# Similar improvements in 5-km performance and maximal oxygen uptake with submaximal and maximal 10-20-30 training in runners, but increase in muscle oxidative phosphorylation occur only with maximal effort training 

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#### Abstract

Objective: The aim of the present study was to examine whether 10-20-30 training (consecutive 1 -min intervals consisting of 30 s at low-speed, 20 s at moderatespeed, and 10 s at high-speed), performed with submaximal effort during the 10-s high-speed runs, would lead to improved performance as well as increased maximum oxygen uptake ( $\mathrm{VO}_{2}$-max) and muscle oxidative phosphorylation (OXPHOS). In addition, to examine to what extent the effects would compare to 10-20-30 running conducted with maximal effort. Design: Nineteen males were randomly assigned to 10-20-30 running performed with either submaximal (SUBMAX; $n=11$ ) or maximal (MAX; $n=8$ ) effort, which was conducted three times/week for 6 weeks (intervention; INT). Before and after INT, subjects completed a 5 -km running test and a $\mathrm{VO}_{2}$-max test, and a biopsy was obtained from m . vastus lateralis. Results: After compared to before INT, SUBMAX and MAX improved ( $p<0.05$ ) 5 -km performance by $3.0 \%$ ( $20.8 \pm 0.4$ (means $\pm$ SE) vs. $21.5 \pm 0.4 \mathrm{~min}$ ) and $2.3 \%$ ( $21.2 \pm 0.4 \mathrm{vs}$. $21.6 \pm 0.4 \mathrm{~min}$ ), respectively, and $\mathrm{VO}_{2}$-max was $\sim 7 \%$ higher ( $p<0.01$ ) in both SUBMAX ( $57.0 \pm 1.3 \mathrm{vs} .53 .5 \pm 1.1 \mathrm{~mL} / \mathrm{min} / \mathrm{kg}$ ) and MAX ( $57.8 \pm 1.2 \mathrm{vs}$. $53.7 \pm 0.9 \mathrm{~mL} / \mathrm{min} / \mathrm{kg}$ ), with no difference in the changes between groups. In SUBMAX, muscle OXPHOS was unchanged, whereas in MAX, muscle OXPHOS subunits (I-IV) and total OXPHOS ( $5.5 \pm 0.3$ vs $4.7 \pm 0.3$ A.U.) were $9 \%-29 \%$ higher ( $p<0.05$ ) after compared to before INT. Conclusion: Conducting 10-20-30 training with a non-maximal effort during the 10 -s high-speed runs is as efficient in improving 5 -km performance and $\mathrm{VO}_{2}$ max as maximal effort exercise, whereas increase in muscle OXPHOS occur only when the $10-\mathrm{s}$ high-speed runs are performed with maximal effort.


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## KEYWORDS

muscle oxidative enzymes, running economy, speed endurance training

## 1 | INTRODUCTION

In the past $\sim 15$ years, the effect of speed endurance training (SET), defined as repeated bouts of $5-40$-s maximalintensity exercise with rest periods greater than five times the exercise bouts, ${ }^{1}$ has been found to improve performance of $10-50 \mathrm{~min}$ events in both untrained ${ }^{2}$ and trained subjects. ${ }^{3-5}$ In recent years, the effect of interval training switching between low-, moderate-, and high-intensity has also been studied..$^{6-8}$ Thus, $10-20-30$ training, consisting of consecutive $1-\mathrm{min}$ intervals of 30 s of low, 20 s of moderate and 10 s of high-intensity exercise, has been shown to improve performance of untrained and trained subjects. ${ }^{9-12}$ Specifically, 10-20-30 training of recreational runners has been demonstrated to improve $5-\mathrm{km}$ performance by $4 \%^{7}$ and intermittent endurance exercise performance of elite soccer players by $18 \%{ }^{13}$ During 10-20-30 training heart rate (HR) reach $90 \%-95 \%$ of maximal $\mathrm{HR},{ }^{7}$ and maximum oxygen consumption $\left(\mathrm{VO}_{2}\right.$-max) has been shown to increase during a period of 10-20-30 training both in not previously trained ${ }^{9,11}$ and in trained ${ }^{7,8}$ subjects, despite a reduced training volume. In these studies, the subjects exercised with maximal effort during the 10 -s high-speed runs, and it is unclear whether similar effects can be obtained with intense, but not maximal effort, during the $10-\mathrm{s}$ high-speed runs.

