Electrically induced vasomotor responses and their propagation in rat renal vessels *in vivo*

Michael Steinhausen, Karlhans Endlich, Rainer Nobiling*, Niranjan Parekh† and Florian Schütt

*Institut für Anatomie und Zellbiologie I, *Institut für Experimentelle Chirurgie and †I. Physiologisches Institut, Universität Heidelberg, D-69120 Heidelberg, Germany*

1. Vasomotor responses (VMR) induced by local electrical stimulation were studied in the vasculature of the split hydronephrotic rat kidney by *in vivo* microscopy.

2. Unipolar pulses, which were applied by a micropipette positioned close to the vessel wall, elicited local and propagated VMR. Depolarizing and hyperpolarizing currents caused vasoconstriction and vasodilatation, respectively.

3. The magnitude of VMR could be controlled within seconds by variation of pulse frequency, pulse width and voltage. VMR were abolished by slight retraction of the stimulating micropipette. Repetitive electrical stimulation resulted in reproducibly uniform VMR.

4. Propagated VMR decayed with increasing distance from the stimulation site. They decayed more rapidly in the upstream than in the downstream flow direction in interlobular arteries. The longitudinal decay was well approximated by an exponential function with significantly different length constants of 150 ± 40 μm (upstream, $n = 5$) and 420 ± 90 μm (downstream, $n = 8$).

5. Our results show that vasomotor responses, which are initiated by changes in membrane potential, are propagated over distances of potential physiological importance in interlobular arteries.

It is largely unknown whether changes in vasomotor activity in response to physical, nerval or humoral stimuli are propagated in the renal vasculature. Functional interdependence of renal vessels is suggested by the presence of vasomotion in the rat hydronephrotic kidney (Steinhausen & Baehr, 1989; Loutzenhiser, Epstein, Hayashi & Horton, 1990), oscillations in renal blood flow in humans (Hollenberg, Meyerovitz, Harrington & Sandor, 1987) and synchronized spontaneous oscillations in tubular pressure of adjacent nephrons which originate from the same interlobular artery (Holstein-Rathlou, 1987; Källskog & Marsh, 1990). Indirect evidence for the propagation of vasomotor responses (VMR) in renal vessels has been provided recently by the finding that the degree of haemodynamic coupling between two nephrons is inversely related to the vascular distance between their glomeruli (Chen, Yip, Marsh & Holstein-Rathlou, 1995). Moreover, it has often been assumed that VMR induced by the tubuloglomerular feedback mechanism in the distal afferent arteriole have to be propagated upstream in order to elicit sufficient changes in renal vascular resistance to autoregulate renal blood flow (Carmines, Inscho & Gensure, 1990; Moore & Casellas, 1990; Øien & Aukland, 1991). This propagation has recently been reported in preliminary studies of the *in vitro* juxtamedullary nephron preparation (Moore, Omar & Casellas, 1994; Wagner, Holstein-Rathlou & Marsh, 1996).

In the hamster cheek pouch and rat cremaster muscle the propagation of VMR *in vivo* is a well-known phenomenon (Duling & Berne, 1970; Segal & Duling, 1986). Vascular diameter changes in response to localized pharmacological stimulation are propagated over considerable distances in these preparations (Segal, Damon & Duling, 1989; Delashaw & Duling, 1991; Segal, 1991). Thus, propagation of VMR results in the functional coupling of small arteries and arterioles. This mechanism is believed to coordinate organ blood flow, e.g. by upstream propagation of metabolic arteriolar vasodilatation during muscle exercise (Segal, 1991, 1992). It has been shown that cell-to-cell coupling of endothelial and/or smooth muscle cells via gap junctions provides the molecular basis for propagation (Segal & Duling, 1987, 1989; Segal & Bény, 1992). By contrast, in the isolated hydronephrotic kidney of the mouse the vascular smooth muscle cells of renal vessels appear to be electrically uncoupled (Nobiling, Gabel, Persson, Dietrich & Bührle, 1991). However, the results of this study may not be applicable to the *in vivo* situation, since the kidney was

perfused with collagenase prior to the electrophysiological measurements. The enzyme may have induced considerable endothelial damage.

In the present study, we analysed propagation of VMR in the split hydronephrotic kidney of rats, which is the only technique to visualize renal vessels *in vivo* over considerable length (Steinhausen, Snoei, Parekh, Baker & Johnson, 1983). In order to overcome the inherent problems of pharmacological microapplication, i.e. unpredictable diffusion and convection of vasoactive agents, we used local electrical stimulation of renal vessels. By this method, we were able to induce local VMR, to observe propagated VMR, and to calculate length constants.

