In summary, for the use of local VF intervals as an index of local refractory periods, a sharp distinction must be made between normal and ischemic myocardium. To our knowledge, a relation between these parameters has not been established in ischemic myocardium. This is not surprising, because even assessment of refractory periods at one site at a time during ongoing ischemia is a cumbersome task because of the changing diastolic threshold of excitability. Finally, it may not be assumed that ischemia develops similarly in normal beating and fibrillating hearts.

Tobias Opthof, PhD
Ruben Coronel, MD
Department of Clinical and Experimental Cardiology
Academic Medical Center
University of Amsterdam
Amsterdam, Netherlands


Regional Hyperinsulinemia Does Not Modulate β-Adrenergic Vasodilator Action

To the Editor:

With regard to the recent interesting and well-written article by Lembo et al concerning the effect of insulin on the β-adrenergic vasodilator pathway, we would like to raise the following points: The authors conclude that insulin augments the β-adrenergic vasodilator response in the human forearm. This conclusion is mainly based on the observation that the response to intra-arterial infusions of isoproterenol was potentiated by concomitant intra-arterial insulin infusion. We feel that there may be a plausible alternative explanation for this main finding: Numerous groups have reported that insulin in itself, also when infused locally into the brachial or femoral artery, will induce a gradual increase in skeletal muscle blood flow. The combination of a gradual, slow-onset vasodilation of insulin itself together with an unaltered response to isoproterenol may well explain the observations reported by Lembo and associates. This combined effect may mimic a potentiating effect of insulin on isoproterenol-mediated vasodilation but in fact simply may be the sum of two independent vasodilator effects.

Although Lembo and associates claimed that 30 minutes of insulin infusion did not change blood flow, this does not exclude the possibility that vasodilation would occur later on, as the effect is slow in onset and continuous. Furthermore, there does exist an important interindividual variability, most probably related to the forearm muscle content. As the subjects in the study of Lembo had a relatively high mean body mass index (BMI) of just under 25 kg·m⁻², some may actually have been slightly obese and could hence show a decreased or attenuated insulin-induced vasodilator effect, stressing even more the need for prolonged evaluation of effects of insulin alone. Furthermore, the chance that the vasodilator effect of insulin may be initially missed as the result of a type I error may be very high, since the different study groups were small. This also may explain why the differences were not found in response to infusion of sodium nitroprusside before and during insulin. Unfortunately, results of pooled data of insulin on blood flow are not given.

Looking more in detail at Fig 2 of the article, it seems that after insulin itself (B2), a slight vasodilation already had occurred (according to Table 1, forearm blood flow [FBF] from 3.0±0.5 to 3.2±0.5). It must be emphasized that the absolute change in FBF is correlated to the baseline FBF, which means that a change in baseline would necessitate expression of the results in percentages above true baseline. The results of the experiments with isoproterenol in the study of Lembo et al would therefore better be expressed as percentage change from the second baseline.

We have performed similar experiments and confirmed the local vasodilatory effect of insulin in a large group of healthy subjects: After 30 minutes of intra-arterial infusion of insulin (leading to exactly comparable forearm venous insulin concentrations: 498±48 pmol·L⁻¹), FBF increased from 2.2±0.3 to 2.8±0.4 mL·dL⁻¹·min⁻¹ (n=27, P=.002). Mean percentage increase in blood flow was 38±11% (P=.001), but the variability was wide and ranged from -23% to +158%.

Furthermore, in experiments in which six dosages of isoproterenol (0.03, 0.1, 0.3, 1.0, 3.0, and 10 ng·dL⁻¹·min⁻¹) were infused into the brachial artery, before and after 60 minutes of concomitant insulin infusion, exactly identical vasodilator responses to isoproterenol were observed. Percentage increases in FBF after the six respective doses were 4.0±4.0%, 34.8±12.6%, 74.7±19.2%, 181.8±35.1%, 316.4±51.6%, and 498.1±95.5% before and 5.6±5.9%, 44.4±19.0%, 67.1±17.1%, 156.0±37.3%, 301.0±56.4%, and 492.8±161.3% during insulin infusion (12 healthy subjects; age, 22.3±2.1 years; BMI, 22.5±1.4 kg·m⁻²; P=.36) (see Figure). The results to our highest isoproterenol dose are shown in the Table.

<table>
<thead>
<tr>
<th>Isoproterenol (ng/dL/min)</th>
<th>FBF (% Increase)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.03</td>
<td>4.0±4.0%</td>
</tr>
<tr>
<td>0.1</td>
<td>34.8±12.6%</td>
</tr>
<tr>
<td>0.3</td>
<td>74.7±19.2%</td>
</tr>
<tr>
<td>1</td>
<td>181.8±35.1%</td>
</tr>
<tr>
<td>3</td>
<td>316.4±51.6%</td>
</tr>
<tr>
<td>10</td>
<td>498.1±95.5%</td>
</tr>
</tbody>
</table>

Effect of six increasing doses of isoproterenol on percentage increase in FBF before and during hyperinsulinemia.
dose were much larger than the responses reported in Lembo and colleagues' study despite a higher isoproterenol dose in that study. These striking differences are not well explained.

