RESEARCH ARTICLE

Acute impact of a single dose of Dynamine®, TeaCrine®, caffeine, and their combination on systemic hemodynamics and associated measures in men and women

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Abstract

Background: Dietary supplements purported to improve focus and "energy" often contain stimulants, which elevate both heart rate (HR) and blood pressure (BP). We evaluated the safety profile of the purine alkaloid methylliberine (Dynamine®) with regards to resting HR, BP, respiratory rate (RR), and body temperature (BT).
Methods: 6 men (aged: 24±4) and 6 women (aged: 22±3) ingested methylliberine (Dynamine®; Compound Solutions, Inc.) at 25mg (M25) and 100mg (M100), caffeine at 150mg (C150), M100+C150, M100 + theacrine (Teacrine®, Compound Solutions, Inc.) at 50mg (M100+T50), and M100+T50+C150. All conditions were assigned using a Latin square design, with approximately one week separating the six different assignments. HR, BP, RR, and BT were collected at baseline and 0.25, 0.5, 1, 1.5, 2, 4, 6, 8, 24, and 48 hours post-dose. Subjective mood was also recorded. Meal replacement bars and shakes were provided after hours 3 and 6.
Results: For SBP and DBP, condition effects were noted ($p<0.05$), with all three caffeine conditions higher than those without caffeine. Condition effects were noted for attentive ($p=0.02$) and energetic ($p=0.02$), with M100+C150 greater than M100+T50; and moody ($p<0.01$), with M100 and M100+T50 greater than M100+C150. Main effects of time were noted for HR ($p<0.001$), BT ($p<0.01$), SBP ($p<0.01$), DBP ($p<0.01$), and focused ($p=0.02$), but no statistically significant interactions were noted.
Conclusion: Methylliberine, alone or in combination with caffeine and/or theacrine, does not result in any significant increase in HR, BP, RR, or BT in healthy adults and has little impact on subjective mood. These findings should be considered with the understanding that subjects began testing in the early morning hours while in a fasted and rested state and were provided with two meals (hours 3 and 6) during the evaluation period, which may have impacted subjective mood.

Key Words: methylliberine, theacrine, caffeine, blood pressure, heart rate

1. Introduction

Dietary supplements designed to increase energy and focus are quite common, in particular within the sports supplement market. These are sold in many retail establishments and appeal to a wide variety of consumers for various purposes. Recently, two novel plant-based ingredients have made their way into the dietary supplement market: purine alkaloids known as theacrine and methylliberine.

Theacrine (1,3,7,9-tetramethyluric acid) is a metabolic derivative of caffeine found in the leaves of Camellia Kucha, *Coffea dewevrei*, *Coffea liberica*, and *Coffea abeokutae* and to a lesser degree in the seeds of cocoa, yerba mate, and Cupuaçu.[1-4] Theacrine, which is commercially available as TeaCrine®, has been found safe for human consumption and results in improved subjective feelings without significant physiological changes to heart rate (HR) or blood pressure (BP).[5-7]

Methylliberine (2-methoxy-1,7,9 tetramethyluric acid) is thought to be a metabolic derivative of theacrine and a precursor to liberine in some coffee leaves.[4] Like theacrine, methylliberine is found in *Coffea dewevrei*, *Coffea liberica*, and *Coffea abeokutae.* Methylliberine is commercially available as Dynamine® and has been shown to be safe in rats.[8] Little scientific research has been done on methylliberine, however it is hypothesized that it will act similarly to theacrine, but with a more rapid and brief effect.[8]

Due to the structural similarities between caffeine and the above plant-based ingredients, some concern has been expressed over the potential pressor effects of these agents. It is well-described that caffeine can either raise[9-10] or lower[9,11] HR and increase BP in a dose-dependent manner.[10-11] Previous studies have found increases in systolic BP (SBP) and diastolic BP (DBP) of 5-15 mmHg and 5-10 mmHg, respectively for caffeine consumption greater than 250 mg.[9] Additionally, small changes in HR have been observed with caffeine consumption over 150 mg, but these were found to be insignificant and diminish with acclimation. Consuming up to 400 mg caffeine daily is considered safe. While the change is typically small, certain individuals may have heightened sensitivity and responsiveness to caffeine and consequently experience a more robust increase in both HR and BP following acute caffeine ingestion. As previous research has indicated, the lack of significant pressor effects in subjects consuming theacrine[5-7,12-13] makes these compounds potential alternatives to caffeine in hypersensitive individuals.

While we have not worked directly with methylliberine, we have conducted prior studies with theacrine and caffeine alone and in combination (with each other and with additional ingredients) and noted only small increases in HR and BP. [6,12-13] For example, in a study of 20 healthy men and women ingesting theacrine (100mg) and/or caffeine (150mg), HR and BP were largely unaffected by treatment (e.g., HR was 3-5 beats per minute (bpm) lower in the hours following ingestion; BP was 1-2 mmHg higher in the hours following ingestion.[6] More recently, in a study of 50 healthy men and women ingesting theacrine or caffeine, only marginal increases (HR ~3 bpm; BP was ~3 mmHg higher) were noted following ingestion.[12]

Other investigators have also studied theacrine. In a double-blinded crossover study of 15 men and women, no significant changes in HR and BP were observed between placebo and theacrine (200 mg) over a 3 hour period.[7] An 8 week study on the safety of regular consumption of theacrine by 60 healthy men and women split between placebo, 200 mg theacrine,

and 400 mg theacrine also found no effects on HR or BP.[5]

There have been very limited controlled laboratory investigations using methylliberine by itself or in combination with other supplements.[8,14] Murbach et al. studied the toxicological effects of methylliberine in rats, with no-observed-adverse-effect level noted to be 150 and 225 mg/kg bw/day for males and females, respectively. To our knowledge, no studies have determined the impact of the combination of methylliberine, theacrine, and caffeine—which has now become the focus of many fitness-specific dietary supplements that are being sold on the market (e.g., Stimpact®, Mr. Hyde® Icon, and ALPHAGEN®). Since methylliberine is being used in many product formulations, alone and in combination with theacrine and caffeine, it is important to understand the potential acute impact of these agents on both HR and BP. We determined the effect of a single dose of methylliberine (as Dynamine®), theacrine (as Teacrine®), caffeine, or their combination on HR and BP in healthy men and women.