METHODS

Preparation of the split hydronephrotic kidney

Experiments were performed on sixteen female Wistar rats weighing between 200 and 250 g in accordance with local and national guidelines for animal protection. The technique of splitting the rat kidney has been described in detail previously (Steinhausen *et al.* 1983). In brief, the ureter of the left kidney was permanently ligated via a small flank incision during pentobarbitone sodium anaesthesia (Nembutal®, 60 mg kg^{-1} I.P., Ceva, Bad Segeberg, Germany). The rats were carefully observed until they had completely recovered from anaesthesia. All the animals survived surgery without apparent ill effects or signs of discomfort. The final experiments were done under thiobutabarbitone anaesthesia (Inactin®, 100 mg kg^{-1} I.P., Byk Gulden, Konstanz, Germany) 2–3 months after induction of hydronephrosis. Body temperature was maintained at 37·0–37·5 °C via a heating table, systemic blood pressure was monitored via a cannula in the left femoral artery, and isotonic saline (50 μl min^{-1}) was continuously infused via a cannula placed in the jugular vein. After exposure of the left hydronephrotic kidney by a flank incision, the kidney was split along the greater curvature with a thermal cautery. The ventral half of the kidney was sutured to a semicircular frame that was attached to the bottom of a Plexiglas bath to visualize the vasculature of cortical glomeruli by transillumination microscopy. The entry of the renal hilus into the bath was sealed with silicone grease, and the bath was filled with an isotonic, isocolloidal solution (Haemaccel®, Behringwerke, Marburg/Lahn, Germany) maintained at 37 °C. A Leitz Ultropak water-immersion objective (UO-55) was combined with a television and video recording system for *in vivo* microscopy. Kidney preparations were allowed to equilibrate in the tissue bath for at least 1 h after the surgical procedure. The animals were killed at the end of the experiments by an overdose of the anaesthetic.

Local electrical stimulation

For local electrical stimulation, glass micropipettes with an outside tip diameter of about 10 μm were made from glass capillary tubes of 1 mm diameter. The pipettes were cleaned with absolute ethanol, backfilled with 2 mol l^{-1} NaCl solution, mounted on a micromanipulator and connected to a Grass stimulator via Ag–AgCl wires. A second wire placed in the tissue bath served as the reference electrode. The pipette was positioned as close as possible to the vessel wall. As inferred from the focal depth, the distance between the vessel and the pipette was about 10–20 μm. Interlobular arteries and afferent and efferent arterioles were identified by their branching pattern. Unipolar pulses with adjustable frequency (2–8 Hz), pulse width (2–8 ms), and voltage (up to 80 V) were used for stimulation. Electrical parameters were chosen to induce local constriction of about 50%. The vessels were stimulated for 10–20 s, the time necessary to reach the steady state diameter. The interval between two consecutive stimulations was at least 90 s to allow for complete recovery of the vessel. In some experiments a single vessel was repetitively stimulated up to 35 times, to determine the reproducibility of electrically induced VMR. All stimulations were recorded on videotape for later off-line analysis.

Propagated vasomotor responses

In order to quantify propagation, propagated VMR were measured at several vascular sites either in the downstream or upstream blood flow direction from the stimulation site. For the measurement of downstream propagation, the stimulating micropipette was placed in the proximal part of an interlobular artery and it was placed in the distal part to measure upstream propagation. Thereby, VMR were propagated through comparable vascular segments of interlobular arteries under both conditions. In the majority of the trials downstream and upstream propagation were measured in separate arteries. After placing the micropipette, the vessel was stimulated several times to ensure that a reproducible local response could be evoked. Thereafter, the microscopic field was moved in a random fashion with the aid of a computer-controlled, motorized stage to several preselected sites (between 3 and 10), which were at variable distances from the stimulation site. The position of the stimulating pipette, which was mounted on the animal stage, remained unchanged relative to the microvessel segment where the measurements were taken. At each site we measured the vasoconstriction in response to one repeated stimulation. At the end, the stage was moved back to the stimulation site and the vessel was again stimulated to check for the stability of the local response. We were forced to adopt this procedure to measure the propagated responses because the high magnification needed to measure vascular diameters restricted the video image to a width of approximately 200 μm.

