Endurance or Sprint Interval Exercise, & Metformin Treatment Differently Modify Insulin-Induced Vasodilation in Skeletal Muscle Arterioles of Obese Insulin Resistant Rats

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1. ABSTRACT

Insulin-induced vasodilation is obligatory for glucose disposal, and is impaired with insulin resistance. We examined the effects of endurance (EXT) and interval sprint (IST) exercise training with and without metformin (MET) treatment on acetylcholine (ACh) and insulin-induced vasodilation in skeletal muscle arterioles of high and low oxidative muscle from obese, insulin resistant OLETF rats. Rats remained sedentary (SED), or were treated with EXT, IST, MET, EXT+MET, or IST+MET from 20-32wks (n=11-13). At sacrifice, 2nd order arterioles from red (G2AR) and white (G2AW) gastrocnemius muscle were isolated for *in vitro* assessment of vasomotor responses to ACh (10^{-9}-10^{-4} M), insulin (1-1000 μIU/mL), and insulin (1-1000 μIU/mL) + tezosentan (ET-1 antagonist; 3 μM). EXT and IST enhanced ACh responses in both G2AR and G2AW to a greater extent than MET alone (all $p<0.05$). In the G2AR, EXT improved insulin-induced vasodilation compared to IST, while MET was greater than IST alone (all $p<0.05$). ET-1 blockade improved insulin-induced vasodilation in IST compared to EXT, and MET in the G2AW, whereas IST+MET exhibited less vasodilation compared to IST (all $p<0.05$). In conclusion, the effects of EXT and IST on insulin-induced vasodilation in 2A arterioles of low and high oxidative muscle fibers are similar; but the mechanisms appear to be different. EXT selectively improved insulin-induced vasodilation in G2AR, and was not affected by ET-1 blockade, suggesting it is mediated by nitric oxide. Conversely, no insulin-induced vasodilation was observed in the G2AW following IST, yet a marked vasodilation occurred with insulin + ET-1 blockade. Overall these data suggest that the type of exercise training, and treatment with MET, have differential effects on ACh- and insulin-induced vasodilation in skeletal muscle arterioles perfusing high vs. low oxidative muscle fibers in the obese, insulin resistant OLETF rat.

2. INTRODUCTION

A key contributor to insulin-mediated glucose uptake is insulin-induced vasodilation of skeletal muscle arterioles, which is impaired with obesity and Type 2 diabetes (T2D). Abnormalities in the vascular reactivity to insulin can limit perfusion, and delivery of glucose and insulin to muscle tissue. In human patients with T2D, exercise improves insulin sensitivity and glucose uptake T2D (1). Furthermore, we have previously shown that daily exercise prevents impairments in insulin-induced vasodilation in OLETF rats (2). However, the efficacy of exercise interventions which utilize different muscle recruitment patterns (i.e. aerobic vs. sprint training) to ameliorate or reverse impairments in microvascular insulin reactivity has not be elucidated.

OBJECTIVE: The current ADA standard of care for T2D is treatment with metformin in combination with a diet and exercise program (3). Therefore, we studied the effects of endurance exercise and interval sprint training, with and without metformin on the vasoreactivity to insulin in skeletal muscle arterioles from red and white muscles.

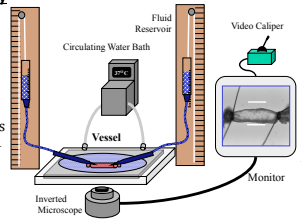
3. METHODS

Otsuka Long Evans Tokushima Fatty (OLETF) rats:

- Mutant CCK Receptor
- Hyperphagic/obese
- Insulin resistant by 12-13 wks
- Type 2 Diabetes by 30-40 wks

We assessed *in-vitro* vasomotor responses to ACh, Insulin, Insulin + Tezosentan (ET-1 blockade), & SNP.

Statistics: We used a repeated measures ANOVA, with post hoc tests for simple effects of group for the dose responses curves with a correction for multiple comparisons (P=0.0167); and one way ANOVAs, with Tukey post-hoc tests, for body composition and HbA1c.



Six groups of OLETF rats:

1. **EXT-** (n=11)- From 20-30wks: treadmill running *30 m/min, (15 % grade), 60 min, 5 d/wk.*
2. **IST-** (n=12)- From 20-30wks: treadmill run *2.5 min 60m/min (15 % grade), 4.5 min rest– x6 bouts, 5 d/wk.*
3. **SED-** (n=13)- 30wks sedentary (type 2D).
4. **SED+MET-** (n=12)-Metformin treatment 20-30 wks.
5. **EXT+MET-** (n=12)-Combined treatment 20-30 wks.
6. **IST+MET-** (n=12)- Combined treatment 20-30 wks.