In conclusion, we do not agree with the conclusions of Lembo and associates and suggest that the increased response to isoproterenol during insulin could be explained simply by a still-ongoing intrinsic vasodilator effect of insulin added to the response to isoproterenol. In fact, the same explanation could hold true for the diminished effect of sympathetic stimulation during insulin and BHT-933. As such, we believe there is currently not sufficient evidence for a specific modulator effect of insulin on $\beta$-adrenergic vasodilator action.

Cees J.J. Tack, MD
Division of General Internal Medicine
Department of Medicine

Paul Smits, MD, PhD
Department of Pharmacology
University Hospital Nijmegen
Nijmegen, Netherlands


Response

Drs Tack and Smits give a different interpretation of our results,1 mentioning a series of studies in which they assert that a vasodilator effect of insulin infused locally was reported. Actually, a careful perusal of these studies reveals that the citations were not entirely appropriate. In particular, in the studies from Steinberg et al2 and Baron et al3 insulin was administered systemically during a euglycemic clamp of long duration (about 3 hours) and not directly into the artery. The same systemic insulin infusion in euglycemic conditions was used in the Utriainen et al investigations.4 Furthermore, in the Creager et al study,6 insulin was infused locally in a dose range from 0.1 to 1.0 mU/kg per minute, but the difference in FBF was observed only at the highest level of intrabrachial insulin infusion rate, which obviously exposed the forearm to elevated pharmacological amounts of the hormone. High insulin levels (104 $\mu$U/mL) were reached in the Nehring et al study,6 which also claimed an insulin spillover into the systemic circulation, as demonstrated by the increase in plasma insulin levels in the contralateral arm. Finally, Louard et al7 unequivocally demonstrated that insulin augments FBF (25%) only at a dose yielding deep venous insulin concentration of 125 $\mu$U/mL,7 about twofold higher than in our study.

In contrast, several studies8,9 have clearly demonstrated that insulin infusion directly into the brachial artery does not increase blood flow. In particular, at lower doses yielding deep venous insulin levels comparable to those reached in our study, no increase in FBF is observed when the infusion of the hormone is extended for longer periods.9,10 However, to satisfy the doubts of Tack and Smits, we have pooled our data and, confirming the results from the studies cited above, we were unable to see any impressive effect of insulin on FBF (3.02±0.2 versus 2.95±0.1 mL/min per 100 mL, n=24; NS). Thus, in this circumstance, their recommendation to express the results as a percentage of the baseline does not appear extremely appropriate.

Therefore, the authors must consider that their novel hypothesis, based on the fact that insulin directly infused into brachial artery evokes vasodilation, needs to be reexamined because such an insulin vasodilator effect is not uniformly observed.

Our conclusions regarding the cross talk between insulin and $\beta$-adrenergic receptor signal transduction pathways at the vascular level were also attested to by a distinct series of studies in which we observed that propranolol, a selective $\beta$-adrenergic-blocking agent, was able to partially restore the reflex sympathetic vascular response, counteracting the modulatory effect of insulin.11 However, supposing that insulin is concretely able to exert a non-specific potentiating effect of the $\beta$-adrenergic—evoked vasodilation through its independent vasodilator action, it is difficult for us to interpret the results obtained with sodium nitroprusside, in which, in contrast, insulin was unable to sensitize that response. Tack and Smits have their personal interpretation for this result also: They claim that we have not studied enough subjects. Thus, it is possible that in a larger group, the insulin-potentiating effect on sodium nitroprusside might be indubitably manifest. Probably they are not informed that Taddei et al12 have recently demonstrated that insulin, locally infused in a dose and time course similar to those used in our study, were unable to modify the response of sodium nitroprusside, entirely supporting our findings and the conclusion that the insulin-potentiating effect on $\beta$-adrenergic vasodilator pathways was not due to a non-specific independent vasorelaxant action of the hormone. Furthermore, Tack and Smits also speculate that an intrinsic vasodilator effect of insulin may account for the diminished effect of sympathetic stimulation during insulin and BHT-933 infusion. However, even in this case, we cannot agree with their interpretation because in our previous studies13 using phenylephrine to evoke a distinct adreno-receptor-mediated vascular effect, we were unable to disclose an insulin effect on phenylephrine-induced vasoconstriction, further sustaining the concept that the insulin effect on the vascular $\alpha_1$- and $\beta$-adrenergic pathways must be considered specific. On this issue, we would like to quote recent basic studies in which has been clearly demonstrated the ability of insulin to increase $\beta$-adrenergic responsiveness,14 and the exact molecular mechanism has been proposed also.15

It remains to be explained why Tack and Smits have attained different results. Actually, this point is more difficult for us, since we did not have an opportunity to preview their manuscript before its publication. However, on the basis of their scant information, we can only speculate that prolonged infusion of insulin into the brachial artery may result in a spillover of the hormone into the systemic circulation, which may recruit a sympathomediatory mechanism.16

Giuseppe Lembo, MD, PhD
IRCCS Istituto Neurologico
Pozzilli (IS)