Data analysis

Data are presented as means ± S.E.M. A special procedure was used to calculate the mean decay of propagation as a function of distance from the stimulation site, since locally induced vasoconstriction differed among the individual trials, and since propagated VMR were measured at variable distances in the individual trials. The locally induced vasoconstriction was set to 100% in each trial. From the data points of each trial a step function (normalized vasoconstriction as a function of distance) was constructed with the data points being centred in the steps. The individual step functions were averaged to obtain the mean decay of downstream and upstream propagation. They were averaged only up to the common maximal distance (590 μm downstream and 300 μm upstream) to avoid a bias towards trials in which VMR were propagated to, and therefore measured at, more distant sites. Since the decay of VMR was reasonably approximated by exponential functions (cf. Fig. 5), we determined the length constant λ for each individual decay. To this end, the percentage changes of diameters $c(x)$ with distances x to the stimulation site were fitted to an exponential function for each trial: $c(x) = c_0 \exp(-x/\lambda)$; c_0 denotes the percentage change of the diameter at the stimulation site. Statistical analysis was done by Student's t test or analysis of variance (ANOVA) as appropriate. The Bonferroni method was used for multiple comparisons. The significance level was set at $P < 0{\cdot}05$.

RESULTS

Local vasomotor responses

Local electrical stimulation with depolarizing currents induced vasoconstriction in renal arcuate and interlobular arteries and afferent and efferent arterioles (Fig. 1). Stable responses were established within seconds. The degree of vasoconstriction could be controlled by variation of the electrical stimulation parameters (pulse frequency, pulse width and voltage). Above a threshold value, vasoconstriction was approximately proportional to the increase in one parameter – the remaining two parameters were kept constant – until saturation was reached (Fig. 2*A*). Doubling of pulse frequency (4 Hz), pulse width (2 ms) or voltage (40 V) resulted in similar significant increases in vasoconstriction: 2·4 ± 0·4-fold ($n = 5$), 3·8 ± 0·9-fold ($n = 5$) or 4·0 ± 0·6-fold ($n = 6$), respectively ($P = 0·25$, analysis of variance among relative increases). Maximal inducible vasoconstriction was: 48 ± 8% ($n = 10$) in interlobular arteries, 78 ± 6% ($n = 15$) in afferent arterioles and 21 ± 5% ($n = 9$) in efferent arterioles, the values being significantly different from each other. In addition, VMR were highly sensitive to the distance between the stimulating micropipette and the vessel wall (Fig. 2*B*). Slight retraction of the stimulating pipette abolished VMR, indicating that the electrical field was focused at the tip of the micropipette. Local electrical stimulation with hyperpolarizing currents resulted in vasodilatation (Fig. 2*C*). Maximal inducible vasodilatation in interlobular arteries was 25 ± 6% ($n = 4$). Electrically induced VMR were highly reproducible during repetitive stimulation (up to 35 times in 1 h) at the same vascular site

