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Angiogenic effects of sequential release of VEGF-A_{165} and PDGF-BB with alginate hydrogels after myocardial infarction

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Abstract

Objective: This study investigates whether local sequential delivery of vascular endothelial growth factor-A_{165} (VEGF-A_{165}) followed by platelet-derived growth factor-BB (PDGF-BB) with alginate hydrogels could induce an angiogenic effect and functional improvement greater than single factors after myocardial infarction.
Methods: Alginate hydrogels were prepared by combining high and low molecular weight alginate. Growth factor release rates were monitored over time *in vitro* with ^{125}I-labelled VEGF-A_{165} and PDGF-BB included in the gels. One week after myocardial infarction was induced in Fisher rats, gels with VEGF-A_{165}, PDGF-BB, or both were given intra-myocardially along the border of the myocardial infarction. Vessel density was analysed in hearts and cardiac function was determined by Tissue Doppler Echocardiography. In addition, the angiogenic effect of sequenced delivery was studied *in vitro* in aortic rings from C57B1/6 mice.
Results: Alginate gels were capable of delivering VEGF-A_{165} and PDGF-BB in a sustainable manner, and PDGF-BB was released more slowly than VEGF-A_{165}. Sequential growth factor administration led to a higher density of α-actin positive vessels than single factors, whereas no further increment was found in capillary density. Sequential protein delivery increased the systolic velocity-time integral and displayed a superior effect than single factors. In the aortic ring model, sequential delivery led to a higher angiogenic effect than single factor administration.
Conclusions: The alginate hydrogel is an effective and promising injectable delivery system in a myocardial infarction model. Sequential growth factor delivery of VEGF-A_{165} and PDGF-BB induces mature vessels and improves cardiac function more than each factor singly. This may indicate clinical utility.

Keywords: Angiogenesis; Growth factors; Tissue engineering; Ventricular function; Infarction

1. Introduction

Therapeutic angiogenesis induced by growth factors is a promising treatment for chronic myocardial ischemia after myocardial infarction and associated remodelling [1]. Because growth factors administered as proteins are rapidly

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broken down and their systemic spread is undesirable, the focus more recently has been on local gene delivery to achieve longer tissue exposure [2]. However, angiogenesis is a complex, multi-step process, and distinct factors are involved in each step [3]. Therefore, delivery of multiple factors may be required to increase the efficacy of angiogenesis, but simultaneous delivery of these factors, whether via recombinant proteins or gene therapy approaches, may not be the optimal approach. A better therapeutic angiogenic efficacy may be attainable with a combination of growth factors delivered by biodegradable polymer, which achieves distinct release kinetics for each factor. In particular, VEGF is an important initiator of angiogenesis [3,4], but delivery of VEGF alone may lead to immature and leaky vasculature with poor function [4]. PDGF-BB stimulates smooth muscle cell recruitment to newly formed vessels [5–7] and is partially responsible for enhancement of vessel functionality with maturation. In therapeutic angiogenesis, it may be useful to deliver VEGF before PDGF-BB to first stimulate vessel formation and then smooth muscle cell investment of the capillaries. This possibility is supported by a previous report where sequential slow release of VEGF-A_{165} and PDGF-BB with a poly-lactide-co-glycolide (PLG) polymer scaffold induced higherdensities of mature vessels than VEGF delivery alone [8]. However, placement of that polymer form requires incision into the tissue [9], which is difficult to apply to the heart. In this study, we used injectable hydrogels to deliver a sequence of recombinant VEGF-A_{165} and PDGF-BB. We hypothesized that sequential delivery of VEGF-A_{165} followed by PDGF-BB would induce similar vessel densities as using VEGF-A_{165} by itself, but that the sequential delivery strategy would produce greater maturation and function of the networks than would result from VEGF delivery alone in a myocardial infarction model.

2. Materials and methods

2.1. Alginate modification

Ultra pure alginates were obtained from ProNova Biomedical (Norway). High guluronic acid content alginate was used as high molecular weight alginate (HMW, 250,000 Da). Low molecular weight alginate (LMW, 5000 Da) was obtained by gamma (γ)-irradiating HMW with a cobalt-60 source for 4 h at a γ-dose of 5.0 Mrad, as specified by Kong et al. [10]. Both alginates were reconstituted in ddH_2O (1% w/v), and 1% of the alginate sugar residues were oxidized with sodium periodate (Aldrich) by maintaining solutions in the dark for 17 h at room temperature to enable hydrolytic degradation of the polymer as previously described (reference 11). The oxidation reaction was stopped by adding an equimolar amount of ethylene glycol (Fisher), and the solution was subsequently dialyzed (MWCO 1000, Spectra/Por®), filtered, and lyophilized as described by Bouhadir et al. [11].