SET has been shown to elevate expression of muscle proteins involved in ion transport, such as the sodium/potassium pump $\left(\mathrm{Na}^{+} / \mathrm{K}^{+}\right)$isoforms ${ }^{2,3,14-16}$ and the sodium/ proton exchanger 1 (NHE1) ${ }^{2,14,16,17}$ as well as muscle oxidative capacity. ${ }^{16,18,19}$ Also, 10-20-30 training has been found to increase muscle ion handling proteins in subelite football players ${ }^{13}$ and in men with Type 2 diabetes. ${ }^{10}$ It is, however, unclear whether 10-20-30 training causes change in muscle oxidative enzymes and whether a submaximal version of the 10-20-30 training leads to muscle adaptations in recreational runners.

Thus, the aim of the present study was to examine whether 10-20-30 training, conducted at a submaximal running speed during the 10 -s high-speed running, would lead to improved performance and increased $\mathrm{VO}_{2}$-max in recreational runners, and to what extent the effects would compare to training conducted with maximal effort. In addition, to study the effect of 10-20-30 training on muscle oxidative enzymes.

## 2 | METHODS

## $2.1 \mid$ Subjects

Nineteen male recreational runners with an age, height, body mass, and $\mathrm{VO}_{2}$-max of $27.2 \pm 4.0$ (means $\pm$ standard deviation [SD]) years, $182.9 \pm 5.6 \mathrm{~cm}, 77.9 \pm 7.1 \mathrm{~kg}$, $53.6 \pm 0.7 \mathrm{~mL} / \mathrm{min} / \mathrm{kg}$, respectively, completed the study. After receiving written and oral information about the study and the possible risks and discomforts associated with the experimental procedures, all subjects gave their written informed consent to participate. The study conformed to the Code of Ethics of the World Medical Association (Declaration of Helsinki) and was approved by the Ethics Committee of the capital region of Copenhagen (Region Hovedstaden; H-17007822).

## 2.2 | Experimental design

Before being included in the study, subjects performed a $5-\mathrm{km}$ running test on an outdoor $400-\mathrm{m}$ running track at Østerbro Stadium, Copenhagen, and an incremental treadmill test to exhaustion (INC) at the laboratory at the Department of Nutrition, Exercise and Sports (NEXS), University of Copenhagen to determine $\mathrm{VO}_{2}$-max. These tests also served as familiarization to the tests conducted in the study. Next, the subjects were randomly assigned, based on 5 -km performance and $\mathrm{VO}_{2}$-max, to $10-20-30$ training performed with either maximal (MAX; $n=8$ ) or submaximal (SUBMAX; $n=11$ ) effort (see below). The subjects completed a 6 -week intervention period (INT) with two experimental days before and after INT.

### 2.3 Experimental days

Tests and sampling procedures were performed at the same time of day for each subject and on separate days interspersed by at least 48 h in either the laboratory at NEXS, University of Copenhagen, or at a $400-\mathrm{m}$ running track. Before the tests, a Polar Team ${ }^{2}$ HR monitor (Polar Electro Oy) was fitted around the chest of the subjects for continuous HR recordings. The subjects refrained from physical activity, alcohol, painkillers, and caffeine 24 h before testing, and were instructed to keep a diary journal in the last

2 days before the first series of tests, and to replicate the diet before being tested again after INT.

On the first experimental day, the subject arrived at the laboratory between 7 and 11 a.m. after an overnight fast; a catheter was inserted in an antecubital vein and a blood and muscle sample (see below) was collected at rest. Next, subjects performed two bouts of 6 -min running at $12 \mathrm{~km} / \mathrm{h}$ on a treadmill separated by 20 min of rest, and the average of the measurements obtained during the two bouts was used to increase the validity of the measure. The second 6-min running bout was followed by INC, where the speed was increased to $14 \mathrm{~km} / \mathrm{h}$ for 1 min and then by $1 \mathrm{~km} / \mathrm{h}$ every minute until exhaustion. Respiratory measurements were obtained in the last 2 min of the 6 -min bouts and continuously during INC by use of Oxycon Pro (Viasys Healthcare), which was calibrated prior to each measurement. In addition, a blood sample was collected immediately after each of the two 6 -min running bouts, as well as immediately after and 3 min after INC. During the last part of INC, the subjects were verbally encouraged to continue their effort until voluntary termination of the test.

On the second experimental day, a $5-\mathrm{km}$ running test was completed at a $400-\mathrm{m}$ running track after a $10-\mathrm{min}$ warm-up period consisting of $5-\mathrm{min}$ of moderate-intensity continuous running, dynamic stretches, various activities such as butt kicks and skips as well as two $\sim 100-\mathrm{m}$ runs where speed was gradually increased to reach nearmaximum speed. Subjects did not receive any verbal encouragement and did not wear a watch or listen to any audio during the test.