2. Materials and Methods

2.1 Subjects

A total of 6 men and 6 women were recruited to complete this study. A prime focus of this study was to determine the pharmacokinetics of the various ingredients alone and in combination (data in preparation and presented elsewhere). Therefore, the sample size of 12 individuals was appropriate and representative of men and women within the stated age range. Subjects were/had:

- age 18-50 years
- male or female
  - males had a body mass between 70-90kg
  - females had a body mass between 50-65kg
- body mass index (BMI) between 18-29.9 kg/m^2 (not obese)
- non-smoker
- no diagnosed history of diabetes
- no diagnosed history of cardiovascular disease
- no diagnosed history of neurological disease
- no consumption of alcohol within 48 hours of testing
- no consumption of caffeine-containing beverages within at least 48 hours of testing
- no consumption of grapefruit-, poppy seed-, or quinine-containing substances within 14 days of enrollment
- normal vital signs (heart rate, systolic and diastolic blood pressure, respiratory rate, and oral temperature)
- non-drug user (e.g., cannabinoids, amphetamines, benzodiazepines, cocaine, opioids, phencyclidine, barbiturates, cotinine)
  - verbal confirmation
- no history of illicit drug or other substances of abuse within 12 months of the screening visit
- no tobacco use for ≥ 90 days prior to screening
- consume < 400mg caffeine daily
- 12-lead ECG without clinically significant abnormalities and blood parameters within normal limits
- physically active: two or more days per week of structured physical activity for a minimum of 30 minutes each day
- if female, not pregnant

Health history, medication and dietary supplement usage, and physical activity questionnaires were completed by all subjects and reviewed by an investigator. Prior to participation, each subject was

informed of all procedures, potential risks, and benefits associated with the study through verbal and/or written form. They signed informed consent, as approved by the University of Memphis Institutional Review Board for Human Subjects Research (Proposal # FY2018-490).

2.2 Initial Laboratory Visit: screening visit

During the initial visit to the laboratory, subjects' HR, BP, height, weight, waist, and hip circumference were measured. A blood sample was taken and analyzed for metabolic panel and lipid panel. A 12-lead resting electrocardiogram was obtained. Women were asked to perform a urine pregnancy test to confirm that they were not pregnant. Upon completion of the screening and confirmation of all measures, eligible subjects were scheduled for their initial testing visit.

2.3 Independent Variable

The dietary ingredient known as methylliberine was evaluated, alone and in conjunction with caffeine and/or theacrine. Methylliberine (Dynamine®) and theacrine (Teacrine®) are ingredients of Compound Solutions, Inc. (Carlsbad, CA) and were provided by this company. A contract manufacturer produced all capsules in accordance with Good Manufacturing Practices. Capsules were of similar appearance and provided in unlabeled bottles. Subjects ingested the assigned condition on each of the six test days in the presence of investigators. Only one capsule was ingested each test day. The following conditions were included and assigned in double-blind manner using a Latin square design:

1. 25 mg methylliberine (M25)
2. 100 mg methylliberine (M100)
3. 150 mg caffeine (C150)
4. 100 mg methylliberine + 150 mg caffeine (M100 + C150)
5. 100 mg methylliberine + 50 mg theacrine (M100 + T50)
6. 100 mg methylliberine + 50 mg theacrine + 150 mg caffeine (M100 + T50 + C150)

The dosage of each studied ingredient was based on prior work [6,12-13] using the ingredients alone and in combination with one another, in addition to anecdotal observations of human subjects ingesting these ingredients alone and in combination. No true placebo condition was used in this experiment, as we used the caffeine only condition (150mg) as our control.

2.4 Test Visit Procedures

Subjects reported to the lab a total of six times during the morning hours. Testing began at approximately 6:00am. Subjects reported in a 10-hour fasted state, without having consumed alcohol or caffeine within the past 48 hours. Subjects were instructed to obtain at least 7 hours of sleep during the night prior to testing. Subjects were instructed to refrain from strenuous physical exercise for 24 hours prior to and during the test day. The time of day was matched for each lab visit, and approximately 7 days separated lab visits.

Resting HR and BP were obtained using an automated unit (Omron HEM 907XL), following a 5-minute seated rest period, with duplicate measures taken at each time. In addition, respiratory rate was counted in one minute and oral temperature was measured using a digital thermometer. Subjective feelings of attentive, energetic, motivated, irritable, focused, jittery, and moody were obtained using a 100mm visual analog scale, with 0 representing the lowest rating (none at all; feeling at the absolute lowest value on this scale for the select variable) and 100 representing the highest

rating (extreme; feeling at the absolute highest value on this scale for the select variable). All data were collected at baseline (pre-dose), and 0.25, 0.5, 1, 1.5, 2, 4, 6, 8, 24, and 48 hours post-dose.

2.5 Dietary Intake and Physical Activity

All subjects were instructed to consume their usual diet throughout the study period and to record all food and drink consumed during the 24 hours prior to each test day. Dietary records were analyzed using Food Processor SQL, version 9.9 (ESHA Research, Salem, OR). Meal replacement food bars (Clif Builder's®) or shakes (Orgain Organic Nutrition™) were provided to subjects after data collection at hour 3 and hour 6 on test days. Subjects were also provided with adequate meal replacement bars (3) and shakes (3) to consume following the 8 hour data collection (during their time outside of the lab between hours 8 and 24). No food other than what was provided to subjects was allowed during each test day. The only beverage that subjects were permitted was water. Subjects returned the following mornings for the 24 and 48 hour data collection, again in a 10 hour fasted state. The same amount of meal replacement bars or shakes were consumed by each subject during each visit (both in lab and outside of lab). Subjects were instructed to maintain typical physical activity levels throughout the study period, with the exception of refraining from strenuous physical activity during the 24 hours prior to each test day and for the actual test day itself.

2.6 Data Analysis

The data are presented as mean ± SD. Data were scanned for outliers prior to being analyzed. Dietary data were analyzed using a 6 (condition) x 2 (gender) analysis of variance (ANOVA). Physiological data were initially analyzed using a 6 (condition) x 2 (gender) x 11 (time) ANOVA, and were reanalyzed using a 2-way design without gender after verifying no significant three-way interactions. Tukey post hoc testing was used in the event of a significant main effect. Analyses were performed using JMP software (SAS, Cary, NC) and statistical significance was set at $p \leq 0.05$.

3. Results

3.1 Overview

All 12 subjects successfully completed all aspects of the study. Descriptive characteristics are provided in Table 1. No subject reported an adverse event and the conditions were well-tolerated. Other than the expected gender differences in dietary data, no differences were noted during the day prior to each test day across conditions ($p>0.05$). Data can be seen in Table 2.

3.2 Outcome Measures

As expected, we noted the usual gender differences in HR and BP, with women generally having higher HR and lower BP as compared to men. However, no three-way interactions were noted ($p>0.05$) and data were analyzed without including gender in the model. No condition x time interactions were noted for any variable ($p>0.05$).

