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Acute caffeine intake improves lower body resistance exercise performance with blood flow restriction

Running head: Exercise with blood flow restriction and caffeine

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ABSTRACT

Purpose: The purpose of this study was to examine the effects of acute caffeine intake on physical performance in three sets of unilateral knee extension with blood flow restriction. Methods: In a double-blind crossover design, 22 trained men ingested 6 mg/kg of caffeine (CAF) or a placebo (PLA), 1 hour prior to performing unilateral knee extension exercise with blood flow restriction until exhaustion (30% of 1RM). Results: There was a significant difference in the number of repetitions between the CAF and PLA conditions in the 1st set (28.3 ± 5.3 vs 23.7 ± 3.2; $P=0.005$), 2nd set (11.6 ± 3.1 vs 8.9 ± 2.9; $P=0.03$), and total repetitions performed across the three sets (44.5 ± 9.4 vs 35.0 ± 6.6; $P=0.001$). Blood lactate was also significantly different ($P=0.03$) after exercise between the CAF (7.8 ± 1.1 mmol.L$^{-1}$) and PLA (6.0 ± 0.9 mmol.L$^{-1}$). In regard to pain perception, there was a difference between the CAF and PLA in the 2nd (6.9 ± 1.5 vs 8.4 ± 1.4; $P=0.04$) and 3rd sets (8.7 ± 0.4 vs 9.5 ± 0.6; $P=0.01$). No differences were found for perceived effort. Conclusion: Acute caffeine intake increases performance, blood lactate concentration and reduces perception of pain in unilateral knee extension exercise with blood flow restriction.
INTRODUCTION

Resistance training of moderate/high load (> 60% of 1 maximal repetition - RM) is considered an integral part of any physical conditioning program to increase muscle strength or muscle mass for health in adults. However, in recent years resistance training with low loads (<30% 1RM), performed with blood flow restriction (BFR), has been suggested as being similarly effective as moderate/high-load resistance training in increasing strength and muscle mass. Although the physiological mechanisms regarding the improvement of strength and muscle mass after a low-load resistance training program with BFR are still unclear, this training model has application both for healthy, non-trained subjects at different ages and athletes. In this context, while resistance training with BFR is not significantly better than traditional moderate/high-load resistance training to improve strength and/or muscle mass, it can be added into the training routine as a variation of exercises and may provide an alternative exercise mode that some individuals find more appealing than traditional resistance training.

In the context of resistance training to increase physical performance, in addition to training per se, it is common for athletes and recreational exercisers to use ergogenic aids as a means to increase or accelerate changes in physical performance. Caffeine, for example, is one of the most widely used ergogenic aids, often employed to increase performance in exercises involving muscular strength, especially when performing several repetitions until exhaustion. The mechanism of action for caffeine has been explained by the high affinity of caffeine with adenosine receptors, inhibiting the action of this substance and, consequently, reducing the perception of effort and pain. Likewise, caffeine can promote greater performance in the propagation of signals between the brain and neuromuscular junction, acting...
peripherally on the ryanodine channels in the release of calcium, optimizing the process of excitation-contraction of the skeletal musculature\textsuperscript{13}. In the context of the BFR-resistance exercise model, sustained blood flow reduction to the muscle during exercise may reduce intramuscular calcium influx\textsuperscript{14}, which could theoretically limit the effect of caffeine on the excitation-contraction coupling process in skeletal muscles. However, the possible ergogenic effects of caffeine have not yet been investigated in low intensity resistance exercise with BFR. Given there is potential for both caffeine ingestion and BFR to act independently and possibly synergistically, it is important to examine if this is the case. No study to date has examined this issue.

In this sense, the purpose of this study was to examine the effects of acute caffeine intake on physical performance in three sets of unilateral knee extension with BFR to failure. Additionally, rating of perceived exertion, perceived pain and blood lactate concentration were analyzed.