2.2. Gel formulation and growth factor incorporation

Modified HMW and LMW alginates were reconstituted in phosphate-buffered solution (Invitrogen, PBS solution with 0.1 g/l of $MgCl_2 \cdot 6H_2O$ and 0.132 g/l of $CaCl_2 \cdot 2H_2O$) to obtain 2% w/v solutions. The resulting solutions were mixed (75% LMW; 25% HMW used in all experiments) and combined with recombinant human VEGF-A_{165} (R&D), PDGF-BB (R&D) or both growth factors by using two syringes coupled by a syringe connector. The resulting alginate solution was cross-linked with aqueous slurries of a calcium sulphate solution (0.21 g $CaSO_4$/ml ddH_2O) in a ratio of 25:1 (40 μl of $CaSO_4$ per 1 ml of 2% w/v alginate solution). Entrapment of air bubbles was prevented during the mixing process. The mixture was allowed to gel for 30 min at room temperature and then maintained at 4 °C before use. The final concentration of the growth factor was 3 μg PDGF-BB and/or 3 μg VEGF-A_{165} per 100-μl alginate gel.

2.3. Release kinetics

^{125}I-VEGF-A_{165} and ^{125}I-PDGF-BB were purchased from PerkinElmer Life Sciences. Binary alginate gels containing radio-labelled growth factors were prepared with the procedure described above. The resulting mixture was then cast between two glass plates separated with 1-mm spacers and allowed to gel for 30 min. The gels were divided into four samples and subsequently incubated in 3-ml of PBS buffer solution at 37 °C. At each experimental time point, the radio-labelled growth factors present in the extracted buffer solution were measured using a gamma counter-1470 WIZARD (PerkinElmer, Life Sciences) and normalized to the initial total radio-labelled growth factor incorporated into samples.

2.4. Alginate degradation detection in vitro

The alginate hydrogels were ionically cross-linked to create gels. After the gelling process, gels were incubated in 1.0 ml of PBS buffer solution at 37 °C. Gel degradation behaviour was monitored by measuring the dry weight loss over time. At least four specimens at each time point were used to obtain the degradation curve. Experiments were done under sterile conditions to prevent bacterial and fungal contamination.

2.5. Animal experiments and design

All procedures involving the use and care of animals conform with the "Guide for the Care and Use of Laboratory Animals" published by the US National Institutes of Health (NIH Publication No. 85-23, revised 1996) and were approved by Stockholm Southern Ethics Review Board.

2.6. Myocardial infarction model and in vivo protein-alginate delivery

Fig. 1 illustrates the experimental procedure. Male Fisher rats (200–250 g) were anaesthetized with midazolam (5 mg/kg) and medetomidine hydrochloride (0.1 mg/kg), intubated and ventilated. The heart was exposed via thoracotomy through the fourth intercostal space. The left anterior descending coronary artery (LAD) was identified and ligated below the atrium. Pallor and regional wall motion abnormality of the left ventricle confirmed occlusion. There was animal loss after LAD ligation. Around 20% of the rats were dead after 24–48 h of infarction. Baseline echocardiography with Tissue Doppler Velocity Imaging (TVI) was done 4 days after myocardial infarction. Rats with visual infarct size below 20% at the papillary level in the short axis view were excluded to minimize the variation of infarct size. Thus only approximately 60% of the animals were used in the experiment. Although experimental rats had large myocardial infarctions with cardiac cavity dilation over time, they survived with negligible loss during the 28 days of the experiment. Seven days after LAD ligation the rats were randomized for treatment and the chest reopened. A suture was applied in the middle area of the myocardial infarction to make a landmark for the injection. Alginate with 3 μg VEGF-A_{165}, 3 μg PDGF-BB, or 3 μg of both proteins, in a total volume of 100 μl was injected intramyocardially in one spot in the peri-infarct region along the border of the myocardial infarction at the same level as the suture. The doses were based on a previous study with the ischemic limb model [12]. Alginate hydrogel blank and saline were injected as controls. Twenty-eight days after the treatment, echocardiography with TVI was performed and the animals were subsequently euthanized with carbon dioxide and their hearts collected for histology. There were 6 or 7 animals in each group.

