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Almonds: Food for Fitness

Sponsored by the Almond Board of California

california almonds®

Almonds: Food for Fitness

Primary Investigator: David C. Nieman, DrPH, FACSM
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Research Facility: Appalachian State University Human Performance Laboratory, North Carolina Research Campus

Sponsor: Almond Board of California
OTR-19-NIEMAND-NR-01; ClinicalTrials.gov: NCT04958018

Nieman et al. Front Nutr. 2023 Jan 9;9:1042719. PMID: 36698469.

Disclosures

AFFILIATION/FINANCIAL INTERESTS (prior 12 months)	ENTITIES
Grants/Research Support	Almond Board of California, National Mango Board, Highbush Blueberry Council, Ocean Spray, LycoRed
Scientific Advisory Board/Consultant/Board of Directors	Vitapod
Speakers Bureau	
Stock Shareholder	
Employee	Appalachian State University
Other	The Almond Board had no role in the study design, data collection, analysis and interpretation, the preparation of the manuscript, or the decision to submit the article for publication.

Systems biology approach

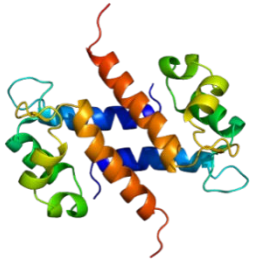
Nat Commun. 2021 Jun 9;12(1):3471;
Med Sci Sports Exerc. 2022;54(5S):S1-S43.

Genomics, transcriptomics, epigenetics, gut microbiome *(sequencers)*

Nutrition interactions

Immune Responses

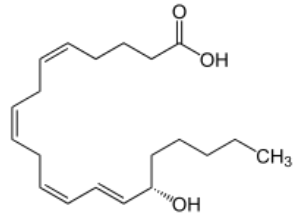
Physiological Stress from Prolonged and Intensive Exercise



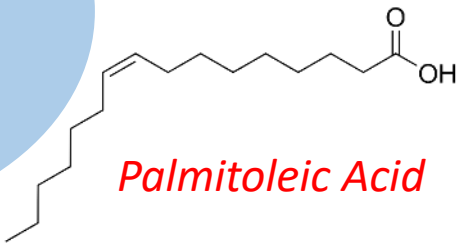
Protein S100-A8

Proteomics *(LC-MS/MS)*

15-Hydroxyicosatetraenoic acid (15-HETE)



Metabolomics, lipidomics, oxylipins *(UPLC-MRM-MS)*



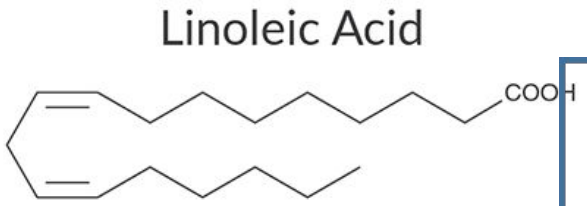
Palmitoleic Acid

Nieman DC, Pence BD. J Sport Health Sci. 2020 Sep;9(5):432-445; Nieman et al. Annu Rev Food Sci Technol. 2019 Mar 25;10:341-363.

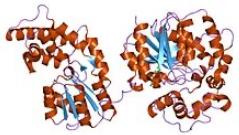
Oxylipins have vital regulatory roles in numerous physiological processes including upstream regulation of inflammation

Oxylipins are bioactive oxidation products derived from n-6 and n-3 PUFAs in the linoleic acid and α-linolenic desaturation pathways; sensitive to exercise and nutrition interventions.

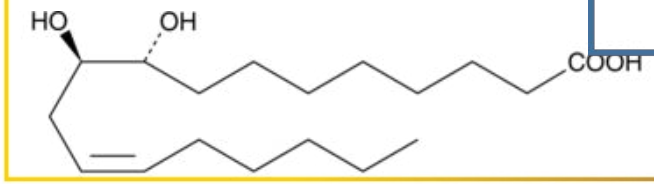
Signini, Nieman et al. Metabolites 2020 Jun 25;10(6):264