MATERIALS AND METHODS

Subjects

Sample size calculation was performed considering a difference between two means of three repetitions, an expected standard deviation of 2, statistical power of 80\%, and level of significance lower than 0.05 in a pilot sample of this study. Thus, the minimum sample required was 16 subjects. Consequently, the sample in the current study included 22 trained men (Table 1) to account for potential drop out during the experimental procedures. The inclusion criteria to take part were: non-smokers, non-users of dietary supplements, non-users of anabolic steroids, the absence of muscular or metabolic problems, body mass index below 30 kg.m\textsuperscript{2}, trained in bodybuilding for at least 12 months (but without experience with BFR exercises), and
non-habitual caffeine users. Individuals were instructed not to engage in vigorous exercise or consume alcoholic beverages for 72 h prior to each testing session until the end of the experiment. All participants were informed about the study procedures and possible effects of caffeine intake and provided informed consent to participate. The study was approved by institutional ethics of the State University of Londrina (application number 1.141.230/2015).

**Experimental design**

The study employed a repeated-measures, within-subjects design and was conducted during four non-consecutive days with intervals of between 48-72 h. Anthropometric measurements and 1RM testing was performed during the first visit. This was followed by familiarization with the scales that were used for subjective perception of effort (RPE)\textsuperscript{15} and pain (PP)\textsuperscript{16}. On the second visit, the 1RM retest was performed. The remaining two visits were assigned to the experimental sessions administered using a randomized double-blind cross-over design. The subjects ingested either one capsule of caffeine (CAF) or a placebo (PLA) and, after 60 min, performed unilateral knee extension exercise with BFR (three sets to exhaustion, 1 min recovery interval between sets, at an intensity of 30% 1RM). The BFR was maintained throughout the whole exercise bout (all sets and repetitions). Subjects were instructed and verbally encouraged to perform the maximum number of repetitions during each set. The repetitions were performed at a rate of 1.5 seconds (via digital metronome) for both concentric and eccentric contractions. The RPE and PP were applied after the end of each set. Blood lactate was collected after the end of the exercise. At the end of the experiment, the subjects were questioned as to whether they were able to distinguish between the two capsules to identify which was caffeine,
in order to determine efficacy of blinding and the individual perception of the effect of this substance.

**Maximum strength test**

The 1RM test and retest were performed within a 48-h interval using a unilateral extensor chair (the dominant leg) (TechnoGym®, Rome, Italy), was determined according to methods accomplished by Seo et al.\textsuperscript{17}. A warm-up was performed with a set of 10 repetitions (~50% of predicted 1RM). Individuals were allowed up to five attempts to determine 1RM, with a recovery interval of 3-5 min. All subjects were instructed and verbally encouraged to perform one correct repetition. The load was considered maximal when the subjects performed only one complete repetition. The highest load obtained in either the test or retest 1RM trials was used in subsequent experimental trials. Test/retest reliability for the 1RM was performed and a high intraclass correlation coefficient (ICC) was found, R = 0.80.

**Caffeine or placebo intake**

For the experimental sessions, each subject ingested a capsule containing 6 mg of caffeine per kilogram of body weight and a placebo capsule (maltodextrin) with 200 ml of water administered in a randomized order. The habitual average caffeine intake of the participants was assessed through a questionnaire\textsuperscript{18} translated to Portuguese. All participants were considered low habitual caffeine users (80.1 ± 10.4 mg.day\textsuperscript{-1}). The same list was used to instruct the individuals not to consume the same substances for 48 h prior to testing.
Blood flow restriction

To elicit BFR, a cuff 18 cm wide and 90 cm long was positioned on the proximal third of the thigh. A vascular Doppler (MARTEC DV600, São Paulo, Brazil) positioned on the posterior tibial artery was used to identify the sound of the passage of blood flow. From identification of the sound, the cuff was inflated until the sound was interrupted and, at that moment, the restriction value was recorded. Cuff pressure during the experimental session was maintained at 80% of the total blood flow restriction value and was released only after the end of the final set.

Blood lactate collection

Blood lactate concentration was obtained at the moment of rest during the first visit to the laboratory and at an interval of up to two minutes after the end of the final set of each experimental session. Prior to the blood sample collection, asepsis was performed with 70% alcohol on the digital pulp of the middle finger of the right hand. The puncture was performed using disposable lancets, the drop of blood (5µl) in suspension being applied to a specific area of the reactive strip and analyzed by means of a portable lactometer (AccutrendPlus, USA).