In order to limit hydration effects, subjects' water intake was regulated during lab testing of each condition. Men and women consumed 44±9 oz and 45±10 oz, respectively.

Table 1. Characteristics of 12 healthy men and women

Variable	Men	Women	P value
Age (years)	24±4	22±3	0.51
Height (cm)	175±3	163±3	<0.01
Weight (kg)	77±6	58±4	<0.01
BMI ($kg \cdot m^{-2}$)	25±1	22±2	0.01
Waist Circumference (cm)	83±9	72±4	0.03
Hip Circumference (cm)	99±8	97±6	0.53
Waist/Hip Ratio	0.83±0.03	0.75±0.02	<0.01
Resting HR (bpm)	65±9	73±8	0.14
Resting SBP (mm Hg)	127±8	114±5	0.01
Resting DBP (mm Hg)	73±11	71±5	0.73
Glucose (mg/dL)	86±5	85±9	0.78
Cholesterol (mg/dL)	169±22	148±24	0.15
HDL-C (mg/dL)	48±9	46±7	0.45
LDL-C (mg/dL)	107±25	91±21	0.65
Triglycerides (mg/dL)	68±34	55±23	0.45
Anaerobic Exercise (yrs)	3±2	3±3	0.59
Anaerobic Exercise (hrs/wk)	3±3	1±1	0.15
Aerobic Exercise (yrs)	4±3	6±4	0.36
Aerobic Exercise (hrs/wk)	3±2	3±1	0.74
Caffeine Intake (mg/day)	155±85	230±103	0.20

Values are Mean ± SD

Table 2. Dietary data of men and women during the 24 hours prior to each test day

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine + Theacrine 100mg+ 50mg	Methylliberine + Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Kilocalories	2026±743	2185±988	1594±569	2001±589	1783±866	1907±754
Protein (g)	91±31	120±62	77±28	91±37	78±43	96±55
Carbohydrate (g)	241±76	253±123	189±77	254±49	201±89	213±78
Fiber (g)	17±6	18±14	15±11	17±5	15±8	16±8
Sugar (g)	62±37	71±32	62±24	68±22	65±31	53±15
Fat (g)	78±41	78±53	60±32	70±34	76±40	75±43
Vitamin C (mg)	37±27	68±99	32±32	42±28	41±28	64±91
Vitamin E (mg)	3±2	7±12	1±1	3±2	2±2	8±12
Vitamin A (RE)	229±237	1081±1425	148±152	492±313	404±357	759±1330
WOMEN						
Kilocalories	1551±491	1302±342	1360±390	1268±451	1585±326	1638±485
Protein (g)	67±26	59±25	64±22	51±28	75±25	68±28
Carbohydrate (g)	178±88	154±69	162±69	174±86	181±82	188±77
Fiber (g)	22±10	18±8	22±7	19±9	20±7	21±14
Sugar (g)	61±50	47±23	54±39	41±27	58±42	54±26
Fat (g)	64±20	53±18	57±20	43±14	64±8	70±22
Vitamin C (mg)	120±76	74±29	78±51	90±80	111±86	95±83
Vitamin E (mg)	6±3	5±2	5±3	4±2	7±6	5±4
Vitamin A (RE)	263±254	324±298	493±438	369±383	405±232	435±486

Values are mean ± SD

Other than expected gender differences, no differences of statistical significance were noted between conditions ($p>0.05$).

For HR, a time effect was noted ($p<0.01$), with values highest between hours 4 and 8, likely in response to the feeding at hours 3 and 6. HR was very stable within the first few hours following ingestion for all conditions, did not appear impacted by treatment, and increased slightly starting at hour 4 for all conditions. Gender-specific data for HR are shown in Table 3, and Figure 1A shows the same data collapsed across genders.

For SBP, a condition effect was noted ($p<0.01$), as was a time effect ($p<0.01$). For the condition effect, M100 + C150 was higher than M25, M100, and M100 + T50, while M100 + T50 + C150 was also higher than M100 + T50—highlighting the impact of caffeine on SBP. For the time effect, in a similar manner as with HR, values were highest between hours 4 and 8, likely in response to the feedings at hours 3 and 6. SBP increased to the greatest degree in the M100 + C150 condition (approximately 10 mm Hg at the 2hr time). Data for SBP are shown in Table 4 (genders included) and Figure 1B (genders collapsed).

Table 3. Heart rate (bpm) data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	64±8	66±9	67±10	64±9	65±7	66±7
0.25hr	65±10	65±11	65±6	60±6	65±9	64±7
0.5hr	64±10	66±5	69±7	63±8	65±8	61±8
1hr	60±8	68±9	62±8	59±5	63±6	60±8
1.5hr	61±7	63±5	63±6	59±7	62±8	62±4
2hr	62±9	63±9	66±10	60±5	62±7	63±6
4hr*	72±7	71±8	70±8	68±5	67±11	70±9
6hr	67±8	68±9	68±7	67±10	66±10	66±5
8hr*	69±10	73±12	74±12	68±7	70±6	70±8
24hr	68±8	68±8	72±6	66±5	67±8	66±8
48hr	70±9	71±4	69±8	70±7	68±6	69±8
WOMEN						
Pre	77±16	72±5	72±9	78±11	76±7	72±10
0.25hr	76±12	70±7	70±9	76±9	73±8	65±12
0.5hr	76±12	72±6	69±9	74±11	70±8	67±7
1hr	75±12	70±8	68±6	69±10	72±8	66±11
1.5hr	75±9	66±8	68±7	75±11	73±6	66±9
2hr	73±12	66±7	70±9	73±10	73±7	70±11
4hr	79±13	76±8	82±14	75±12	77±12	73±6
6hr	76±8	72±8	75±7	76±13	76±12	71±11
8hr	75±7	72±5	76±5	79±11	77±10	76±8
24hr	77±9	74±4	74±7	73±9	74±6	70±8
48hr	76±7	71±3	76±5	74±9	74±6	72±8

Values are mean ± SD (* time effect with values significantly higher than other time points ($p<0.01$).

For DBP, a condition effect was noted ($p=0.02$), as was a time effect ($p<0.01$). For the condition effect, M100 + C150 and M100 + T50 + C150 were both higher than M100 + T50; however, values for all conditions were very similar. DBP also increased to the greatest degree in the M100 + C150 condition (approximately 6 mm Hg at the 2hr time). Data for DBP are shown in Table 5 (genders included) and Figure 1C (genders collapsed). Only negligible increases in SBP and DBP were noted with methylliberine ingestion alone.