Before the first and final 10-20-30 training session, the subjects performed three maximal $60-\mathrm{m}$ sprints, separated by 1-min rest periods. The running speed was measured by GPS based tracking units (Catapult MinimaxV4, Catapult Innovations) worn on the upper part of the torso by the subjects. The highest speed obtained was defined as maximal speed.

## 2.4 | Training

Before being included in the study, the subjects were running at moderate speed two to three times a week for 20-40 min. During INT, the subjects only performed 10-20-30 training, which was carried out as previously described. ${ }^{7}$ In brief, a 10-20-30 training session consisted of two to four $5-6 \mathrm{~min}$ bouts of running interspersed by $2-3 \mathrm{~min}$ of rest. In each bout, the subjects carried out five to six consecutive 1-min intervals of 30-s at low-speed $(\sim 30 \%$ of maximum speed $/ \sim 8 \mathrm{~km} / \mathrm{h} / \mathrm{i} . \mathrm{e}$, jogging), followed by 20 s at moderate-speed ( $40 \%-50 \%$ of maximum speed/11-14 km/h/i.e., regular continuous running pace) and lastly 10 s of high-speed running ( $74 \%-84 \%$ of maximum speed; Figure 1). While there was no difference between the low- and moderate-speed of the two groups, the high-speed runs were performed with either maximal (MAX) or submaximal effort (SUBMAX). Thus, MAX ran longer than SUBMAX during a 10-s high-speed run. So, to ensure that the total volume of high-speed running was the same in the two groups,


FIGURE 1 Example of relative heart rate (\%max) and velocity (\%max) during three bouts of 10-20-30 training with submaximal (SUBMAX; red lines) or maximal (MAX; blue lines) exercise intensity.

MAX and SUBMAX conducted five and six 10-20-30 intervals, respectively. As seen on Figure 1, peak speed during the 10 -s high-speed runs in MAX declined over the course of the bout ultimately resulting in an average speed of $84 \%$ of maximal speed even though effort (i.e., perceived exertion) was maximal in each of the highspeed runs. In comparison, peak speed during the $10-\mathrm{s}$ high-speed runs in SUBMAX were kept at $\sim 75 \%$ of maximal speed.

Training was performed on Mondays, Wednesdays, and Fridays at an outdoor $400-\mathrm{m}$ running track. A standardized 10 -min warm-up was conducted for each training session consisting of $5-\mathrm{min}$ jogging, followed by dynamic stretches and various activities, as well as two $\sim 100-\mathrm{m}$ runs where speed was gradually increased to reach nearmaximum speed.

During INT the subjects completed 18 sessions of 10-20-30 training. All training sessions were supervised. If the subjects were unable to participate in the supervised training, they performed the training on their own. To obtain a progressive training load, the first, second and third training session consisted of two, three and four 5- to 6min bouts, respectively, separated by two (SUBMAX) or three (MAX) min of rest. Then, the subjects interchanged between doing three or four 5 - to $6-\mathrm{min}$ bouts of 10-20-30 for the remainder of INT.

For all training sessions during INT, subjects were wearing GPS and HR monitors.

## 2.5 | Blood sampling and analysis

Blood samples were stored on ice until being analyzed for pH , lactate, and $\mathrm{K}^{+}$(ABL800 Flex; Radiometer Medical).

## 2.6 | Muscle biopsy and analysis

Using the Bergstrom procedure, ${ }^{20}$ a biopsy was collected with a $5-\mathrm{mm}$ needle from a standardized depth of 5 cm at the middle of m . vastus lateralis of the right leg at rest using local anesthesia ( $1 \mathrm{~mL} ; 20 \mathrm{mg} / \mathrm{L}$ lidocaine without adrenaline). The muscle sample ( $\sim 100 \mathrm{mg}$ wet weight) was immediately frozen in liquid $\mathrm{N}_{2}$ and stored at $-80^{\circ} \mathrm{C}$ until further analysis.