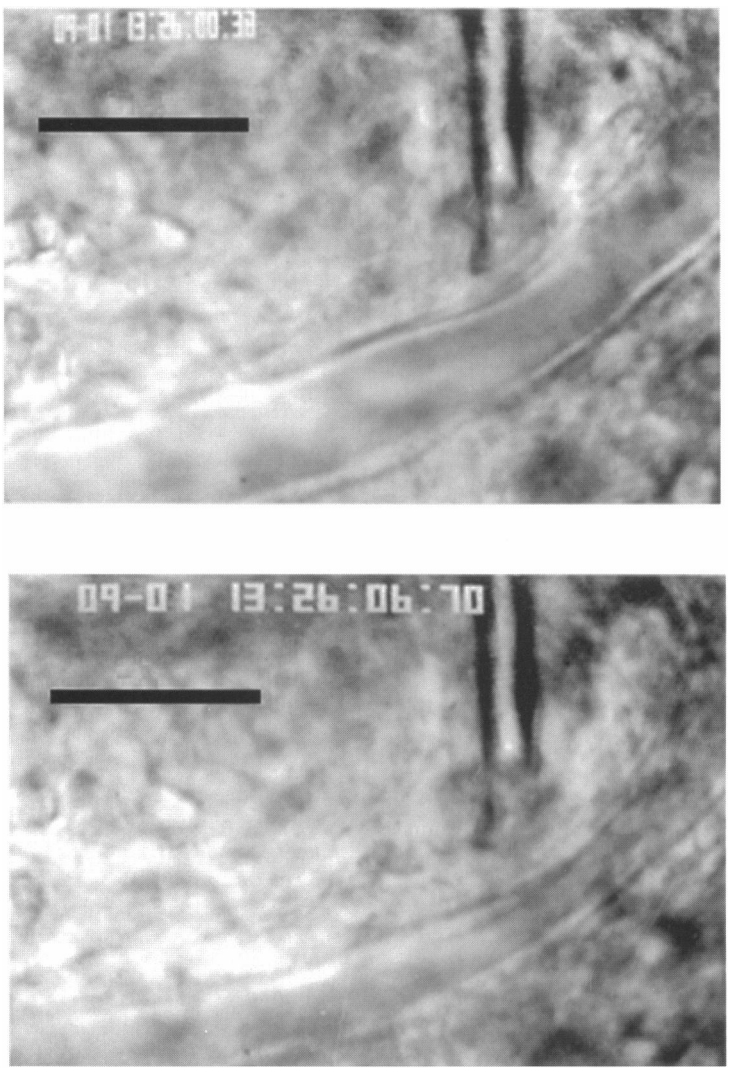

Figure 1. Vasoconstriction induced by local electrical stimulation

Local electrical stimulation of renal vessels with depolarizing currents, which were applied via a micropipette positioned close to the vessel wall, induced vasoconstriction. Microphotographs illustrate the response of an interlobular artery. The diameter of the vessel was about 20 μm during control conditions (upper panel) and decreased in response to depolarizing currents (lower panel). Scale bars, 30 μm.

(Fig. 3). The accuracy of the maximal vasoconstriction during repetitive stimulation was around 5% of the baseline diameter of the vessel.

Propagation of vasomotor responses

VMR induced by local electrical stimulation were not only observed in proximity to the micropipette but also at more distant sites in the upstream and downstream direction along the vessel. Responses decayed with increasing distance from the tip of the micropipette. Figure 4 summarizes the mean decay of propagated VMR in the downstream ($n = 8$) and upstream ($n = 5$) flow direction as measured in different interlobular arteries in seven animals. Neither the mean parameters of electrical stimulation nor the vasoconstriction induced at the stimulation site were statistically different in the downstream and upstream propagation experiments. Along 300 μm in the upstream direction, the propagated VMR gradually decreased to 18 ± 1% of the initial vasoconstriction measured at the stimulation site. Along the same distance in the downstream direction, the propagated VMR decreased to only 54 ± 11% (unpaired $P < 0{\cdot}05$ upstream *vs.* downstream). In order to compare the localization of the stimulation sites between the different experiments, we measured the vascular length from the stimulation site to the last glomerulus that was fed by the same artery. The length between the stimulation site and the last glomerulus was 530 ± 160 μm in the upstream propagation trials and 1080 ± 160 μm in the downstream propagation trials ($P < 0{\cdot}05$). The mean length of the afferent arteriole of the last glomerulus (330 ± 50 μm) did not differ significantly between the upstream and downstream trials. Thus, upstream and downstream propagation of VMR were studied in similar vascular segments of interlobular arteries. In addition, in both the upstream and downstream trials, the interlobular artery showed a narrowing of 5 ± 1% per 100 μm length in the downstream direction.

Since VMR has been demonstrated to decay exponentially in the hamster cheek pouch and the rat cremaster muscle (Segal *et al.* 1989; Delashaw & Duling, 1991; Segal, 1991), we fitted exponential functions to the decays. VMR decayed with significantly different length constants λ of 420 ± 90 μm ($n = 8$) and 150 ± 40 μm ($n = 5$) in the downstream and upstream direction, respectively. The validity of an exponential description of the decays was checked by plotting all data points ($n = 70$) in a normalized diagram (Fig. 5). The following parameters for an exponential fit were obtained: coefficient of variation of 7% for λ, $R^2 = 0{\cdot}80$, and $P < 0{\cdot}0001$. Thus, the decays of VMR were well approximated by exponential functions.