Table 4. Systolic blood pressure (mm Hg) data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	121±7	122±8	123±7	122±8	124±6	126±9
0.25hr	121±9	125±10	125±7	124±10	123±5	127±8
0.5hr	121±11	125±7	130±4	132±8	124±9	132±4
1hr	127±7	126±10	133±7	134±6	124±7	131±7
1.5hr	117±9	123±8	124±5	133±6	124±4	131±16
2hr	123±9	126±8	127±5	135±7	122±4	132±8
4hr*	127±5	127±6	135±8	126±7	128±9	132±7
6hr	131±12	122±7	126±8	132±7	124±10	131±11
8hr*	131±4	129±4	135±9	134±12	126±9	133±10
24hr	127±4	127±5	127±9	125±4	125±5	128±5
48hr	122±4	126±4	128±9	128±6	126±6	128±10
WOMEN						
Pre	112±7	108±7	111±6	111±4	110±5	111±4
0.25hr	113±10	106±11	113±8	116±4	109±5	111±7
0.5hr	116±7	111±6	113±4	114±8	111±5	117±5
1hr	114±5	109±2	114±8	114±7	110±9	114±8
1.5hr	111±4	110±6	115±9	116±7	108±5	116±6
2hr	109±7	112±9	117±7	120±7	113±8	109±5
4hr	118±8	117±9	118±5	117±9	113±9	116±4
6hr	115±8	113±5	115±5	115±4	112±3	113±5
8hr	117±6	118±4	115±8	121±13	112±7	114±5
24hr	115±9	111±4	112±5	113±6	112±5	112±6
48hr	113±8	111±5	112±8	116±4	113±6	108±3

Values are mean ± SD (* time effect with values significantly higher than other time points ($p<0.01$).

Table 5. Diastolic blood pressure (mm Hg) data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	70±9	74±5	72±6	71±8	71±9	74±8
0.25hr	75±7	73±6	74±2	74±10	71±5	78±6
0.5hr	72±7	75±5	79±7	76±10	72±8	80±5
1hr	76±5	75±6	77±6	79±8	74±5	77±8
1.5hr	75±9	75±7	76±6	78±6	72±6	76±6
2hr	75±8	72±6	74±8	83±11	72±4	81±9
4hr	72±9	75±8	75±6	72±6	70±7	75±6
6hr	76±10	73±5	73±7	74±12	71±5	76±8
8hr	73±6	75±4	75±5	73±5	76±5	76±8
24hr	74±7	75±6	72±6	73±6	74±5	76±6
48hr	71±8	76±5	75±5	76±4	74±5	74±8
WOMEN						
Pre	74±7	70±7	70±8	71±6	71±8	70±4
0.25hr	70±7	72±3	72±9	75±7	69±5	70±11
0.5hr	75±11	72±4	73±8	77±9	72±4	73±8
1hr	74±8	74±6	76±10	75±7	73±5	76±7
1.5hr	70±10	74±7	74±9	75±9	72±5	74±9
2hr	71±10	72±8	75±9	73±6	73±4	69±6
4hr	74±7	72±5	72±3	70±8	68±10	68±8
6hr	70±6	69±9	74±9	69±8	68±4	69±8
8hr	69±7	68±5	73±6	72±10	71±8	68±6
24hr	73±5	71±4	71±7	71±5	70±7	71±5
48hr	73±4	73±3	70±7	71±6	68±3	73±5

Values are mean ± SD

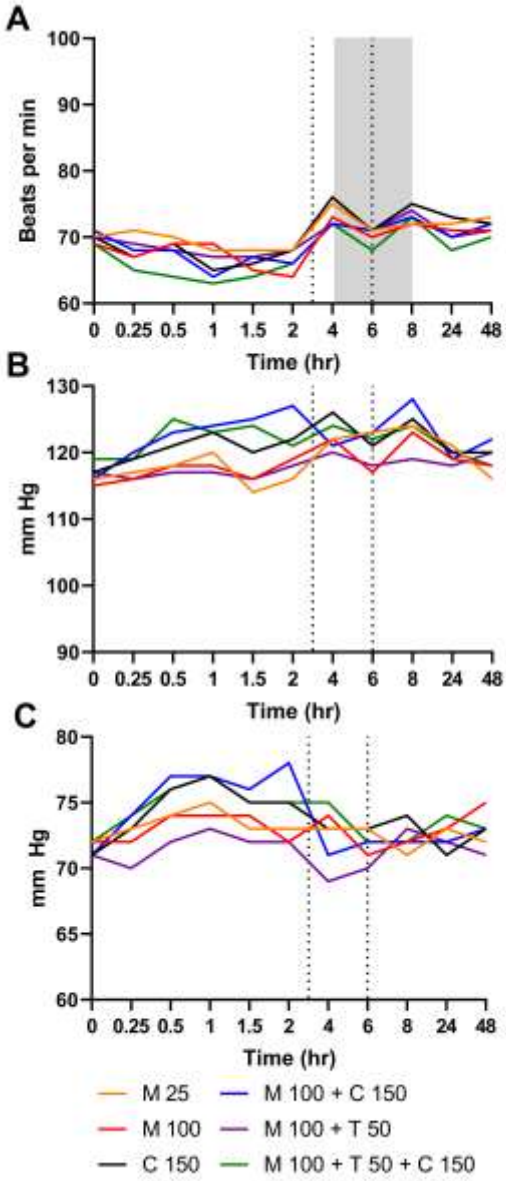

Figure 1. Heart rate (A), systolic (B) and diastolic (C) blood pressure of men and women ingesting methylliberine, caffeine, theacrine, or their combination. Note: 25 mg methylliberine (M 25), 100 mg methylliberine (M 100), 150 mg caffeine (C 150), 100 mg methylliberine + 150 mg caffeine (M 100 + C 150), 100 mg methylliberine + 50 mg theacrine (M 100 + T 50), and 100 mg methylliberine + 50 mg theacrine + 150 mg caffeine (M 100 + T 50 + C 150). Vertical dashed lines indicate feedings. Grey box indicates time in which time effects were observed.