4. RESULTS

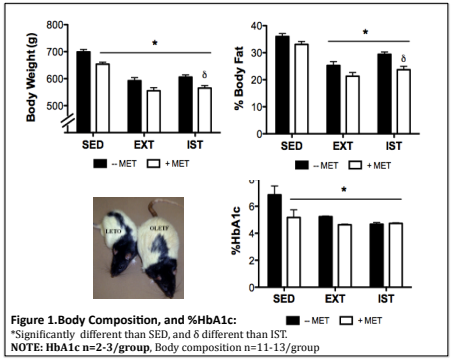


Figure 1.Body Composition, and %HbA1c: *Significantly different than SED, and δ different than IST.
NOTE: HbA1c n=2-3/group, Body composition n=11-13/group

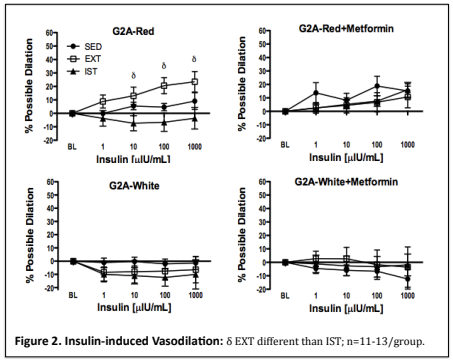


Figure 2. Insulin-induced Vasodilation: δ EXT different than IST; n=11-13/group.

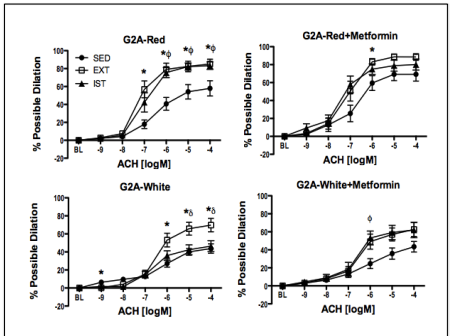


Figure 4. ACh Induced Vasodilation: * EXT different than SED, Φ IST different than SED, and δ EXT different than IST; n=11-13/group.

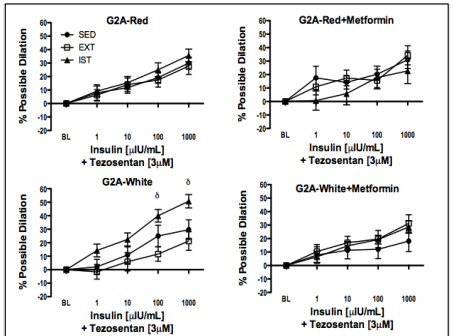


Figure 3. Insulin-induced Vasodilation + ET-1 Blockade: δ EXT different than IST; n=11-13/group.

5. SUMMARY

Body Composition and HbA1c (Fig. 1):

- Body weight was reduced in all treatment groups, and %body fat was decreased in all exercise rats.
- Metformin & interval sprint training reduced body weight and %body fat more than interval sprint training alone.
- HbA1c was reduced in all treatment groups.

Insulin-Induced Vasodilation, & ET-1 Blockade (Fig. 2 & 3):

- Endurance exercise selectively improved insulin-induced vasodilation in arterioles perfusing the high oxidative, red portion of the gastrocnemius muscle.
- ET-1 blockade normalized differences between groups, suggesting that improvements in the dilatory effects of insulin with exercise are ET-1 dependent.
- Effects of interval sprint training on insulin-induced vasodilation in the white portion of the gastrocnemius, only occurred with ET-1 blockade, suggesting that ET-1 vasoconstriction prevented insulin-induced vasodilation in arterioles perfusing low oxidative muscle.
- Effects of exercise on insulin-induced vasodilation were mainly abolished in rats treated with Metformin.

Ach-Induced Vasodilation (Fig. 4):

- Endurance exercise and interval sprint training improved ACh-induced vasodilation in arterioles perfusing the high oxidative red portion of the gastrocnemius muscle; whereas improvements in the low oxidative white portion of the muscle was only found with endurance exercise.

6. CONCLUSIONS

Insulin-induced vasodilation in 2nd order arterioles perfusing the high oxidative, red portion of the gastrocnemius muscle appear to be most responsive to exercise. However, these effects are largely dependent on the mode of exercise and absent when animals are concomitantly treated with Metformin.

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REFERENCES

1. **Kirwan JP, et al.** *Am J Physiol-Endoc M*, 2009.
2. **Mikus CR, et al.** *J Appl Physiol* , 2010.
3. **American Diabetes Association**. *Diabetes Care*, 2011.