Rating of perceived exertion and perceived pain

To measure perceived exertion and perceived muscle pain, the OMNI 0-10 rating of perceived exertion (RPE) and a perceived pain (PP) visual analog scales were used respectively. Familiarization with the scales was performed on the first day of the individuals' visit to the laboratory. Subsequently, during experimental trials, at the end of each set, the RPE and PP values were also collected.
Statistical analyses

The Shapiro-Wilk test was used to verify the distribution of the data and the Levene’s test to verify the homogeneity of the variances. Considering the normal distribution of data, for the comparison of the number of repetitions, RPE, and PP a two-way ANOVA with repeated measures was used (caffeine/placebo x number of sets). The Student T-test for dependent samples was applied for the blood lactate analysis. Two-way ANOVA was used to compare the performance in the total number of repetitions, RPE, and PP, among those who identified correctly and those who made a mistake about the intake of the caffeine capsule. In all cases, the Tukey post-hoc test was used to identify significant results. Additionally, to determine the magnitude of the findings, Cohen’s d effect sizes (ES) were calculated for the differences between PLA and CAF, following the classification: small (0.20<ES<0.50), medium (0.50≤ES<0.80) or large (ES≥0.80). The level of significance adopted was P<0.05. The data were analyzed in Statistica 12.0 software (Statsoft, Tulsa, OK, USA).

RESULTS

Values are expressed as mean and standard deviation.

Table 1 presents the general characteristics of the sample. Randomization showed that 10 subjects started with the CAF session and 12 subjects started with the PLA session.

Table 2 shows the number of repetitions performed, RPE, and PP in the CAF and PLA conditions. For the number of repetitions performed, there was a significant interaction between CAF and PLA (F=18.45; P=0.02) with the Tukey post-hoc test identifying a significant difference between the 1\textsuperscript{st} and 2\textsuperscript{nd} sets. The RPE analysis demonstrated no significant inter-group interaction. However, there was a
significant intra-group interaction in CAF (F=14.37; P=0.04) and PLA (F=16.65; P=0.03) conditions. RPE scores was significantly lower after the 1st set of CAF and PLA in relation to the other sets. For the PP analysis, there was significant interaction between CAF and PLA (F=23.78; P=0.01). The PP results demonstrated a progressive increase in the CAF condition from the 1st to 3rd sets. Conversely, in the PLA condition PP was only significantly different in the 1st set compared to the 2nd and 3rd sets. Furthermore, PP was significantly different between CAF and PLA in the 2nd and 3rd sets.

The analysis of blood lactate, by Student’s T-test, showed no significant differences in resting values between the CAF (2.1 ± 0.3 mmol.L⁻¹) and PLA (2.2 ± 0.2 mmol.L⁻¹; Cohen’s d ES = 0.39). After the end of the exercise, the values were significantly higher (P=0.002) than those observed at rest, and a significant difference (P=0.03) was observed between the CAF (7.8 ± 1.1 mmol.L⁻¹) and PLA (6.0 ± 0.9 mmol.L⁻¹; Cohen’s d ES = 1.79) conditions.

Table 3 shows the two-way ANOVA results for the total number of repetitions performed between subjects who correctly determined which condition was the caffeine condition and which the placebo. Nine subjects correctly identified the caffeine trial (true positive) and also performed more repetitions in this condition than after placebo intake. Similarly, those failing to correctly identify the caffeine trial also performed more repetitions than after taking the placebo. However, there was a significant difference in the number of repetitions completed after caffeine intake between those who identified and those who did not correctly identify caffeine (P <0.05). No differences were observed for RPE, PP, or lactate.