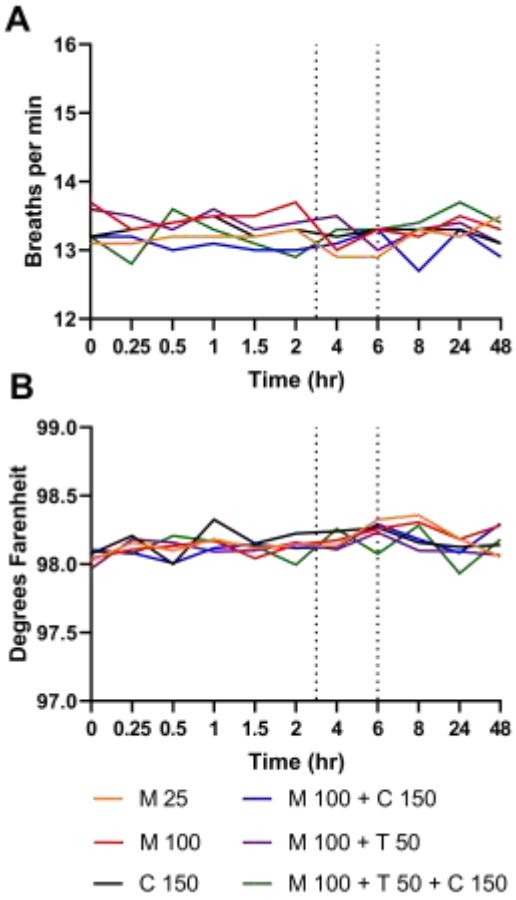

Figure 2. Respiratory rate (A) and body temperature (B) of men and women ingesting methylliberine, caffeine, theacrine, or their combination. Note: 25 mg methylliberine (M 25), 100 mg methylliberine (M 100), 150 mg caffeine (C 150), 100 mg methylliberine + 150 mg caffeine (M 100 + C 150), 100 mg methylliberine + 50 mg theacrine (M 100 + T 50), and 100 mg methylliberine + 50 mg theacrine + 150 mg caffeine (M 100 + T 50 + C 150). Vertical dashed lines indicate feedings.

Both respiratory rate (RR) and body temperature (BT) remained nearly identical across time for all conditions, with only a time effect noted for BT ($p<0.01$). Values remained relatively constant across time and very close to 98 degrees for all conditions at all times. Data are shown in Tables 6 and 7, as well as Figure 2A and Figure 2B, respectively.

Table 6. Respiration (breaths per minute) data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	12.5±0.8	13.3±1.4	13.2±1.2	13.0±1.3	13.3±1.0	13.2±0.8
0.25hr	12.7±1.0	13.0±0.6	13.3±1.2	13.0±1.1	13.7±0.5	12.7±0.8
0.5hr	12.8±1.0	13.2±1.0	13.5±1.4	12.8±0.8	13.3±0.8	13.8±0.8
1hr	13.0±1.1	13.5±1.2	13.8±1.0	12.7±1.0	13.7±1.4	13.0±0.9
1.5hr	12.8±0.8	13.7±1.2	12.8±1.0	12.8±0.8	13.2±0.8	12.8±1.0
2hr	13.0±1.3	13.8±1.7	13.5±1.2	12.8±1.2	13.5±0.5	12.7±0.8
4hr	12.5±0.8	13.0±1.1	13.0±0.9	12.8±1.2	13.7±1.0	13.3±1.5
6hr	12.2±0.8	13.2±0.8	13.2±1.0	13.2±1.0	12.8±0.8	13.2±1.7
8hr	13.0±0.9	13.0±1.1	13.5±0.8	12.7±1.2	13.2±1.0	13.3±1.2
24hr	12.5±0.8	13.5±0.5	13.3±1.2	13.0±0.9	13.5±0.8	13.7±1.0
48hr	13.3±1.0	13.2±1.2	13.2±1.2	12.5±1.0	13.0±0.9	13.2±1.3
WOMEN						
Pre	13.7±1.4	14.0±1.7	13.2±0.8	13.3±1.0	13.8±0.8	13.2±1.2
0.25hr	13.5±0.8	13.5±0.8	13.2±0.8	13.3±1.0	13.3±0.5	13.0±0.6
0.5hr	13.5±1.0	13.7±1.0	13.3±0.8	13.2±1.0	13.2±0.8	13.3±1.2
1hr	13.3±0.8	13.4±0.9	13.2±0.8	13.5±0.8	13.5±0.8	13.7±0.5
1.5hr	13.5±0.8	13.3±1.2	13.5±0.5	13.2±1.0	13.5±0.8	13.3±0.8
2hr	13.7±0.8	13.5±1.0	13.2±0.8	13.2±1.0	13.3±0.8	13.2±0.8
4hr	13.3±1.0	13.0±0.9	13.3±0.8	13.3±1.2	13.3±0.8	13.3±0.8
6hr	13.7±1.4	13.5±0.8	13.3±0.5	13.3±1.2	13.2±0.8	13.3±0.8
8hr	13.5±1.0	13.3±1.5	13.0±0.9	12.8±0.8	13.5±0.5	13.5±1.0
24hr	13.8±1.0	13.5±1.0	13.2±0.4	13.7±0.5	13.3±0.8	13.7±1.4
48hr	13.7±1.0	13.5±0.5	13.0±0.9	13.2±0.8	13.2±1.2	13.6±1.3

Values are mean ± SD

Table 7. Body temperature (degrees Fahrenheit) data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	98.0±0.3	98.0±0.3	98.1±0.3	98.2±0.2	98.0±0.4	98.0±0.2
0.25hr	98.1±0.3	98.1±0.2	98.1±0.4	97.9±0.2	98.1±0.2	98.0±0.4
0.5hr	98.1±0.2	98.0±0.6	98.0±0.2	98.1±0.1	98.1±0.4	98.2±0.4
1hr	98.1±0.2	98.1±0.3	98.3±0.3	98.0±0.3	98.1±0.2	98.1±0.3
1.5hr	98.1±0.3	97.8±0.3	98.1±0.2	98.1±0.3	98.1±0.2	98.0±0.5
2hr	98.0±0.3	97.9±0.4	98.2±0.3	98.1±0.2	98.2±0.2	97.9±0.3
4hr	98.1±0.3	98.1±0.2	98.2±0.4	98.1±0.4	98.1±0.3	98.2±0.3
6hr	98.3±0.2	98.1±0.2	98.2±0.5	98.4±0.1	98.1±0.3	98.2±0.3
8hr	98.3±0.3	98.2±0.3	98.1±0.2	98.1±0.6	98.0±0.4	98.2±0.4
24hr	98.1±0.2	98.2±0.1	98.2±0.3	98.1±0.3	98.2±0.4	97.9±0.3
48hr	98.1±0.2	98.3±0.3	98.2±0.2	98.3±0.3	98.0±0.2	98.2±0.1
WOMEN						
Pre	98.1±0.3	98.1±0.4	98.1±0.4	98.0±0.4	97.9±0.4	98.2±0.4
0.25hr	98.2±0.2	98.1±0.3	98.3±0.6	98.2±0.2	98.2±0.2	98.2±0.2
0.5hr	98.1±0.5	98.2±0.2	98.0±0.2	98.0±0.3	98.2±0.2	98.3±0.2
1hr	98.3±0.2	98.3±0.2	98.4±0.2	98.3±0.2	98.1±0.3	98.2±0.2
1.5hr	98.2±0.2	98.3±0.2	98.2±0.2	98.2±0.2	98.1±0.3	98.3±0.1
2hr	98.4±0.2	98.4±0.3	98.3±0.3	98.1±0.2	98.1±0.2	98.1±0.3
4hr	98.2±0.1	98.3±0.4	98.3±0.4	98.2±0.2	98.1±0.3	98.3±0.2
6hr	98.3±0.4	98.5±0.5	98.3±0.4	98.2±0.3	98.4±0.2	98.4±0.2
8hr	98.4±0.3	98.4±0.5	98.3±0.3	98.3±0.1	98.2±0.3	98.4±0.3
24hr	98.3±0.3	98.2±0.3	98.0±0.5	98.1±0.2	98.0±0.2	98.0±0.4
48hr	98.0±0.3	98.3±0.3	98.1±0.4	98.3±0.2	98.1±0.2	98.2±0.5

