect size. Dotted line represents no effect. n, number of groups for comparison.
Figure 4 Meta-regression of sex. Effect size per sex for the primary outcome IS/AAR (A) and secondary outcome IS/LV (B). IS/AAR, infarct size as percentage of area at risk; IS/LV, infarct size as percentage of the left ventricle. Dots and vertical lines represent the mean and 95% CI per group; horizontal grey bar represents the 95% CI of the mean effect size. Dotted line represents no effect. n, number of groups for comparison.

Figure 5 Meta-regression of infarction model and blinded outcome assessment. Meta-regression of model of infarct creation (permanent vs. temporary occlusion of the coronary artery) on the secondary outcome IS/LV (A). Meta-regression of the effect of blinded vs. non-blinded outcome assessment on the primary outcome IS/AAR (B) and secondary outcome mortality (C). IS/AAR, infarct size as percentage of area at risk; IS/LV, infarct size as percentage of left ventricle. Dots and vertical lines represent the mean and 95% CI per group; horizontal grey bar represents the 95% CI of the mean effect size. Dotted line represents no effect. n, number of groups for comparison.
reperfusion are directly targeted at reperfusion injury in these models, a possible explanation for the discrepancy of efficacy of anti-inflammatory compounds in animals vs. patients could be that reperfusion injury does not play a critical role in human subjects.

### 4.2 Clinical translation and preclinical study design

Although comparison of the effect of different drug groups may be useful to select agents for future clinical testing, none of the compounds that have been tested in clinical trials have been implemented in daily practice.

A major reason for this translational failure may be the presence of publication bias, which is of concern since previous reports have shown that the effect of compounds in animal research may be overestimated due to under-reporting of small, neutral studies. Our analysis also presents evidence of small study bias, and after correction for this bias, the overall effect of anti-inflammatory compounds is estimated to be 10% lower than the observed effect.

As presented in this study, the presence of methodological flaws also contributes to the translational failure from bench to bedside. In this perspective, both internal and external validity of preclinical studies are important factors. Lack of blinding of both the operator and outcome assessor poses threats to internal validity. Less than a third of the included studies in our systematic review reported blinding of treatment administration or outcome assessment. Moreover, we observed an effect of investigator blinding on outcome in the present study. This underlines the importance of high methodological quality to ensure internal validity.

In the perspective of external validity in large animal research, studies should resemble the clinical situation as closely as possible with regard to study design to enable extrapolation of study outcome to the clinical situation. Some of the disparities are inherent with the use of animals for modelling human disease, like the artificial induction of MI in animal models vs. the gradual process of MI by coronary stenosis and plaque rupture in patients. However, many methodological factors can be controlled for and efforts should be made to optimize preclinical study design and to prevent avoidable discrepancies between preclinical and clinical research.

Furthermore, differences between animal species must be acknowledged. The presence of an evident collateral coronary circulation in dogs compared with pigs must be accounted for when therapies for ischaemia–reperfusion injury are tested. In our analysis, the majority of animals were dogs, and animal species did not have an effect on the effect size.

Our study shows that these aspects influence outcome, since infarct size reduction in the included large animal studies is highest during the first hours post-MI, and this effect is attenuated when the time of assessment is extended. Any effect limited to the first hours post-MI only is of no clinical relevance and fails to be detected in clinical trials given their long-term follow-up. One important explanation of this observation might be the effect of anti-inflammatory compounds on myocardial oedema. First of all, myocardial oedema arises in the first hours after MI and leukotriene inhibitors, have been shown to be involved in reducing tissue oedema. Since the effect is strongest during the first hours after MI and leukotriene inhibitors seem to be most effective, the observed effect might partially be caused by the effect of anti-inflammatory treatments on oedema reduction. This phenomenon is only temporary and could be considered as a major confounder that directly influences translational success.

Similarly, we observed sex to be a significant predictor of outcome. Since most studies used both male and female animals, translational failure is not necessarily attributable to the difference in effect size. However, the more pronounced effect in studies, using male animals only, emphasizes the importance of external validity and limits the generalizability to female patients. Although the exact mechanism behind this phenomenon remains unidentified, differences between male and female subjects regarding outcome after MI have been observed in clinical and preclinical studies. One explanation of this difference in effect size between male and female animals could be the direct anti-inflammatory effect of estrogens, thereby diminishing the effect of anti-inflammatory compounds after MI in female subjects. To the best of our knowledge, this is the first study showing that attenuating the inflammatory response in male subjects is more efficacious compared with inhibiting this response in female subjects, a phenomenon that should be taken into account when designing large animal MI studies.

### 4.3 Meta-analysis of translational studies

Although our analysis shows significant effects of anti-inflammatory treatments on infarct size, it should be kept in mind that meta-analyses...
cannot reveal causal relationships. Hence, we do not intend to provide mechanistic insights with our study.

The strength of a meta-analysis depends on the strength and quality of included studies. For example, reporting of several functional outcomes in preclinical models varies between studies, disallowing pooling of these data. Also, reporting of mortality is considered to be less rigorous compared with clinical studies, which may introduce bias into the data set.

In contrast to meta-analyses of clinical studies, heterogeneity in preclinical studies can be of great additional value, since parameters responsible for heterogeneity can be used as predictors for success and can help design future clinical and preclinical studies. The extent of our data set was insufficient for a multivariate meta-regression analysis; therefore, we cannot completely rule out co-linearity of the different sources of heterogeneity (see Supplementary material online, Table S3). Furthermore, other clinically relevant parameters like open vs. closed chest experiments or the use of anesthesia (anesthetized vs. conscious animal models) either did not show to have a significant effect on outcome in our analysis or could not be tested due to statistical limitations (1 parameter for every 10 included study groups). Moreover, we chose not to include rabbits, as we did not consider these animals to be large animal MI models in the perspective of the current study given their size and the inability to perform transluminal coronary interventions in this animal species.

5. Conclusion

Our systematic review and meta-analysis of large animal MI studies showed reduced infarct size in animals treated with anti-inflammatory compounds compared with control animals. We found methodological aspects, like time of outcome assessment, sex of the animals, and blinding of the operator to be significant predictors of effect size. We, therefore, stress the use of clinically relevant animal models and study designs to increase the translational value of preclinical research.

Supplementary material

Supplementary material is available at Cardiovascular Research online.

Conflict of interest: none declared.

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References


8. van der Worp HB, Macleod MR. Preclinical studies of human disease: time to take myofiber


10. van der Worp HB, Macleod MR. Preclinical studies of human disease: time to take myofiber