Figure 1 presents the individual responses between CAF and PLA in terms of the total number of repetitions performed.
DISCUSSION

The main findings of the present study were: 1) caffeine intake increased both the number of repetitions performed and capillary blood lactate whilst also reducing pain sensation in knee extension exercise with BFR; 2) subjects who accurately interpreted the caffeine trial as such performed more repetitions than those who did not perceive it accurately. In both cases, the Cohen’s d ES were large, ratifying the significant level identified. No study to date has examined the concurrent effects of caffeine ingestion and BFR on strength performance and few prior studies examining caffeine ingestion have also examined whether the participant’s perception of the substance ingested influences the response to the subsequent exercise protocol. As such the results of the present study are novel and extend the literature pertaining to effects of caffeine ingestion on exercise performance.

One of the possible mechanisms of action of caffeine on physical performance occurs through the increase in the release of calcium in the sarcoplasmic reticulum, boosting the excitation-contraction process\textsuperscript{13}. However, it is possible that calcium availability may be impaired under hypoxia conditions\textsuperscript{14}. Thus, one of the potential explanations is that exercise with BFR reduced the availability of calcium and, consequently, compromised performance. In the present study, as performance did not decrease, on the contrary, it increased, we suggest two hypotheses to explain the
seemingly paradoxical findings we present. A BFR threshold of 80% BFR was employed in the present study as this threshold is commonly used in the literature\textsuperscript{19}. As a consequence the experimental model did not apply vascular occlusion that interrupted 100% of the blood flow. This means, in the current study, there was no total hypoxia condition, despite less oxygen availability. Thus, even assuming a lower availability of calcium, this may not have been sufficient to compromise performance. Secondly, the release of calcium into the sarcoplasmic reticulum is not the only mechanism of action by which caffeine influences performance\textsuperscript{20}. Given that there is debate in regard to the mechanism by which caffeine is ergogenic, the results of the current study would imply that increased caffeine availability in the sarcoplasmic reticulum might not be the prime mechanism by which caffeine ingestion enhances muscular performance.

It has previously been established that caffeine can also act in the central nervous system by blocking adenosine receptors\textsuperscript{21}, attenuating the action of adenosine and increasing the release of adrenergic neurotransmitters to reduce PP\textsuperscript{22}. In the context of the current study, this may have helped individuals to continue performing the exercise for a longer duration and, consequently, increasing the number of repetitions performed with a concomitant increase in blood lactate values post exercise. The observed increase in blood lactate might also be associated with increased PP. However, in the present study, PP was lower in the caffeine condition (with large Cohen’s d ES) throughout the sets. In addition, lactate appears to influence PP depending on the concentration of protons and ATP\textsuperscript{24}. Thus, high lactate values may not increase PP. This finding aligns with other studies that have also reported dampened PP during resistance exercise with caffeine ingestion\textsuperscript{11}. Conversely, no significant differences in RPE were identified as a consequence of the substance
ingested. Such a finding is congruent with other resistance exercise data, where the authors hypothesized that caffeine may be able to improve performance by maintaining similar levels of perceived exertion to those who produced less work\textsuperscript{23}. Concerning the number of repetitions during exercise with BFR, there was a significant difference between CAF and PLA until the 2\textsuperscript{nd} set (P<0.05 and large Cohen’s d ES). The non-significant difference in the 3\textsuperscript{rd} set may be associated with fatigue in both groups. In this context, the ergogenic effect of caffeine may not be realized when several sets are performed to exhaustion and could be indicative that caffeine enhances peak strength performance. It is important to note that there was a significant difference in repetitions between subjects who correctly identified caffeine intake and those who did not identify the intake of caffeine. Some studies have carried out an individual analysis on the effects of caffeine on performance and identified that some subjects did not present an ergogenic effect\textsuperscript{25}. In other studies, the authors analyzed the side effects of caffeine action to see if the sample could identify the substance ingested\textsuperscript{26}. In the present study, we did not perform individual performance analysis and also did not verify side effects - we only asked the subjects at the end of the experiment if they thought that caffeine had been the first or second capsule consumed. It is important to reinforce that, irrespective of whether subjects correctly identified the caffeine ingestion trial or not, the participants performed more repetitions when they ingested caffeine compared to when placebo was ingested. However, subjects who correctly identified caffeine performed more repetitions than those who did not. Our results agree with the study by Saunders et al.\textsuperscript{27} The authors found that cyclists who correctly identified caffeine improved cycling performance to a greater extent than the overall effect of caffeine; and the performance also improved when participants ingested caffeine while believed they were ingesting placebo. Therefore,
the results of this study reinforces the possibility that caffeine has an individualized physiological and psychological action, allowing some subjects to have a superior ergogenic effect to others as a consequence of their expectancy of the effect of the substance they have ingested.