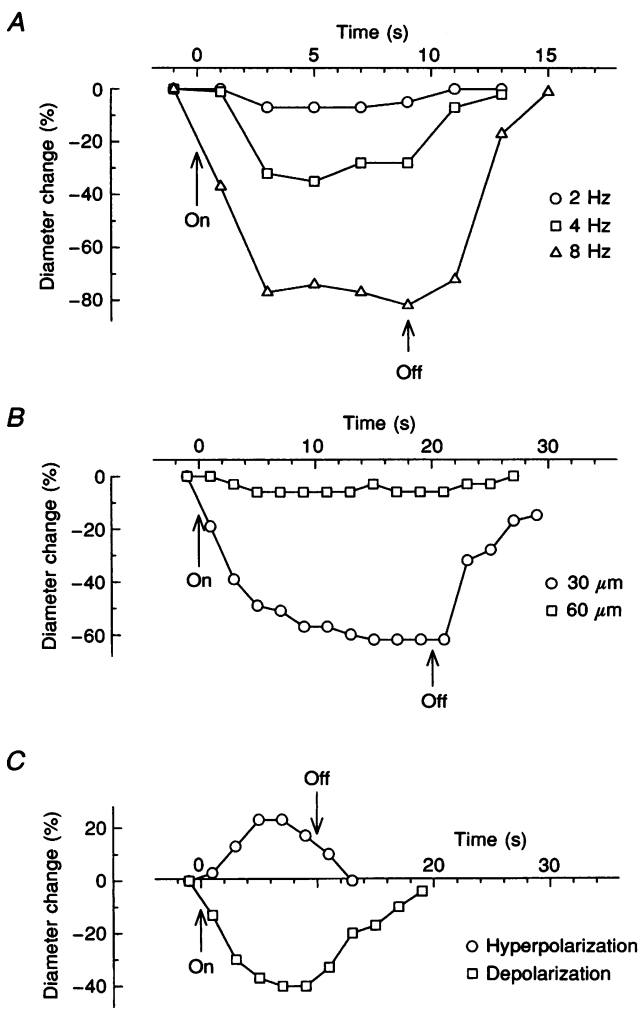

Figure 2. Stimulus dependence of electrically induced vasomotor responses

Vasomotor responses induced by local electrical stimulation could be controlled by variation of the stimulation parameters. Representative examples of stimulations in interlobular arteries are shown. *A*, variation of pulse frequency (2 ms, 70 V). *B*, variation of distance between vessel and stimulating micropipette (8 Hz, 8 ms, 40 V). *C*, vasoconstriction and vasodilatation in response to depolarizing and hyperpolarizing currents, respectively (±50 V DC). Electrical parameters (frequency, pulse width, voltage) for the stimulations are given in parentheses.

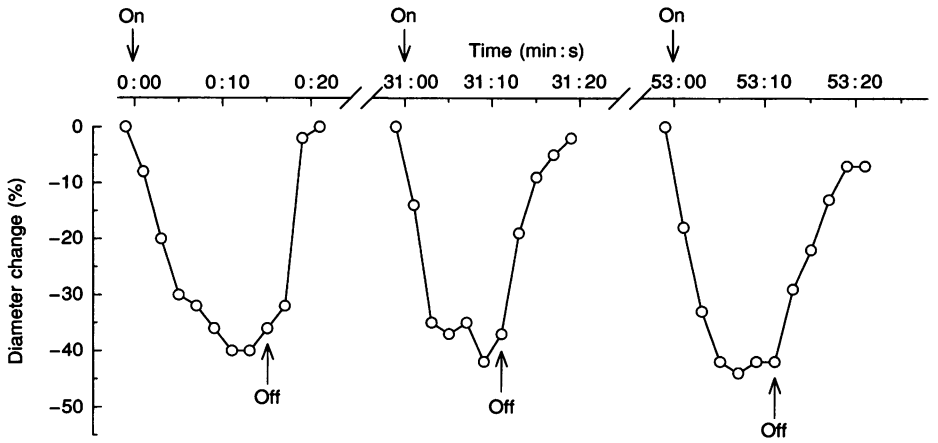

Figure 3. Repetitive electrical stimulation

Repetitive local electrical stimulation (4 Hz, 2 ms, 40 V) at the same vascular position induced reproducible vasomotor responses. Representative tracings of diameter changes in response to the first, the 20th and the last stimulation of an interlobular artery are shown. The vessel was stimulated 35 times during 54 min with a duration of less than 20 s for each stimulation.