Muscle western blot analysis were performed to determine protein expression as described previously. ${ }^{17}$ In short, $\sim 2.5 \mathrm{mg}$ dry weight (dw; freeze-dried for a minimum of 24 h ) of each muscle sample was dissected free from blood, fat, and connective tissue. Samples were homogenized for $3 \times 30-\mathrm{s}$ periods at 28.5 Hz
(Qiagen Tissuelyser II; Retsch) in a fresh batch of icecold buffer containing (in mM) $10 \%$ glycerol, 20 Napyrophosphate, $150 \mathrm{NaCl}, 50 \mathrm{HEPES}(\mathrm{pH} 7.5), 1 \% \mathrm{NP}-40$, $20 \beta$-glycerophosphate, $2 \mathrm{Na}_{3} \mathrm{VO}_{4}, 10 \mathrm{NaF}, 2 \mathrm{PMSF}, 1$ EDTA (pH 8), 1 EGTA (pH 8), $10 \mu \mathrm{~g} / \mathrm{mL}$ aprotinin, $10 \mu \mathrm{~g} /$ mL leupeptin, and 3 benzamidine, after which they rotated for 1 h at $4^{\circ} \mathrm{C}$, and centrifuged at 18320 g for 20 min at $4^{\circ} \mathrm{C}$ to exclude non-dissolved structures. The supernatant (lysate) was collected and used for further analysis. Total protein concentration in each sample was determined by a BSA standard kit (Thermo Scientific), and samples were mixed with $6 \times$ Laemmli buffer ( 7 mL $0.5 \mathrm{~mol} / \mathrm{L}$ Tris-base, 3 mL glycerol, 0.93 g DTT, 1 g SDS, and 1.2 mg bromophenol blue) to reach equal protein concentration before protein expression was determined by western blotting.

Equal amounts of total protein were loaded in each well of precast gels (Millipore). All samples from each subject were loaded on the same gel. Proteins were separated according to their molecular weight by SDSPAGE and semi-dry transferred to a $0.45 \mu \mathrm{~m}$ PVDF membrane (Bio-Rad). The membranes were blocked in either $2 \%$ skimmed milk or $3 \%$ BSA in TBST, including $0.1 \%$ Tween -20 before an overnight rocking incubation in primary antibody at $4^{\circ} \mathrm{C}$. The membranes were then incubated for 1 h at room temperature in horseradish peroxidase conjugated secondary antibody (dependent on the primary antibody source).

The protein bands were visualized with ECL (Millipore) and recorded with a digital camera (ChemiDoc MP Imaging System, Bio-Rad Laboratories). For each muscle sample, protein expression was determined in duplicate on individual gels. Quantification of the band intensity was performed using Image Lab version 4.0 (Bio-Rad Laboratories). Each band was normalized to total protein content of each sample from the stain-free image as previously described. ${ }^{21}$

## 2.7 | Calculations

Running economy (RE) was calculated using the formula:

$$
\begin{aligned}
& \mathrm{RE}\left(\mathrm{mLO}_{2} / \mathrm{kg} / \mathrm{km}\right)=\mathrm{VO}_{2}(\mathrm{~mL} / \mathrm{min}) \\
& \quad \times 60 \mathrm{~min} / \mathrm{h} / \text { body mass }(\mathrm{kg}) \times \text { running speed }(\mathrm{km} / \mathrm{h})
\end{aligned}
$$

where $\mathrm{VO}_{2}$ is the average value during the last 2 min of the two $6-\mathrm{min}$ bouts at the running speed of $12 \mathrm{~km} / \mathrm{h}$.

During INC, $\mathrm{VO}_{2}$-max was determined as the highest average value achieved over a 30 -s period, ${ }^{22}$ with attaining
maximal HR (compared to the screening test) and a respiratory exchange ratio (RER) value $>1.15$ used as criterions. During INC, peak running speed ( $V_{\text {peak }}$ ) was calculated as:

$$
V_{\text {peak }}=V_{\mathrm{f}}(\mathrm{~km} / \mathrm{h})+\left(T_{\mathrm{i}} / 60 \times 1 \mathrm{~km} / \mathrm{h}\right)
$$

where $V_{\mathrm{f}}(\mathrm{km} / \mathrm{h})$ is the final velocity obtained and $T_{\mathrm{i}}(\mathrm{s})$ is the time spent at the final speed level.

## 2.8 | Statistics

Data were evaluated using two-way ANOVAs with repeated measures and the level of significance was set at $p<0.05$. A Student-Newman Keuls post hoc test was applied in case significance was reached in the ANOVA. Absolute data values are used and presented as means $\pm$ standard error of the mean (SE) unless otherwise stated. Sample size was determined based on the predicted effect of $10-20-30$ training on $5-\mathrm{km}$ performance. ${ }^{7}$

## 3 | RESULTS

## 3.1 | Distance, HR and running speed during training

In SUBMAX, mean distance covered during a 10-20-30 bout was $1233 \pm 13 \mathrm{~m}$, which was longer $(p<0.05)$ than in MAX $(1097 \pm 12 \mathrm{~m})$, but with the distance covered during the 10 -s high-speed runs being equal. Mean HR during a $10-20-30$ bout was $83 \pm 0.5 \%$ and $81 \pm 0.8 \%$ in SUBMAX
and MAX, respectively, with no difference between groups.