In regard to blood lactate responses to exercise, we observed an increase in this variable after exercise in both the placebo and caffeine conditions. Studies have shown that resistance exercise with BFR may result in blood lactate concentration values similar or higher when compared to high intensity exercise without BRF. It is worth mentioning that this type of exercise has some peculiarities in relation to conventional training of moderate-high intensity, among them is maintaining the blood flow restriction even in the recovery periods, which can significantly decrease the removal of blood lactate in the target muscles.

However, in the current study, blood lactate was higher after the exercise performed with caffeine intake. Such a finding is congruent with recent meta-analytical data identifying that acute caffeine intake significantly increases plasma lactate. In the present study, a possible explanation the greater number of repetitions performed after caffeine ingestion, would have also been associated with a longer duration of effort and a longer time in the BFR condition. As a consequence of this, blood lactate may have been elevated simply because of the greater work performed in the caffeine condition rather than because of the caffeine ingested. Despite this potential explanation, some studies have shown that plasma lactate did not change after caffeine intake even with increased performance. In this sense, the effect of caffeine on plasma lactate is still inconclusive and requires further investigation.

PRACTICAL APPLICATIONS
The present study is the first to investigate the acute effects of caffeine on an exercise performed with BFR. The positive effect of acute caffeine intake to increase the number of repetitions during unilateral knee extension with BFR may help practitioners, athletes, and coaches to optimize the performance in this model of resistance exercise. Notwithstanding, regardless of the results presented, there are some limitations of the current study. We used a single unilateral lower limb exercise as our outcome measure of resistance exercise performance. The results of the present study are only reflective of this type of exercise and we cannot confirm that the results presented herein would be reproducible in bilateral exercises and/or with different muscle mass. Plasma caffeine concentration was not measured and thus we cannot confirm the bioavailability of this substance in all study subjects. We did not test the reliability of the measurements during the exercises protocols. Although our subjects were familiar with resistance exercise protocols, they were not regular practitioners of resistance exercise with BFR. Exercise with BFR may feel different to exercise without BFR and as such some of the observed changes might be attributable to the feeling of the exercise, the error of measurement or to learning effects. Finally, as this is an acute study, we cannot verify whether the greater number of repetitions performed after caffeine intake would be significant to promote a superior effect on strength or muscle mass when analyzed in the long term. Future research would be welcome examines these issues and also seeks to replicate the results of the current study.

CONCLUSION

Acute caffeine supplementation increases physical performance and decreases PP in an exercise session of unilateral knee extension with BFR.
This study was partially supported by grants from the Brazilian Council for Research Development (CNPq).

The authors declare that they have no conflicts of interest.
REFERENCES


15. Lagally KM, Robertson RJ. Construct validity of the OMNI resistance exercise


Table 1. General characteristics of the sample (n=22).

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Value</th>
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<tr>
<td>Age (years)</td>
<td>23.4 ± 4.1</td>
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<tr>
<td>Height (cm)</td>
<td>177.2 ± 3.9</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>76.7 ± 4.0</td>
</tr>
<tr>
<td>Time of training (years)</td>
<td>2.6 ± 1.1</td>
</tr>
<tr>
<td>Level of BFR during exercise (mmHg)</td>
<td>131.4 ± 14.6</td>
</tr>
<tr>
<td>Habitual caffeine consumption (mg.day⁻¹)</td>
<td>80.1 ± 10.4</td>
</tr>
<tr>
<td>1 RM (kg)</td>
<td>87.2 ± 61.1</td>
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<td>30% of 1 RM (kg)</td>
<td>26.2 ± 2.5</td>
</tr>
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</table>
Table 2. Results of the two-way ANOVA with repeated measures to effect of caffeine supplementation on the number of repetitions, rating of perceived exertion and pain perceived.