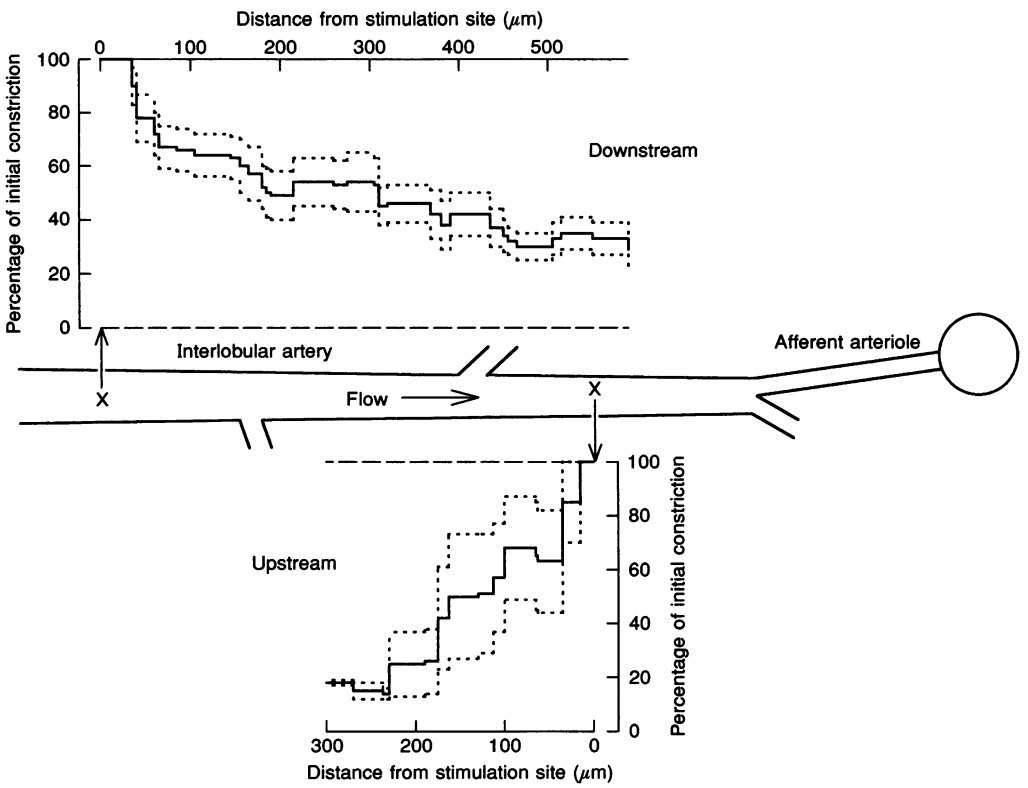

Figure 4. Longitudinal decay of propagated vasomotor response

Locally induced vasoconstriction decayed with increasing distance from the site of stimulation and was propagated more efficiently in the downstream direction. The longitudinal decay is shown for the downstream ($n = 8$) and for the upstream ($n = 5$) direction (continuous line, mean; dotted lines, S.E.M.). Vasoconstriction at the site of stimulation was set to 100% in each experiment. The sites of stimulation (marked by crosses) for downstream and upstream propagation were located in the interlobular artery at a distance of 1080 ± 160 μm and 530 ± 160 μm from the last glomerulus, respectively. The mean length of the afferent arteriole of the last glomerulus was 330 ± 50 μm in these experiments.

Concerning the velocity of propagation of VMR in renal vessels, we could not detect a time delay between the onset of local and propagated vasoconstriction even for separations of more than 1000 μm. The time resolution in our experiments was at best 1 s, since local and propagated VMR could not be visualized simultaneously and since maximal vasoconstriction needed a few seconds to develop (cf. Fig. 2). VMR are therefore propagated with a velocity of at least 1 mm s^{-1}.

DISCUSSION

Until now propagation of VMR *in vivo* has been studied in arterioles of the hamster cheek pouch or rat cremaster muscle by pulsed local application of vasoactive agents with micropipettes (Duling & Berne, 1970; Segal & Duling, 1986; Segal *et al.* 1989; Delashaw & Duling, 1991; Segal, 1991). Likewise, upstream propagation of VMR in afferent arterioles has been studied by local application of KCl with micropipettes in the *in vitro* juxtamedullary nephron preparation (Moore *et al.* 1994; Wagner *et al.* 1996). The present study introduces electrical stimulation of renal vessels as a novel approach to investigate VMR in the microcirculation. In contrast to pharmacological stimulation, electrical stimulation appears to be easier to control. Variation of the stimulation parameters allows for a relatively rapid adjustment in the magnitude of VMR. Moreover, only a small area of the vessel is directly affected by electrical stimulation owing to the high density of the electrical field at the tip of the micropipette. Accordingly, VMR were abolished by a slight retraction of the stimulating micropipette. During pharmacological stimulation by pressure ejection we have always been uncertain about the strictly local nature of stimulation, since dye-coloured solutions, which we applied by micropipettes, spread over considerable distances in an unpredictable manner due to convection.

During local electrical stimulation, we always observed propagation of local VMR. Since VMR were characterized by an approximately exponential decay, length constants for propagation could be derived. Surprisingly, the length constant for the downstream direction (420 μm) was more than twice as high as that for the upstream direction (150 μm). At present, it remains unclear what mechanism accounts for a directional propagation of VMR as observed in this study. Differences in vessel depths, in electrical conductance of the surrounding tissue and in vascular reactivity between upstream and downstream stimulation sites as well as conduction of electrical current and transport of biochemical factors in the vessel lumen could selectively attenuate or amplify the upstream or downstream propagation of VMR. Therefore, the possibility of an artifact cannot be completely ruled out. However, the confinement of the electrical field to a small area around the tip of the micropipette and the observation of a true difference in reactivity between afferent and efferent arterioles, embedded in the same surrounding tissue, favour an interpretation of physiological significance. On the one hand, downstream narrowing of the interlobular artery (cf. Results) could be accompanied by a reduction of the smooth muscle layer, leading to an increase in current density. On the other hand, the efficacy of electromechanical coupling in smooth muscle could vary in different renal vessels. Accordingly, local electrical stimulation induced decreasing VMR in different renal vessels in the order: afferent arteriole, interlobular artery, efferent arteriole. Along the same line, membrane depolarization by local