In SUBMAX, mean peak velocity during the 10-s highspeed runs was $20.9 \pm 0.1 \mathrm{~km} / \mathrm{h}(73.9 \pm 0.4 \%$ of maximal speed) and lower $(p<0.05)$ than in MAX $(23.3 \pm 0.2 \mathrm{~km} / \mathrm{h}$; $84.4 \pm 0.5 \%$ of maximal speed).

## 3.2 | 5-km performance

5-km performance was better $(p<0.05)$ after compared to before INT in both SUBMAX $(3.0 \%, 20.8 \pm 0.4$ vs. $21.5 \pm 0.4 \mathrm{~min}$ ) and MAX ( $2.3 \%, 21.2 \pm 0.4$ vs. $21.6 \pm 0.4 \mathrm{~min}$ ) with no difference in the change between groups (Figure 2; Table 1).

## 3.3 | Maximal measurements

$V_{\text {peak }}$ during INC was higher $(p<0.01)$ after than before INT in both SUBMAX $(1.9 \%, 19.0 \pm 0.3$ vs $18.6 \pm 0.3 \mathrm{~km} / \mathrm{h})$ and MAX $(4.2 \%, 18.8 \pm 0.2$ vs. $18.1 \pm 0.2 \mathrm{~km} / \mathrm{h})$, with the increase being less $(p<0.05)$ in SUBMAX than MAX (Table 1).
$\mathrm{VO}_{2}$-max was higher $(p<0.01)$ after than before INT in both SUBMAX ( $6.4 \%, 57.0 \pm 1.3$ vs. $53.5 \pm 1.1 \mathrm{~mL} / \mathrm{min} / \mathrm{kg}$ ) and MAX $(7.5 \%, 57.8 \pm 1.2$ vs. $53.7 \pm 0.9 \mathrm{~mL} / \mathrm{min} / \mathrm{kg})$ with no difference in the change between groups (Table 1).

Plasma $\mathrm{K}^{+}$immediately after and 3 min after exhaustion, was not different in SUBMAX, whereas in MAX, plasma $\mathrm{K}^{+}$was lower $(p<0.05) 3 \mathrm{~min}$ after exhaustion after compared to before INT. Blood lactate and pH did not change in neither SUBMAX nor MAX (Table 2).

■SUBMAX ロMAX


FIGURE 2 Performance during the 5-km running test before (Pre) and after (Post) 6weeks of 10-20-30 training with submaximal (SUBMAX; black bars) or maximal (MAX; white bars) exercise intensity. *Different ( $p<0.05$ ) from Pre.

|  | SUBMAX |  | MAX |  |
| :---: | :---: | :---: | :---: | :---: |
|  | Pre | Post | Pre | Post |
| 5-km (min) | $21.5 \pm 0.4$ | $20.8 \pm 0.4^{*}$ | $21.6 \pm 0.4$ | $21.2 \pm 0.4^{*}$ |
| $V_{\text {peak }}(\mathrm{km} / \mathrm{h})$ | $18.6 \pm 0.3$ | $19.0 \pm 0.3^{*}$ | $18.1 \pm 0.2$ | $18.8 \pm 0.2^{*}$ |
| $\mathrm{VO}_{2}$-max ( $\mathrm{mL} \mathrm{O} 2 / \mathrm{min} / \mathrm{kg}$ ) | $53.5 \pm 1.1$ | $57.0 \pm 1.3^{*}$ | $53.7 \pm 0.9$ | $57.8 \pm 1.2^{*}$ |
| $\mathrm{VO}_{2}$-max (mL O2/min) | $4053 \pm 100$ | $4270 \pm 95^{*}$ | $4330 \pm 136$ | $4618 \pm 193 *$ |

Note: Values are means $\pm \mathrm{SE} . \mathrm{VO}_{2}$-max, maximal oxygen consumption; $V_{\text {peak }}$, peak running speed during INC. See Training for description of 10-20-30 training.
*Different ( $p<0.05$ ) from Pre.

TABLE 1 Performance during the $5-\mathrm{km}$ running test, $\mathrm{VO}_{2}$-max and Vpeak before (Pre) and after (Post) 6 weeks of 10-20-30 training with submaximal (SUBMAX) or maximal (MAX) exercise intensity.

## 3.4 | Sub-maximal measurements

$\mathrm{VO}_{2}, \mathrm{RE}$ and RER during running at $12 \mathrm{~km} / \mathrm{h}$ did not change during INT in neither SUBMAX nor MAX (Table 3).

In SUBMAX, plasma $\mathrm{K}^{+}$, blood lactate and pH during submaximal running did not change during INT, whereas in MAX, plasma $\mathrm{K}^{+}$was lower $(p<0.05)$ after both bouts of submaximal running, and blood lactate was lower ( $p<0.05$ ) after the second bout of submaximal running after compared to before INT (Table 2).