Values are mean ± SD

With regards to the subjective mood measures, no interactions were noted ($p>0.05$). Condition effects were noted for attentive ($p=0.02$) and energetic ($p=0.02$), with M100 + C150 greater than M100 + T50; and moody ($p<0.01$), with M100 and M100 + T50 greater than M100 + C150. A time effect was noted for focused ($p=0.02$), with a pattern of increase observed from baseline through 4 hours post ingestion, with the three caffeine conditions appearing most influential in driving this effect. Values followed a very similar pattern within all conditions, in that they improved from baseline through 8 hours, likely due to the early morning start time. As subjects "woke up" and started feeling better throughout the day, values improved. While very little noted differences in the subjective mood measures were noted, data are shown in Table 8.

Table 8. Subjective mood data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

A. Attentive

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	51±34	54±39	58±32	60±36	48±36	53±34
0.25hr	58±32	56±36	63±29	63±31	47±33	55±36
0.5hr	62±32	69±32	64±26	66±29	51±35	60±34
1hr	60±33	64±32	66±24	68±26	51±35	58±33
1.5hr	58±34	63±33	68±25	66±26	52±33	62±35
2hr	62±34	66±29	71±26	70±26	58±32	61±34
4hr	62±31	64±30	73±21	67±27	54±35	67±34
6hr	61±33	64±28	66±25	65±31	59±31	60±35
8hr	57±35	56±33	65±25	63±32	53±34	56±35
24hr	52±36	55±37	61±29	60±34	47±36	54±35
48hr	27±18	57±36	54±34	41±26	47±36	66±32
WOMEN						
Pre	67±21	49±28	51±21	64±15	52±31	56±26
0.25hr	57±27	45±22	51±24	61±19	53±23	57±24
0.5hr	57±27	53±21	56±21	64±14	51±25	58±22
1hr	57±21	47±19	59±20	68±13	53±22	65±20
1.5hr	57±24	57±17	58±18	68±15	53±19	69±19
2hr	54±21	59±23	60±16	68±14	50±22	67±16
4hr	59±24	56±28	61±20	65±16	59±19	61±21
6hr	56±27	54±18	60±23	57±14	60±14	68±18
8hr	54±25	47±19	62±23	58±23	61±16	64±22
24hr	65±23	54±18	53±27	69±21	54±21	60±29
48hr	53±30	54±25	50±26	66±14	55±23	52±23

Values are mean ± SD

Table 8. Subjective mood data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

B. Energetic

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	45±36	47±35	57±33	57±39	45±35	51±37
0.25hr	52±36	54±36	61±29	63±30	45±32	56±35
0.5hr	58±34	66±34	63±28	65±29	51±34	59±34
1hr	59±34	61±32	64±25	69±25	52±35	61±33
1.5hr	60±33	62±32	66±25	69±26	51±33	62±35
2hr	65±33	62±28	72±26	71±26	53±33	66±34
4hr	62±31	64±31	78±18	65±29	54±32	62±33
6hr	57±34	62±30	52±37	61±34	56±32	59±35
8hr	60±37	57±34	68±27	63±32	55±34	62±36
24hr	49±37	50±36	61±30	59±36	45±32	52±35
48hr	24±16	59±36	57±33	38±27	49±35	60±40
WOMEN						
Pre	60±22	39±28	48±26	57±25	51±29	53±30
0.25hr	55±24	40±26	51±29	56±23	51±23	51±23
0.5hr	54±23	49±22	57±21	65±15	51±24	56±18
1hr	48±21	48±22	52±26	68±14	49±28	58±17
1.5hr	54±20	53±18	57±22	68±13	55±19	63±20
2hr	50±17	58±23	57±22	63±15	52±21	62±17
4hr	53±24	52±25	59±26	65±16	58±18	57±19
6hr	53±29	46±20	61±26	53±13	59±15	65±14
8hr	51±26	46±18	62±25	58±24	60±13	57±22
24hr	56±27	42±22	47±34	74±14	53±30	54±28
48hr	55±31	60±33	50±30	63±13	51±30	39±25

Values are mean ± SD

Table 8. Subjective mood data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

C. Motivated

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	54±35	51±38	60±31	58±37	46±29	57±36
0.25hr	59±31	55±35	62±28	60±34	43±33	58±33
0.5hr	63±32	61±33	59±36	63±32	51±34	61±34
1hr	61±32	59±28	67±23	68±25	54±33	61±33
1.5hr	61±33	63±32	68±25	67±27	57±30	66±34
2hr	64±32	65±31	74±22	69±26	61±30	65±33
4hr	66±31	67±32	79±17	67±28	60±31	69±34
6hr	61±33	63±27	67±23	62±34	62±28	60±35
8hr	61±37	58±33	69±26	52±40	60±33	61±34
24hr	54±36	54±36	54±36	58±35	47±30	54±36
48hr	24±16	57±38	53±36	36±25	58±35	63±35
WOMEN						
Pre	56±25	40±33	48±27	65±22	51±31	53±38
0.25hr	55±29	44±29	51±25	56±25	47±29	56±32
0.5hr	48±31	50±24	54±21	61±18	49±29	57±24
1hr	50±29	48±20	57±22	63±15	51±26	62±23
1.5hr	55±28	56±21	56±22	67±15	53±4	63±19
2hr	51±26	59±23	57±23	66±18	52±25	66±22
4hr	55±28	52±26	59±29	66±18	61±20	64±20
6hr	53±33	52±17	60±25	48±17	59±18	66±20
8hr	51±28	45±18	58±28	57±22	58±22	59±26
24hr	55±36	41±24	46±34	68±22	50±36	55±28
48hr	52±34	54±33	48±31	62±19	51±32	47±33