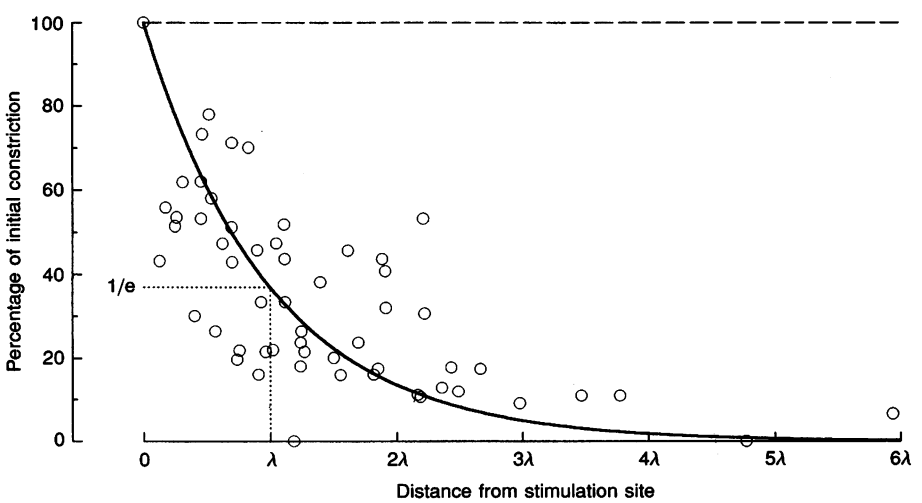

Figure 5. Exponential fit to the decay of vasomotor response

Normalized data points of 13 trials were reasonably approximated by an exponential function (with a coefficient of variation of 7% for λ, $R^2 = 0{\cdot}80$ and $P < 0{\cdot}0001$). The data points of each trial were normalized with respect to their length constant λ, as calculated for each trial separately, and the vasoconstriction at the site of stimulation.

application of KCl results in less pronounced constriction of efferent as compared with afferent arterioles (Carmines, Fowler & Bell, 1993; Conger & Falk, 1993).

In preliminary reports from the *in vitro* blood-perfused juxtamedullary nephron preparation, upstream propagation of VMR, which was evoked in the distal part of the afferent arteriole by local microapplication of KCl, has been demonstrated (Moore *et al.* 1994; Wagner *et al.* 1996). A 'mechanical' length constant of 430 ± 90 μm (Wagner *et al.* 1996) and a 'mean length of propagation' of 410 ± 80 μm (Moore *et al.* 1994) were calculated. Propagation of VMR in the downstream direction was not investigated in these reports. There are several reasons which could account for the at least 2-fold higher length constants in the upstream direction given in these reports as compared with our present results. First, afferent arterioles in the juxtamedullary nephron preparation are about 700 μm long (Moore *et al.* 1994), which is more than three times the length of cortical afferent arterioles. Teleologically reasoning, length constants in juxtamedullary afferent arterioles could be proportionally higher to obtain vascular interdependence between juxtamedullary nephrons similar to cortical nephrons. Second, since propagated VMR were evoked by microapplication of KCl in juxtamedullary afferent arterioles, propagation could have been artificially enhanced by KCl convection. Third, the strength of vascular coupling could differ between afferent arterioles and interlobular arteries. Finally, if propagation of VMR is mediated via gap junctions, differences in length constants could also be explained by a different level of expression or phosphorylation of gap junctional proteins, which control gap junctional conductance and are regulated through protein kinase A and C (Kolb & Somogyi, 1991).