2.7. Alginate observation in the heart tissue

After the hearts were frozen in OTC compound, frozen sections were prepared. Hematoxylin–eosin staining was performed to visualize overall tissue structure and remaining alginate gel in the myocardium.

2.8. Vessel densities

For the analysis of capillary density, frozen sections were incubated with Griffonia Badeiraea Simplicifolia Isolectin B4 (GSL-I-B4, Vector Laboratories), followed by a second incubation with ABC Complex. Capillaries were visualized by DAB with supplementation of 0.03% hydrogen peroxide. The capillaries were counted from images obtained at a magnification of 400× taken with a LCD camera (Olympus, Japan) connected to a microscope. Photomicrographs of eight fields around the injection site were taken, and the capillary count was analyzed using a blinded procedure with an image analysis program (Micro Image, Olympus). For the analysis of density of vessels associated with smooth muscle cells, the lectin-stained sections were incubated with primary antibody against α-actin (Sigma). Rabbit anti-mouse secondary antibody (FITC, Dako) was used to visualize the positively stained blood vessels. Positively stained blood vessels around the injection site were counted under 200× magnification using a fluorescent microscope. All analyses were performed in a blinded manner.

2.9. Left ventricular function

Cardiac function was assessed by Echocardiography using a Vingmed Vivid 5 (Vingmed A/S, Norway) ultrasound system equipped with a 10 MHz transducer. Echocardiography was performed on the rats four days after ligation and four weeks after alginate injection. Tissue Doppler Echocardiography was performed to detect systolic velocity-time integral. Colour Tissue Velocity Images were collected at the apical chamber view at frame rates close to 230 fps. The probe position was adjusted under the guidance of Pulsed Tissue Doppler to the maximum velocity of the mitral valve plane. The recording was obtained by positioning a sample volume in each basal septal left ventricular wall segment. Analysis was performed off-line from acquired cine loops with Echopac (version 6.1). Peak systolic velocity was not measured because of the unfavourable noise to signal ratio. Systolic velocity-time integral, a parameter for displacement of myocardium, is less noise dependent and was assessed from 2–3 beats to minimize the variability. The final value of regional systolic myocardial function was calculated as a mean value from 2–3 peak velocity-time integral at systole.

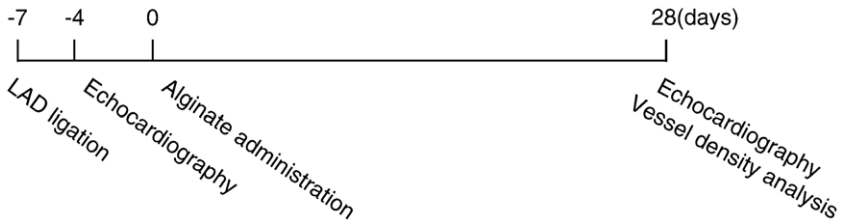

Fig. 1. Experimental design for the delivery of growth factors in alginate gel in a rat myocardial infarction model.

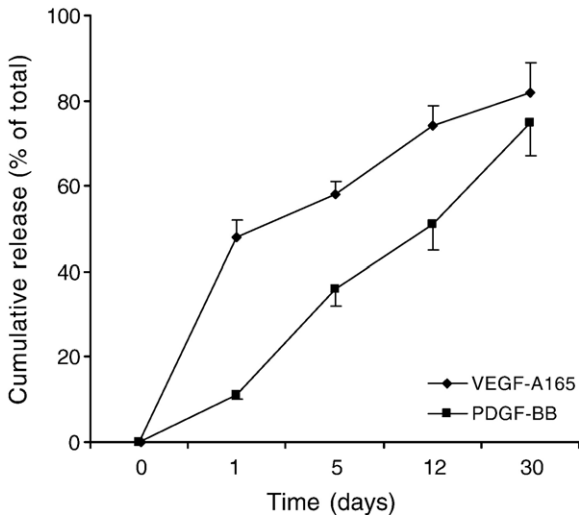

Fig. 2. The cumulative release of VEGF-A_{165} and PDGF-BB from alginate hydrogels *in vitro* following incubation in PBS at 37 °C. Values are given as mean±SEM, $n=4$ at each datapoint.