Values are mean ± SD

Table 8. Subjective mood data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

D. Irritable

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	15±32	13±21	19±39	11±16	19±30	13±23
0.25hr	18±38	20±40	19±36	20±39	25±34	18±38
0.5hr	19±38	23±43	19±38	20±40	31±42	18±39
1hr	16±36	18±38	20±39	20±39	28±40	17±40
1.5hr	17±38	17±35	20±38	19±39	28±42	17±36
2hr	18±38	17±32	24±40	19±39	27±40	17±37
4hr	15±30	17±30	22±41	19±39	18±33	16±35
6hr	15±32	20±35	22±37	20±39	24±38	15±32
8hr	17±36	21±38	20±38	19±39	22±40	14±31
24hr	13±28	17±33	9±13	4±6	17±31	15±33
48hr	13±18	12±25	7±9	3±6	19±34	20±37
WOMEN						
Pre	8±11	11±14	4±5	3±3	14±20	15±23
0.25hr	6±9	9±12	2±4	4±4	11±18	6±7
0.5hr	6±7	4±4	4±5	3±3	7±11	6±7
1hr	5±5	5±5	4±5	4±4	7±11	5±7
1.5hr	4±4	7±7	3±3	3±4	7±7	4±3
2hr	3±3	3±3	4±6	4±4	4±3	9±13
4hr	4±4	4±5	2±2	3±4	8±14	6±8
6hr	8±12	8±10	2±2	1±1	4±6	8±12
8hr	3±3	7±11	2±3	1±1	5±8	3±4
24hr	3±3	14±24	10±11	4±3	5±7	18±26
48hr	2±1	3±3	5±8	2±1	5±6	10±11

Values are mean ± SD

Table 8. Subjective mood data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

E. Focused

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	48±33	49±39	53±35	57±39	43±34	48±36
0.25hr	52±31	50±34	63±25	63±33	43±35	52±35
0.5hr	63±32	62±35	67±24	63±32	49±35	59±34
1hr	60±36	57±33	67±23	61±30	51±36	54±38
1.5hr	60±37	62±31	70±22	67±28	58±33	63±34
2hr	59±35	65±30	73±22	68±28	59±30	64±37
4hr	64±31	60±35	78±19	68±33	55±37	65±33
6hr	64±34	61±31	71±22	57±42	60±30	57±35
8hr	56±36	57±33	64±29	56±41	51±34	53±32
24hr	47±37	54±38	47±32	50±44	43±32	50±34
48hr	29±20	55±37	54±36	37±29	49±34	62±37
WOMEN						
Pre	51±28	40±36	43±28	58±28	48±34	44±31
0.25hr	51±32	44±30	48±30	58±24	48±30	50±35
0.5hr	49±32	50±24	52±24	57±20	52±29	51±30
1hr	52±31	51±22	57±23	66±16	50±27	61±25
1.5hr	55±29	57±16	58±20	65±15	55±24	62±24
2hr	48±28	61±20	58±22	66±18	55±29	61±22
4hr	51±32	60±28	62±29	67±17	54±21	67±19
6hr	51±37	54±18	60±28	51±18	58±19	60±26
8hr	46±31	46±19	60±29	57±25	60±22	57±24
24hr	50±36	39±29	36±37	67±24	51±33	56±29
48hr	52±30	48±32	53±32	64±15	49±34	39±32

Values are mean ± SD

Table 8. Subjective mood data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

F. Jittery

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	15±33	10±18	8±10	7±11	14±26	10±21
0.25hr	16±33	19±39	10±15	19±40	17±28	17±38
0.5hr	17±37	23±43	12±17	21±44	23±35	18±39
1hr	18±38	20±39	11±20	19±40	23±34	23±39
1.5hr	17±38	18±35	17±30	22±39	22±32	25±40
2hr	18±35	18±31	20±34	23±40	24±37	24±40
4hr	15±31	19±40	19±33	23±39	18±31	19±36
6hr	17±35	21±39	16±31	20±39	18±32	14±31
8hr	18±36	19±40	27±37	19±40	18±32	13±27
24hr	13±28	15±31	4±6	4±6	20±35	13±29
48hr	1±1	12±24	7±8	3±6	15±32	19±34
WOMEN						
Pre	14±19	20±21	14±21	4±4	7±7	10±11
0.25hr	6±9	12±16	6±7	5±4	5±4	6±7
0.5hr	6±7	4±6	6±5	4±3	2±2	9±6
1hr	5±4	4±7	5±3	4±3	4±4	9±12
1.5hr	5±4	5±4	3±3	4±4	3±3	13±16
2hr	5±3	2±2	3±5	10±13	4±6	13±19
4hr	6±6	3±3	3±3	5±4	6±9	11±13
6hr	3±3	7±11	3±2	3±5	2±2	7±10
8hr	2±2	7±10	2±3	2±1	5±8	3±4
24hr	3±2	15±20	5±7	6±9	4±5	8±10
48hr	3±2	8±8	3±2	3±2	2±2	8±10

Values are mean ± SD

Table 8. Subjective mood data of men and women ingesting methylliberine, caffeine, theacrine, or their combination

G. Moody

Variable	Methylliberine 25mg	Methylliberine 100mg	Caffeine 150mg	Methylliberine+ Caffeine 100mg+ 150mg	Methylliberine+ Theacrine 100mg+ 50mg	Methylliberine+ Theacrine+ Caffeine 100mg+ 50mg+ 150mg
MEN						
Pre	7±15	11±14	5±5	5±6	7±7	8±17
0.25hr	8±13	10±12	6±8	5±6	12±13	9±16
0.5hr	4±4	25±34	4±5	4±6	14±14	7±12
1hr	7±11	14±19	6±7	4±4	19±19	4±6
1.5hr	3±3	9±11	8±8	4±7	14±16	5±9
2hr	2±3	7±6	9±9	19±38	12±17	8±14
4hr	6±9	3±3	8±10	3±3	8±9	6±11
6hr	7±12	7±7	8±11	3±2	11±16	7±11
8hr	4±4	10±12	7±12	3±4	9±13	8±9
24hr	4±3	7±8	3±4	3±3	9±14	6±10
48hr	20±33	6±8	5±4	2±4	6±7	4±5
WOMEN						
Pre	6±7	13±13	7±7	3±3	14±18	10±17
0.25hr	6±5	9±11	4±5	4±4	8±8	5±6
0.5hr	7±7	5±8	4±4	5±4	6±6	6±9
1hr	7±6	6±9	5±6	4±3	7±8	6±10
1.5hr	5±6	6±7	7±9	4±3	5±6	5±6
2hr	5±5	4±5	4±6	3±3	5±5	13±28
4hr	5±5	6±8	13±20	6±5	18±21	7±11
6hr	5±6	9±14	3±2	2±2	3±2	5±6
8hr	3±3	6±6	4±4	4±3	7±9	5±8
24hr	5±4	12±18	8±8	4±3	7±7	14±16
48hr	2±2	4±6	3±3	3±3	3±4	15±13