## 3.5 | Muscle oxidative protein expression

Muscle oxidative phosphorylation (OXPHOS) complex I-V and total OXPHOS were unchanged in SUBMAX, whereas in MAX, muscle OXPHOS complex I-IV and total OXPHOS were higher $(p<0.05)$ after compared to before INT $(15 \pm 6 \%, 24 \pm 11 \%, 29 \pm 10 \%, 9 \pm 3 \%$, and $17 \pm 7 \%$, respectively) (Table 4).

## 4 DISCUSSION

The major finding of the present study was, that a group of recreational runners, conducting 10-20-30 training with submaximal effort during the 10 -s high-speed runs, improved $5-\mathrm{km}$ performance and increased $\mathrm{VO}_{2}$-max to a similar extent as the group exercising with maximal effort during the $10-\mathrm{s}$ high-speed runs. In addition, only the group that performed the training with maximal effort had an increase in muscle oxidative enzymes and it had a more marked improvement in peak running speed during the incremental test compared to the submaximal group.

Both the SUBMAX and MAX group had an increase of $\sim 3 \%$ in $5-\mathrm{km}$ performance. In accordance, Gunnarsson et al. ${ }^{7}$ studied 10-20-30 training in recreational runners and found $4 \%$ improvement in $5-\mathrm{km}$ performance with 7 weeks of training three times per week and a $54 \%$ reduction in training volume. Likewise, Gliemann et al. ${ }^{8}$
observed a $3 \%$ increase in 5 -km performance with 8 weeks of 10-20-30 training twice a week, and Faelli et al. ${ }^{23}$ found an improvement of $2.5 \%$ in a $1-\mathrm{km}$ run after 8 weeks of two 10-20-30 training sessions and one continuous training session a week with $25 \%$ reduction in training volume. The novel finding in the present study is that the improvement in performance can be achieved even with nonmaximal effort during the $10-\mathrm{s}$ high-speed runs.

Also, maximum oxygen uptake increased to a similar extent in SUBMAX and MAX $(6.4 \%$ and $7.5 \%$, respectively). Likewise, Gunnarsson et al. ${ }^{7}$ and Gliemann et al. ${ }^{8}$ observed increases in $\mathrm{VO}_{2}$-max of $4 \%$ and $3 \%$, respectively, whereas the increase in the study by Faelli et al. ${ }^{23}$ was higher (about $8 \%$ ), which may be due to the subjects having a low $\mathrm{VO}_{2}-\max (43 \mathrm{~mL} / \mathrm{min} / \mathrm{kg})$ at the start of the intervention. The higher $\mathrm{VO}_{2}$-max after a period with $10-$ 20-30 training may be explained by the subjects during training reaching peak HRs higher than $90 \%$ of maximum HR even with non-maximal effort during the 10-s highspeed runs (Figure 1), which is higher than during their normal training. ${ }^{7}$ Also, a period of SET has been shown to increase $\mathrm{VO}_{2}$-max in recreational active males $(10 \%)^{18}$ and trained subjects despite a marked reduction in training volume and high baseline $\mathrm{VO}_{2}-\max (\sim 60 \mathrm{~mL} / \mathrm{min} / \mathrm{kg})$ before the intervention. ${ }^{24}$ Apparently, maximal or near maximal effort exercise, even for a relative short duration, does have a significant influence on $\mathrm{VO}_{2}$-max, at least when performed as 10-20-30 training.

Taken together, the present findings confirm that the 10-20-30 training is very effective in improving performance and increasing maximum oxygen uptake, and the novel observation is that these changes, in recreational runners, can be obtained with a non-maximal effort during the 10 -s high-speed runs. It should be noted that the MAX group had greater improvement in the final speed of the incremental test to exhaustion than the SUBMAX group ( $4 \%$ vs. $2 \%$ ). This difference may be explained by a larger improvement in the anaerobic capacity in the MAX compared to the SUBMAX group, which is supported by the finding, that blood lactate after the exhaustive test in the MAX group tended to be higher after compared to before the intervention (Table 2).
TABLE 2 Whole blood pH , lactate, and $\mathrm{K}^{+}$before (Pre) and after (Post) 6 weeks of 10-20-30 training with submaximal (SUBMAX) or maximal (MAX) exercise intensity before (Rest), after two submaximal running bouts at $12 \mathrm{~km} / \mathrm{h}$ (Sub1 \& Sub2) separated by 20 min of recovery, and after ( $\mathrm{E} \times \mathrm{h}$ ) as well as 3 min after (Exh +3 ) running to exhaustion during an incremental test.