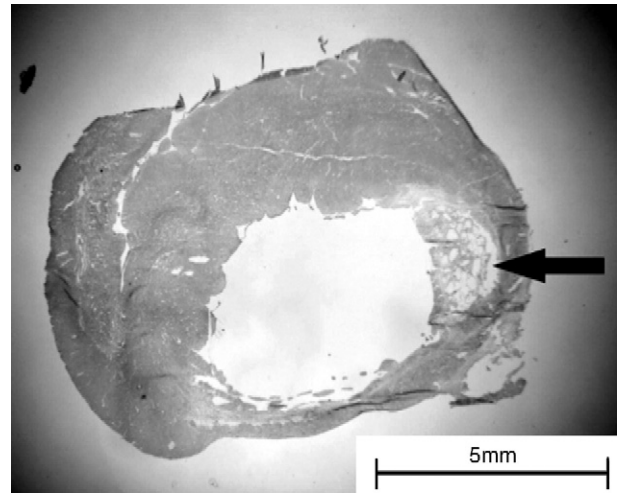

Fig. 4. Photomicrograph of hematoxylin and eosin-stained cryosection of the heart 4 weeks after delivery of hydrogel (arrow indicates residual gel fragments) into the periinfarction area. Residual hydrogel fragments were observed in 30% of the animals receiving gel injection.

For the measurement of left ventricular end-systolic dimension (LVDd) and ejection fraction the heart was imaged in the 2-D mode in the long axis view at the level of the largest left ventricular diameter.

2.10. Angiogenic effect of sequenced VEGF and PDGF-BB delivery on aortic rings

The thoracic aortas from mice (male C57B1/6 mice, 20–30 gm and 6–8 weeks old, B&K, Sweden) were removed and sectioned into 1-mm lengths of aortic rings. Next, four ring-shaped aortic rings were embedded into growth factor-reduced matrigel (Matrigel™, BD Biosciences) per well in 24 well cell culture plates. After 1-ml of the mixture of endothelial and smooth muscle medium 200 and 231 had been added (Cascade Biologics), each well received growth factor treatments daily for 6 continuous days. Four treatments were tested: (1) 20 ng/ml VEGF-A_{165}; (2) 20 ng/ml PDGF-BB; (3) VEGF and PDGF-BB in sequence delivery (VEGF for the first 3 days followed by PDGF-BB for the last 3 days); (4) control: only media with no growth factor administration. The aortic ring experiment was carried out under hypoxia (3% oxygen) in a 37 °C incubator and the medium was changed every day. The vessel growth was quantified by counting the total number of the newly formed vessel branches of each aortic ring using a microscope.

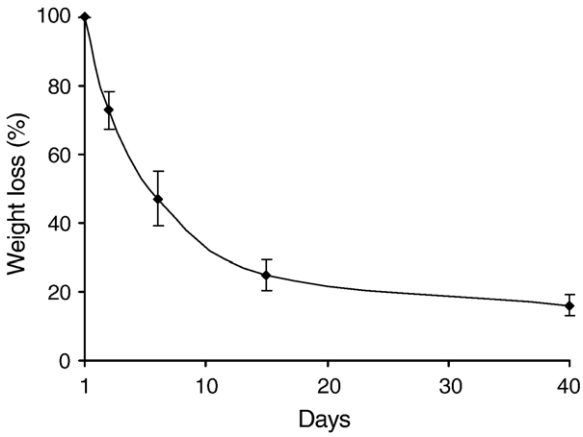

Fig. 3. Alginate degradation rate *in vitro* as measured by the dry mass loss (as percentage of initial dry mass) of hydrogels following incubation in a saline solution at 37 °C for various time periods. Values are given as mean± SEM, $n=4$ at each data point.

2.11. Statistical analysis

Data are presented as mean±SEM. Comparison between the groups was made by means of 1-way ANOVA (STATVIEW), followed by Fisher's PLSD test. Paired t-tests were performed to compare the differences between baseline and after treatment. Values were considered to be statistically significant at a value of $p \leq 0.05$.