Indirect evidence for the propagation of VMR in the renal vasculature is also available. A hypothetical, retrograde (upstream) propagation of the VMR, initiated by the tubuloglomerular feedback at the distal afferent arteriole, has been proposed to explain synchronized variations in tubular pressure of nephrons fed by the same interlobular artery (Holstein-Rathlou, 1987; Källskog & Marsh, 1990). Our results suggest that propagation of VMR in the upstream direction with a length constant of 150 μm is in the range of the afferent arteriolar length. This is compatible with the results obtained by Källskog & Marsh, who observed that the covariation in tubular pressure variations of interacting nephrons is about 25% (Källskog & Marsh, 1990). Furthermore, Chen *et al.* (1995) measured the interaction strength in pairs of nephrons, which originated from a common interlobular artery, by measuring stop flow pressure changes in response to perfusion of only one nephron. In addition, they also measured the arteriolar separation between the two glomeruli in vascular casts, which is the distance that one has to travel from one glomerulus to the other along the vessels. Nephron interaction strength was inversely related to the arteriolar separation with a length constant of about 600 μm. However, it is by no means clear how this length constant of 600 μm for changes in stop flow pressure relates to a length constant for propagation of VMR. Stop flow changes are caused by resistance changes, which in turn depend on the integrated diameter changes of the interlobular artery and two afferent arterioles. Despite the discrepancies discussed above, the available data suggest, however, that length constants for the propagation of VMR in the kidney are in the range of only a few hundred micrometres.

Length constants for the propagation of VMR *in vivo* have also been determined in the hamster cheek pouch and rat cremaster muscle during pharmacological stimulation. In these preparations, VMR induced by various vasodilators and constrictors were propagated bidirectionally with similar length constants ranging from 1·6 to 2·5 mm (Segal *et al.* 1989; Delashaw & Duling, 1991; Segal, 1991). Thus, VMR are communicated over considerably longer distances in these tissues as compared with the kidney. This may represent an important mechanism to increase blood flow by the upstream propagation of arteriolar vasodilatation in these tissues (Segal, 1992). In the kidney, on the other hand, relatively small length constants may be crucial for a differential regulation of preglomerular vessels (Steinhausen & Endlich, 1996).

Propagated VMR in the hamster cheek pouch were shown to be conducted via gap junctions in the vessel wall (Segal & Duling, 1987, 1989). Direct evidence for a pathway for electrotonic conduction in the vessel wall has been provided by Segal & Bény in the hamster cheek pouch *in vivo* (Segal & Bény, 1992). They observed dye-coupling of endothelial cells only. In the rat kidney, a dense coupling of endothelial cells (Mink, Schiller, Kriz & Taugner, 1984) and the presence of gap junctions in myoendothelial contacts (Taugner, Kirchheim & Forssmann, 1984) has been demonstrated in the renal vasculature by electron microscopy, whereas smooth muscle cells in afferent arterioles and interlobular arteries appear to be only sparsely coupled (Taugner & Hackenthal, 1989). In agreement with the latter finding, Nobiling *et al.* (1991) reported that vascular smooth muscle cells of afferent arterioles in the isolated hydronephrotic kidney of the mouse are practically uncoupled with electrical length constants of less than 10 μm. However, these results were obtained in the absence of a functional endothelium, since the vasculature was perfused with collagenase for several minutes to facilitate micropuncture. Thus, the renal vascular endothelium might play a predominant role in the propagation of VMR.

In the present study we used the split hydronephrotic kidney, an established model for the rat renal microcirculation (Navar, Inscho, Majid, Imig, Harrison-Bernard & Mitchell, 1996). Besides obvious alterations – the hydronephrotic kidney is a non-filtering kidney devoid of tubular structures and the tubuloglomerular feedback mechanism – kidney-specific vascular responses are well preserved in this model, e.g. the myogenic vasodilatation in

response to renal perfusion pressure reduction (Steinhausen & Endlich, 1996). However, propagation of VMR in this model could differ to some extent from that in the normal kidney due to possible vascular alterations related to the reduced blood flow in this preparation.

Propagation of VMR in renal vessels may have important functional implications. Propagation smoothes VMR, prevents focal constriction and provides the basis for collective vascular responses in the kidney, e.g. vasomotion (Hollenberg *et al.* 1987; Steinhausen & Baehr, 1989; Loutzenhiser *et al.* 1990). Furthermore, the impact of the tubuloglomerular feedback mechanism on the autoregulation of renal blood flow may crucially depend on the upstream propagation of the VMR initiated at the distal part of the afferent arteriole (Carmines *et al.* 1990; Moore & Casellas, 1990; Øien & Aukland, 1991). On the other hand, if VMR are preferentially propagated in the downstream direction, as suggested by our results, myogenic activation of the interlobular artery will be powerfully transmitted to downstream vascular segments.

In summary, our study presents the first direct evidence for propagation of VMR in the renal vasculature *in vivo*. Length constants for the propagation of VMR are in the range of a few hundred micrometres, which permit physiologically important, but at the same time spatially confined, interaction between vascular segments and nephrons.

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Author's email address

M. Steinhausen: michael.steinhausen@urz.uni-heidelberg.de

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