|  |  | Rest |  | Sub1 |  | Sub2 |  | Exh |  | Exh +3 |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | SUBMAX | MAX | SUBMAX | MAX | SUBMAX | MAX | SUBMAX | MAX | SUBMAX | MAX |
| pH | Pre | $7.36 \pm 0.01$ | $7.36 \pm 0.01$ | $7.30 \pm 0.02$ | $7.30 \pm 0.03$ | $7.32 \pm 0.02$ | $7.30 \pm 0.04$ | $7.16 \pm 0.02$ | $7.18 \pm 0.04$ | $7.16 \pm 0.02$ | $7.18 \pm 0.04$ |
|  | Post | $7.38 \pm 0.01$ | $7.35 \pm 0.00$ | $7.30 \pm 0.02$ | $7.27 \pm 0.01$ | $7.32 \pm 0.01$ | $7.30 \pm 0.01$ | $7.15 \pm 0.02$ | $7.16 \pm 0.02$ | $7.17 \pm 0.02$ | $7.17 \pm 0.02$ |
| Lactate mmol/ | Pre | $1.2 \pm 0.1$ | $1.1 \pm 0.1$ | $4.4 \pm 0.4$ | $4.2 \pm 0.5$ | $4.5 \pm 0.4$ | $4.3 \pm 0.5$ | $13.6 \pm 1.0$ | $11.5 \pm 0.6$ | $15.2 \pm 1.0$ | $13.2 \pm 0.9$ |
|  | Post | $1.0 \pm 0.1$ | $1.2 \pm 0.2$ | $4.1 \pm 0.5$ | $3.7 \pm 0.5$ | $4.0 \pm 0.3$ | $3.4 \pm 0.3^{*}$ | $13.6 \pm 0.8$ | $12.9 \pm 0.8$ | $14.4 \pm 0.9$ | $13.7 \pm 0.9$ |
| $\mathrm{K}^{+} \mathrm{mmol} / \mathrm{l}$ | Pre | $4.0 \pm 0.3$ | $4.4 \pm 0.3$ | $5.0 \pm 0.2$ | $5.6 \pm 0.3$ | $5.1 \pm 0.2$ | $5.5 \pm 0.3$ | $4.8 \pm 0.2$ | $5.3 \pm 0.3$ | $4.1 \pm 0.2$ | $4.5 \pm 0.3$ |
|  | Post | $3.9 \pm 0.1$ | $4.1 \pm 0.2$ | $4.6 \pm 0.1$ | $4.6 \pm 0.2^{*}$ | $4.7 \pm 0.1$ | $4.9 \pm 0.1^{*}$ | $4.7 \pm 0.1$ | $4.8 \pm 0.2$ | $3.8 \pm 0.0$ | $3.9 \pm 0.2^{*}$ |

Note: Values are means $\pm$ SE. See Training for description of 10-20-30 training.

A novel finding in the present study was that the $10-$ 20-30 training with maximal effort led to an increase in the expression of muscle OXPHOS complexes. Similarly, Fiorenza et al. ${ }^{18}$ observed that muscle OXPHOS complex I-IV and total OXPHOS were enhanced after a 6 -week period with three training sessions a week of $4-10$ repetitions of 20 -s sprints, and in addition, they found that the muscle respiratory capacity was elevated. In contrast to the MAX group, the SUBMAX group had no change in muscle OXPHOS complexes suggesting that the effort during the high-speed running must be maximal to achieve changes in muscle oxidative capacity. It may be explained by a greater recruitment of fast twitch fibers when the effort is maximal, and, thus, enhancement of the expression of oxidative enzymes in these fibers, which have lower oxidative capacity than slow twitch fibers. ${ }^{5,25}$ However, some studies of adaptations of muscle oxidative enzymes in slow and fast twitch fibers with a period of SET have not been able to find increases in fast twitch fibers. ${ }^{5,25}$ Thus, the reason for the difference in the muscle adaptations with the two types of 10-20-30 training needs to be clarified. As the SUBMAX and MAX group had similar increases in $\mathrm{VO}_{2}$-max, the findings of different muscle adaptations with the two interventions, indicate that the muscle oxidative capacity is not limiting $\mathrm{VO}_{2}$-max in these already trained subjects. Thus, supporting the notion that $\mathrm{VO}_{2}$-max in trained subjects mainly is limited by central factors. ${ }^{26}$

The higher $\mathrm{VO}_{2}$-max after the training intervention is one of the explanations for the improvement in $5-\mathrm{km}$ performance in both groups. It was not related to running economy, as it was unchanged in both groups. In MAX, blood lactate during submaximal running was lower after the intervention period, which fits well with the finding of elevated expression of muscle oxidative enzymes. This indicates a better endurance capacity ${ }^{27}$ and may have led to higher relative intensity during the $5-\mathrm{km}$ test, which can have contributed to the better performance. On the other hand, the SUBMAX group did not change blood lactate during submaximal running with the intervention and had the same improvement in $5-\mathrm{km}$ performance.