3. Results

3.1. Characterization of the gel delivery system

The ability of the gel system to provide both a sustained delivery of growth factors, and a sequential release of VEGF-A_{165} and PDGF-BB was first assessed *in vitro*. Following an initial burst on the first day, VEGF-A_{165} displayed a sustained release from alginate gels at a rate of ~75 ng/day from days 1–12, and ~10 ng/day from days 12–30 (Fig. 2). In contrast, PDGF-BB release was delayed, compared to VEGF-A_{165} release, with a much smaller

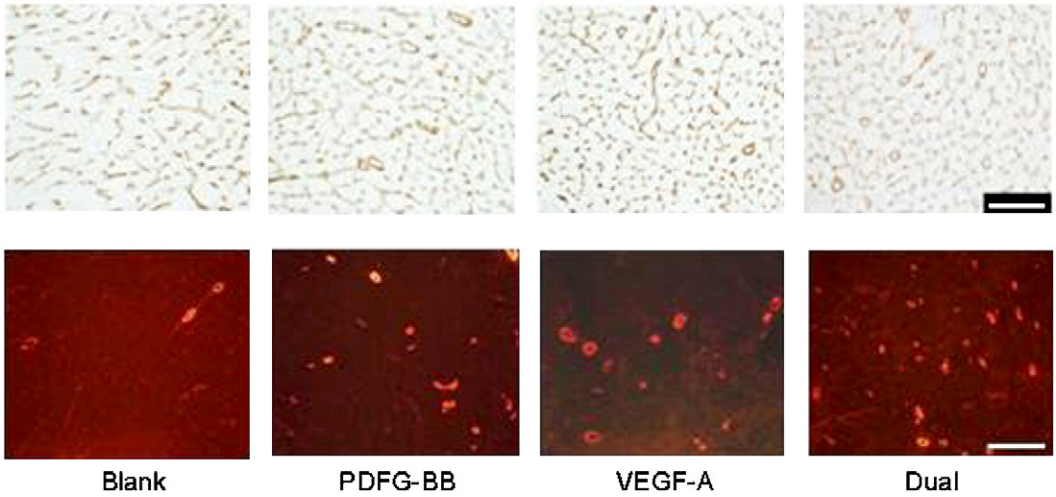

Fig. 5. Periinfarct myocardial capillary vessels identified by lectin staining (above), and α-SMC positive vessels (below) in tissues 4 weeks after delivery of alginare blank gel, alginate gel with PDGF-BB, VEGF-A_{165} or Dual (VEGF-A_{165} and PDGF-BB). The bars indicate 100 μm.

amount of factor released in the first day. PDGF-BB release was maintained at ~70 ng/day from days 1–30.

The degradation of this delivery system was next assessed *in vitro* and *in vivo*. As an initial indication of the ability of the gel system to be cleared from an injection site, gel degradation was monitored *in vitro* by measuring dry weight loss as function of time following incubation in PBS at 37 °C (Fig. 3). Alginate gels exhibited a relatively rapid degradation *in vitro*; within 7 days, approximately 60% of the initial gel mass was lost. Following injection *in vivo*, small gel fragments could be observed in tissue sections of the myocardium 4 weeks after delivery in approximately one-third of the rats (Fig. 4). No gel fragments were found at autopsy or in histological sections at this time point in the other two-thirds of the animals. No obvious fibrosis was observed in any animal receiving the gels. The clearance of the gels from the myocardium agreed well with the measured *in vitro* degradation profile, as expected for these hydrolytically degrading gels.

3.2. Capillary and arteriolar densities

Examination of myocardial periinfarct tissue sections four weeks after intervention revealed that delivery of alginate gels with no factor (alginate blank) or with PDGF-BB alone did not modify the capillary density, as compared to delivery