Values are mean ± SD

4. Discussion

Caffeine is a common ingredient contained within dietary supplements marketed to boost energy, performance, and weight loss.[15] There is considerable variation in the caffeine content of pre-workout dietary supplements, although the average of the 100 most popular supplements is approximately 250 mg per serving.[16] As mentioned above, 250 mg is enough to elicit an increase in BP.[9] While 400 mg of caffeine is considered safe for healthy adults, there is some concern about adverse effects that may occur with interactions with other supplement ingredients or additional caffeine consumption.[17] On a survey on pre-workout supplements, over a third of respondents reported that they consume additional caffeine with their pre-workout supplement at least some of the time. Moreover, nearly 20% reported that they sometimes use pre-workout supplements more than once a day,[18] suggesting the amount of caffeine

consumed by some respondents could exceed the recommended caffeine limit. The supplement dosage may also be unsafe for those who are caffeine naïve, sensitive to caffeine, or have certain health conditions such as hypertension.[19-22] Therefore, while most supplements contain a putatively safe level of caffeine, ingestion of caffeine at the provided doses in combination with other ingredients or with additional caffeine may lead to adverse health outcomes.

Theacrine, and more recently methylliberine, have been explored for their potential for boosting "energetic feel" without augmenting undesirable physiological effects associated with caffeine (e.g., increase in BP). Theacrine has repeatedly been found to have little effect on HR and BP while having a positive effect on psychometric parameters.[5-7,12-13]

In the current study, we sought to determine whether methylliberine consumption led to pressor effects alone or in conjunction with caffeine and theacrine, as these are co-ingredients in some dietary supplements. Additionally, we sought to determine whether methylliberine presented similar subjective mood enhancing properties observed in previous studies with theacrine.[5-7]

Similar to what was previously observed with theacrine,[12-13] methylliberine either alone or in combination with caffeine had no significant effect on HR and BP when compared to caffeine alone. Furthermore, we found no significant changes in HR and BP with the addition of theacrine to methylliberine and caffeine (M100 + T50 + C150 versus M100 + C150). In a previous study, methylliberine did not result in a significant change in HR and BP.[14] Both the current and the previous study also found no significant differences in HR and BP with the addition of theacrine to methylliberine.[14] These findings suggest methylliberine has little to no significant pressor effect alone or in combination with theacrine.

All conditions that lead to higher BP were associated with the presence of caffeine. The combination of caffeine and methylliberine resulted in an increase in SBP compared to non-caffeinated treatments, and the combination of theacrine, methylliberine, and caffeine had an increased SBP compared to the combination of methylliberine and theacrine. Caffeine and methylliberine with or without theacrine resulted in a higher DBP compared to the combination of theacrine and methylliberine. The presence of caffeine within all treatments with higher BP suggests that caffeine is the main factor for the increased BP.

The time effects observed for HR and BP appear to be due to experimental design rather than actual treatments, as no time x condition effects were noted. The HR increase at 4 and 8 hours is most likely associated with feedings from the previous hours. The initial increase in BP, particularly during the first hour of the experiment, may be explained by the waking phenomenon.

The current study also explored the effects of methylliberine on subjective moods. Subjects using methylliberine with caffeine felt modestly more energetic and attentive than when they were using methylliberine with theacrine, with no improvement compared to methylliberine alone or other caffeinated combinations. The combination of methylliberine and caffeine was also found to augment mood compared to treatments with the higher dose of methylliberine (100 mg) with or without theacrine (50 mg). This suggests that methylliberine and caffeine may have a synergistic relationship that may improve subjective moods. Unlike theacrine which has repeatedly been shown to have positive effects on subjective mood,[5-7,12] methylliberine alone was not found to

positively alter subjective mood compared to other treatments. Without a negative control, we can't definitively state whether methylliberine altered subjective moods. Previous work exploring combinations of methylliberine (as Dynamine®) and theacrine (as Teacrine®) versus placebo also found changes in main effects for energy, alertness, productivity, and motivation to perform mental tasks, although it was unclear which treatments were different.[14] However, both of these studies were relatively small, 12 subjects with 6 treatments in a crossover design (current study) and 24 subjects split separately into five treatments groups.[14] A larger study will be necessary to assess whether or not methylliberine, either at the current or higher doses, positively alters subjective moods.

While the known physiological effects of caffeine are relatively mild and generally well-tolerated, some degree of caffeine hypersensitivity (likely due to genetic differences in the polymorphic cytochrome P450 1A2 enzyme) is known to exist, leading to more robust changes in HR and BP in some individuals. The lack of significant pressor effects of theacrine and methylliberine in current studies therefore suggest that these compounds could be alternatives to caffeine for sensitive individuals. Caffeine derivatives such as theacrine and methylliberine may also be used to boost the desirable effects of caffeine when the latter is used in lower doses. However, because the currently available information on theacrine and methylliberine is universally derived from studies using small numbers of subjects, we cannot rule out that higher doses of these substances could lead to pressor effects. Additionally, it is possible that some degree of hypersensitivity to these compounds may exist in the general population, as this is unlikely to be captured in studies using small numbers of participants.

5. Conclusion

Acute ingestion of methylliberine alone at doses up to 100mg does not result in a significant increase in HR, BP, respiratory rate, or body temperature in a sample of healthy men and women. Future, larger-scale studies are needed to further evaluate the mood-enhancing effects of methylliberine, alone and in combination with other dietary ingredients.

6. Authors' contributions

RJB was responsible for the study design, data analysis, and manuscript preparation. MB was responsible for study coordination, subject recruitment, data collection, and data entry. JP was responsible manuscript preparation. All authors read and approved the final manuscript.

7. Conflict of Interest Statement

RJB has been a Consultant for and/or Principal Investigator on research studies funded by various dietary ingredient and supplement companies. Other authors declare no competing interests.

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