In summary, 10-20-30 training with non-maximal effort during the $10-\mathrm{s}$ high-speed running was as efficient in improving $5-\mathrm{km}$ and $\mathrm{VO}_{2}$-max as when performed with maximal effort. However, adaptations in muscle oxidative enzymes occurred only in the group performing the 10-2030 training with maximal effort.

## 4.1 | Perspective

"Lack of time" is a common barrier to regular physical activity, and the 10-20-30 training has been identified

TABLE $3 \quad \mathrm{VO}_{2}$, running economy and RER at $12 \mathrm{~km} / \mathrm{h}$ before (Pre) and after (Post) 6 weeks of 10-20-30 training with submaximal (SUBMAX) or maximal (MAX) exercise intensity.

|  | SUBMAX |  | MAX | Post |
| :--- | :--- | :--- | :--- | :--- |
|  | Pre | Post | Pre | $3564 \pm 116$ |
| $\mathrm{VO}_{2} 12 \mathrm{~km} / \mathrm{h}(\mathrm{ml} \mathrm{O} 2 / \mathrm{min})$ | $3306 \pm 96$ | $3314 \pm 90$ | $3499 \pm 95$ | $223.1 \pm 3.6$ |
| RE $12 \mathrm{~km} / \mathrm{h}\left(\mathrm{ml} \mathrm{O}_{2} / \mathrm{min} / \mathrm{kg}\right)$ | $217.9 \pm 3.2$ | $219.0 \pm 2.6$ | $217.2 \pm 2.7$ | $0.94 \pm 0.0$ |
| RER $12 \mathrm{~km} / \mathrm{h}$ | $0.95 \pm 0.0$ | $0.93 \pm 0.0$ | $0.95 \pm 0.0$ |  |

Note: Values are means $\pm$ SE. $\mathrm{VO}_{2}$, oxygen consumption; RE, running economy; RER, respiratory exchange ratio. See Training for description of 10-20-30 training.

|  | SUBMAX |  |  |  |  | MAX |  |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :---: |
|  | Pre | Post |  | Pre | Post |  |  |
| OXPHOS (complex I) | $1.19 \pm 0.09$ | $1.20 \pm 0.06$ |  | $1.08 \pm 0.05$ | $1.25 \pm 0.08^{*}$ |  |  |
| OXPHOS (complex II) | $1.16 \pm 0.14$ | $0.93 \pm 0.08$ |  | $1.07 \pm 0.12$ | $1.33 \pm 0.11^{*}$ |  |  |
| OXPHOS (complex III) | $0.82 \pm 0.08$ | $0.79 \pm 0.05$ |  | $0.78 \pm 0.06$ | $1.00 \pm 0.06^{*}$ |  |  |
| OXPHOS (complex IV) | $1.02 \pm 0.07$ | $1.01 \pm 0.05$ |  | $1.01 \pm 0.05$ | $1.10 \pm 0.03^{*}$ |  |  |
| OXPHOS (complex V) | $0.93 \pm 0.05$ | $0.90 \pm 0.03$ |  | $0.89 \pm 0.04$ | $0.97 \pm 0.03$ |  |  |
| OXPHOS (total; I-V) | $5.06 \pm 0.37$ | $4.76 \pm 0.21$ |  | $4.73 \pm 0.25$ | $5.52 \pm 0.27^{*}$ |  |  |

Note: Values are means $\pm$ SE. OXPHOS, oxidative phosphorylation. See Training for description of 10-2030 training.
*Different ( $p<0.05$ ) from Pre.

TABLE 4 Muscle protein expression before (Pre) and after (Post) 6 weeks of 10-20-30 training with submaximal (SUBMAX) or maximal (MAX) exercise intensity.
as a time-efficient exercise strategy to improve performance and health. ${ }^{7}$ In addition, the perceived effort during 10-20-30 training is lower compared to other interval training forms. ${ }^{7,8}$ The present findings show that 10-20-30 training performed with a non-maximal effort during the 10 -s high-speed running is also efficient. Thus, specifically applicable to people that are not highly motivated or able to do maximal-intensity training.

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## CONFLICT OF INTEREST STATEMENT

Jens Bangsbo is the author of the book "10-20-30 træning" describing in Danish how to conduct 10-20-30 training.

## DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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