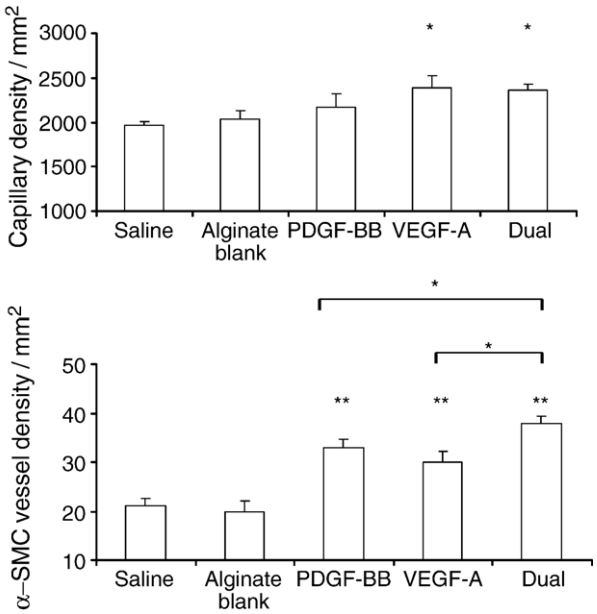

Fig. 6. Periinfarct myocardial capillary densities (above) and α-actin SMC positive vessel densities (below) 4 weeks after injection of saline ($n=6$), alginate blank ($n=6$) and alginate gel with PDGF-BB alone ($n=6$), VEGF-A_{165} alone ($n=7$) or Dual (VEGF-A_{165} and PDGF-BB, $n=7$). *$p<0.05$, ** $p<0.01$. ANOVA ($p<0.05$ for overall capillary densities and $p<0.01$ for α-SMC positive vessels) followed by PLSD test.

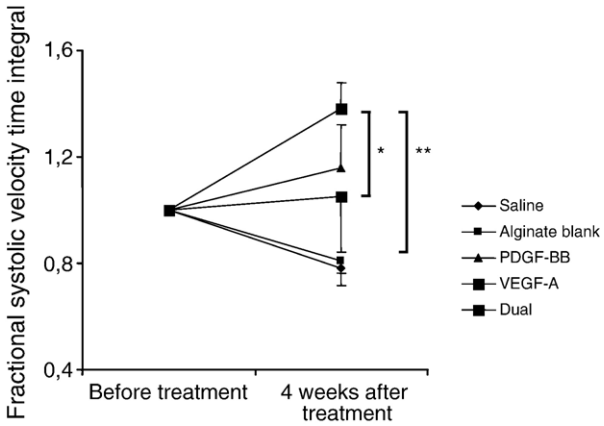

Fig. 7. Fractional systolic velocity-time integral 3 days before treatment and 4 weeks after delivery of VEGF-A_{165} alone ($n=7$), PDGF-BB alone ($n=6$), or Dual (VEGF-A_{165} and PDGF-BB, $n=7$) in alginate hydrogel, alginate blank ($n=6$) or saline ($n=6$). Normalization was done by dividing individual baseline and 4 week values with baseline value. * $p<0.05$ and ** $p<0.01$, ANOVA ($p<0.01$) followed by PLSD test.

of saline (Figs. 5 and 6). In contrast, delivery of VEGF-A_{165} and sequential delivery of VEGF-A_{165}+PDGF-BB increased ($p<0.05$) the capillary densities, as compared to alginate blank.

Alginate blank also did not modify the density of vessels with associated cells staining positive for α-actin smooth muscle cells (SMC). Compared to alginate blank, both delivery of VEGF-A_{165} ($p<0.01$) and PDGF-BB ($p<0.01$) alone increased the density of á-actin SMC associated vessels Further, sequential delivery of VEGF-A_{165} and PDGF-BB resulted in an even higher α-actin SMC positive vessel density, as compared to alginate blank ($p<0.01$), and the differences between this condition and VEGF-A_{165} or PDGF-BB alone were also statistically significant ($p<0.05$).

3.3. Left ventricular function

The systolic velocity-time integral was analyzed as a marker of myocardial function, and this deteriorated with time in both controls (alginate blank or saline administration, $p<0.01$) (Fig. 7). Delivery of either VEGF-A_{165} or PDGF-BB alone prevented the decrease in the systolic velocity-time integral, compared to the baseline, and tended to improve function compared to alginate blank at 4 weeks after treatment. Strikingly, sequential delivery of VEGF-A_{165} and PDGF-BB increased the systolic velocity-time integral, compared with alginate blank ($p<0.01$), and compared to delivery of VEGF-A_{165} alone ($p<0.05$). No significant differences in the LVDd and ejection fraction were observed between the groups. The LVDd values at 4 weeks (normalized to the baseline) were as follows: VEGF-A_{165} alone: 1.18±0.04; PDGF-BB alone: 1.11±0.04; VEGF-A_{165} and PDGF-BB: 1.18±0.05, alginate blank 1.26±0.05 and saline 1.20±0.05. The corresponding ratios for the ejection fractions were (again, normalized to baseline): VEGF-A_{165} alone 0.82±0.03; PDGF-BB alone: 1.02±0.12; VEGF-A_{165} and PDGF-BB: 0.94±0.05 and alginate blank 1.12±0.06.