<table>
<thead>
<tr>
<th></th>
<th>Caffeine</th>
<th>Placebo</th>
<th>Inter-group P value (caffeine vs placebo)</th>
<th>Cohen’s d effect size</th>
<th>Cohen’s d classification</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Repetitions</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1st set</td>
<td>28.3 ± 5.3*</td>
<td>23.7 ± 3.2*</td>
<td>0.005</td>
<td>1.05</td>
<td>Large</td>
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<tr>
<td>2nd set</td>
<td>11.6 ± 3.1†</td>
<td>8.9 ± 2.9†</td>
<td>0.03</td>
<td>0.90</td>
<td>Large</td>
</tr>
<tr>
<td>3rd set</td>
<td>4.6 ± 3.6</td>
<td>2.4 ± 3.0</td>
<td>NS</td>
<td>0.66</td>
<td>Medium</td>
</tr>
<tr>
<td>Total</td>
<td>44.5 ± 9.4</td>
<td>35.0 ± 6.6</td>
<td>0.001</td>
<td>1.17</td>
<td>Large</td>
</tr>
<tr>
<td><strong>Rating of perceived exertion</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1st set</td>
<td>6.3 ± 1.5*</td>
<td>6.1 ± 1.9*</td>
<td>NS</td>
<td>0.11</td>
<td>Small</td>
</tr>
<tr>
<td>Set</td>
<td>Mean ± SD 1st set</td>
<td>Mean ± SD 2nd set</td>
<td>p-value</td>
<td>Effect size</td>
<td></td>
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<tr>
<td>---------</td>
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<tr>
<td>2nd set</td>
<td>8.4 ± 1.1</td>
<td>8.5 ± 1.5</td>
<td>NS</td>
<td>0.07</td>
<td>Small</td>
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<tr>
<td>3rd set</td>
<td>9.4 ± 0.3</td>
<td>9.6 ± 0.8</td>
<td>NS</td>
<td>0.33</td>
<td>Small</td>
</tr>
</tbody>
</table>

**Pain perceived**

<table>
<thead>
<tr>
<th>Set</th>
<th>Mean ± SD 1st set</th>
<th>Mean ± SD 2nd set</th>
<th>p-value</th>
<th>Effect size</th>
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</thead>
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<tr>
<td>1st set</td>
<td>5.2 ± 1.0*</td>
<td>5.9 ± 2.2*</td>
<td>NS</td>
<td>0.40</td>
</tr>
<tr>
<td>2nd set</td>
<td>6.9 ± 1.5†</td>
<td>8.4 ± 1.4</td>
<td>0.04</td>
<td>1.03</td>
</tr>
<tr>
<td>3rd set</td>
<td>8.7 ± 0.4</td>
<td>9.5 ± 0.6</td>
<td>0.01</td>
<td>1.57</td>
</tr>
</tbody>
</table>

* Intra-groups significant difference (P=0.02) from 1st and 2nd sets; † Intra-groups significant difference (P=0.04) from 3rd set; NS = non-significant difference
Table 3. Results of the two-way ANOVA to the total number of repetitions performed between subjects who scored correctly and those who made a mistake about caffeine intake.

<table>
<thead>
<tr>
<th></th>
<th>Caffeine</th>
<th>Placebo</th>
<th>Inter-group P value (caffeine vs placebo)</th>
<th>Cohen’s d effect size</th>
<th>Cohen’s d classification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Correct identification of caffeine (n=9)</td>
<td>48.8±4.8</td>
<td>35.7±7.7</td>
<td>0.002</td>
<td>2.04</td>
<td>Large</td>
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<tr>
<td>Non-correct identification of caffeine (n=13)</td>
<td>41.5±7.1</td>
<td>34.5±6.0</td>
<td>0.001</td>
<td>1.06</td>
<td>Large</td>
</tr>
<tr>
<td>Intra-groups P value (correct vs non-correct)</td>
<td>0.04</td>
<td>NS</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

NS = non-significant difference
Figure 1. Individual responses between caffeine and placebo sessions