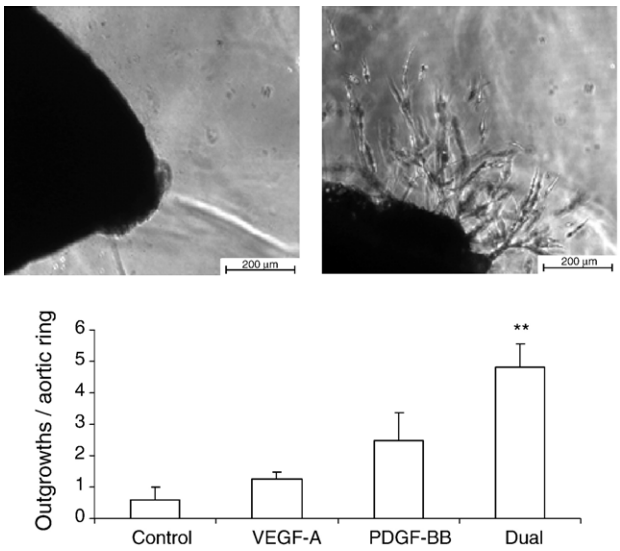

Fig. 8. Number of vascular outgrowths from aortic rings cultured under conditions of 3% oxygen: Above, left: no growth factor administration (no outgrowth from aortic ring); Above, right: sequenced exposure to VEGF-A_{165} and PDGF-BB (six outgrowths from aortic ring); Below: quantification of vascular outgrowths from aortic rings with exposure to control (media without growth factors, $n=5$), VEGF-A_{165} ($n=4$), PDGF-BB ($n=4$), or sequenced exposure to VEGF-A_{165} followed by PDGF-BB ($n=4$). ** $p<0.01$, versus control. ANOVA ($p<0.01$) followed by PLSD test.

3.4. Angiogenic effect in vitro of sequenced VEGF-A_{165} and PDGF-BB delivery

Finally, the influence of sustained and sequenced VEGF-A_{165} and PDGF-BB exposure on angiogenesis was confirmed using an aortic ring angiogenesis assay performed under hypoxic conditions (3% oxygen tension). The control condition led to minimal vessel outgrowth, while continuous exposure to VEGF-A_{165} or PDGF-BB alone for 6 days each led to an increase in the number of outgrowths, although not statistically significant increases. In contrast, sequential exposure to VEGF-A_{165} (3 days) followed by PDGF-BB (3 days) led to a statistically significant increase in vessel outgrowth, as compared to control ($p<0.01$) (Fig. 8).

4. Discussion

In this study, sequential and sustained delivery of VEGF-A_{165} and PDGF-BB using an injectable alginate hydrogel induced greater remodelling of the vasculature and improvement of cardiac function more than single growth factor delivery.

VEGF is an endothelial specific growth factor that mainly induces angiogenesis [13,14], and our results are consistent with the accepted mechanism of VEGF-A_{165} action. Sustained VEGF-A_{165} delivery also slightly increased the density of α-actin SMC containing vessels, but the mechanisms underlying this effect of VEGF-A_{165} on SMCs have not been completely elucidated. VEGFR2 is expressed on SMCs [15] and VEGF-A_{165} can stimulate vascular SMC migration and have an arteriogenic effect [16–20].

Maturation of blood vessels may be induced by SMC recruitment to nascent blood vessels, and PDGF-BB, a stimulator of SMC growth [5–7], appears to be instrumental in this maturation process. This suggests PDGF-BB should be present in the later phases of VEGF-A_{165} delivery in order to boost VEGF-A_{165}-induced angiogenesis via the recruitment of SMCs to newly formed capillaries. In this study, sequential growth factor delivery induced a higher percentage of α-actin SMC positive vessels than VEGF-A_{165} delivery alone, a result that confirmed that PDGF-BB enhanced the VEGF-A_{165} action. PDGF-BB may further stimulate and enhance the VEGF-A_{165} maturation process in two ways: recruitment of SMCs and pericytes to the newly formed capillaries, and proliferation of SMCs recruited by VEGF-A_{165}. The sequenced delivery of VEGF-A_{165} and PDGF-BB also may increase angiogenesis compared to single factor exposure, as indicated in the aortic ring *in vitro* study. Although the

capillary density did not change significantly with sequential delivery *in vivo*, this might reflect the enlargement and maturation of capillaries remodelled into α-actin SMC positive vessels. It is possible that the sequenced delivery might stimulate both angiogenesis and vessel maturation into α-actin-SMC positive vessels.

The more pronounced vessel maturity induced by sequential delivery of the proteins would be expected to translate into improved blood flow, leading to less ischemia in the peri-infarct region. Tissue velocity reflects myocardial function and cardiac perfusion [21,22], and the improved tissue velocity integral with sequential delivery provided functional evidence for improved perfusion in this condition. This observation also confirms that angiogenic therapy can improve the systolic function following myocardial infarction [23–25]. The ejection fraction and LVDd did not change significantly in this randomised study. Thus the results suggest that left ventricular function improved as regards tissue velocity, but not to such an extent that ejection fraction and LVDd were effected. These results might relate to random error introduced by the variability of infarct size in this study, and this would infer a need for much larger experimental groups to detect the effects on LVDd and ejection fraction. Alternatively, these results may arise from the methodological differences in these measurements. LVDd and ejection fraction were measured at the 2D echo in the long axis view at the level of the largest left ventricular diameter. The measurements thus reflect mainly only the measured cross-sectional plane of the left ventricle. On the contrary, TVI is measured from the base of the left ventricle at the mitral valve plane in the longitudinal direction towards the apex and reflects subendocardial more than subepicardial changes [26]. Thus, in addition to its higher resolution than the echocardiographic signal, it indexes the integrated change of the left ventricle instead of measuring only a cross-sectional plane, as analyzed by the LVDd and ejection fraction measurements.

In this study, injectable alginate gels were used to release VEGF-A_{165} and PDGF-BB in the peri-infarction heart region. Blank alginate gels led to similar vessel densities as saline injection, suggesting that alginate did not induce angiogenesis itself or any significant inflammatory reaction. Alginates are naturally occurring biocompatible polysaccharides [9], but alginate hydrogels typically present low and uncontrolled degradation [9]. However, partial oxidation of alginate and the use of a combination of polymers with distinct molecular weight distributions to form gels can provide controlled degradation kinetics [27,28], allowing control of the release kinetics of incorporated factors [12].

PDGF-BB released from this injectable system showed a slower release than VEGF-A_{165}. This likely relates to the different affinity to alginate for the two growth factors. Delivery of growth factors with this gel system leads to ~95% retention of the released factors in the muscle tissue immediately surrounding (<5 mm) the injection site, as directly measured by quantitatively digesting serial tissue samples and quantifying tissue lysates with an ELISA [12]. This gel delivery system also maintains the local factor concentrations at physiologically relevant concentrations for more than 15 days following a single injection. In contrast, injection of a solution of growth factor leads to rapid loss of the factor from the site of injection, and concentrations in the muscle drop below detectable levels by 72 h. The localized and sustained gel delivery of factors also led to minimal systemic exposure; peak serum levels never rose above 0.2 ng/ml with this mode of delivery [12]. We have previously also noted this minimal rise in serum levels in other polymeric systems that enable localized and sustained delivery of VEGF-A_{165} and PDGF-BB [8,29]. The experimental findings of tissue and serum distribution are consistent with mathematical models of growth factor tissue distribution with polymer system delivery [30].

Localized and sustained gel delivery of these factors was not found to lead to significant inflammation or fibrosis in the surrounding muscle tissue in this study or in past studies [12]. Sustained and localized VEGF-A_{165} delivery can lead to short-term increases in the number of infiltrating macrophages [31], a result consistent with its known biological effects. However, no increase in muscle fibrosis has been observed in several studies using polymer delivery of VEGF-A_{165} and a combination of VEGF-A_{165} and PDGF-BB in skeletal muscle [8,12,31]. The increased macrophage infiltration may actually aid in revascularization and maturation of the new vasculature due to the multiple cytokines and factors released from these cells. The ability of the gel delivery to enhance vascularisation, while avoiding fibrosis, likely relates to the ability of this delivery approach to avoid the extremely high and transient factor concentrations that result from the typical injections of factors in solution, and instead provide a lower and longer-term tissue exposure.

5. Conclusions

An alginate hydrogel delivery system is useful for growth factor delivery in a myocardial infarction model. Sequential delivery of VEGF-A_{165} and PDGF-BB induces a greater formation of mature vessels and improves cardiac function compared to delivery of single factors. Growth factor delivery with these alginate gels provides precise doses for a desired time-frame, preventing multi-time deliveries that are not practical for myocardial delivery, indicating alginate is a promising biomaterial for myocardial delivery of growth factors. This may indicate its clinical usefulness.

Acknowledgements

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