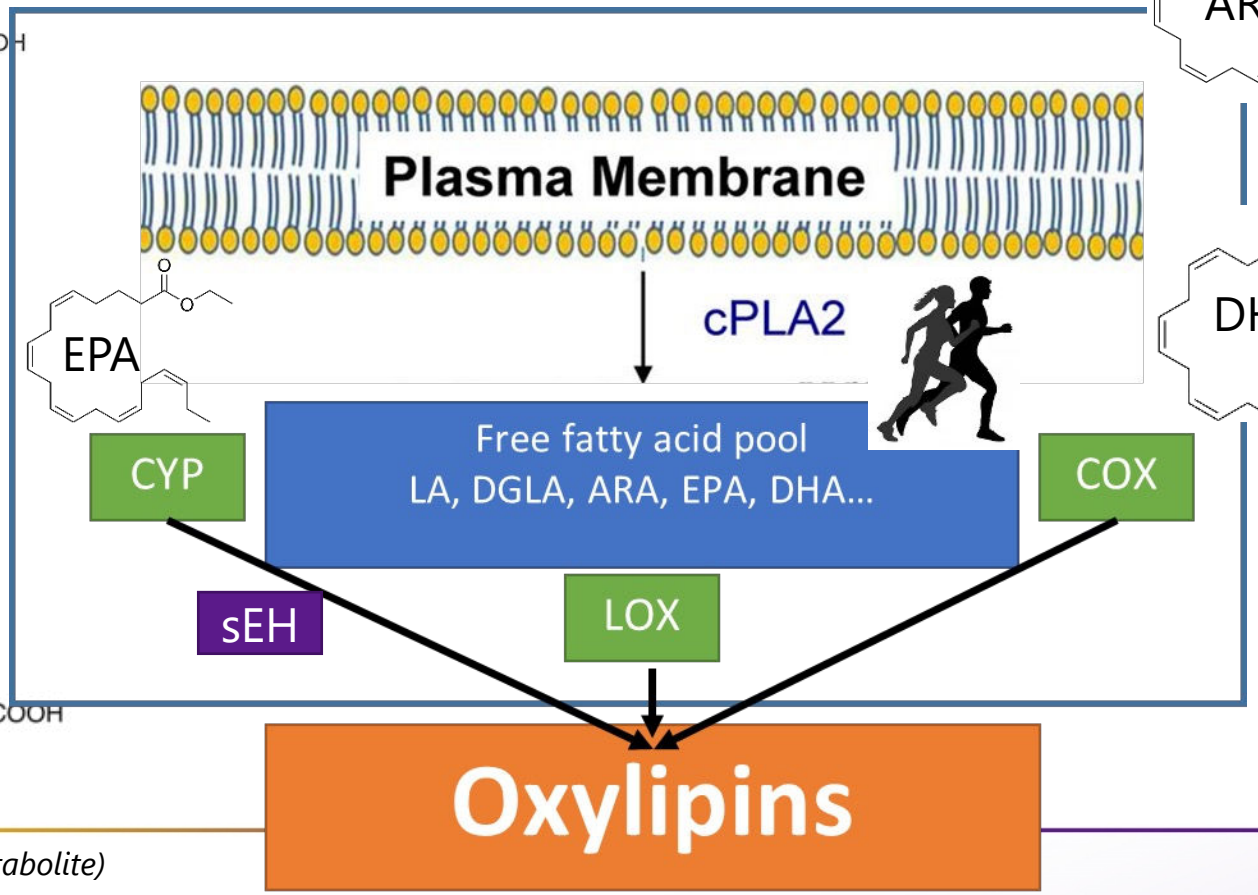
Cytochrome P450



Soluble epoxide hydrolase



9,10-diHOME (oxidized signaling metabolite)



Dr. Qibin Zhang *(Associate Professor, Center for Translational Biomedical Research, UNC Greensboro)*

Physical Exercise

Duration

Workload

Intensity

PLA_2

CYP

LOX

COX

PUFAs
(ω-6 ω-3)

Pro and anti-inflammatory oxylipins

Pro-resolving oxylipins

DiHETrEs
DiHOMEs
HETEs

HDoHEs
DiHETEs
DiHDPEs

SPMs

HETEs
HODEs
LTs

HEPEs
HDoHEs
HOTrEs

SPMs

PGs

TXs

Oxylipins Upstream Regulation

Function depends on the physiological context

Post-exercise condition:
⬆ level of most oxylipins (0-5h)

Inflammatory Process

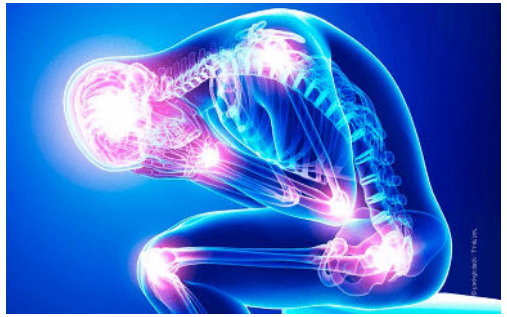
Immune Function

Tissue Repair

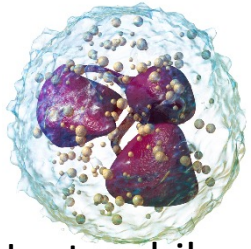
Cardiovascular Functions

Renal Functions

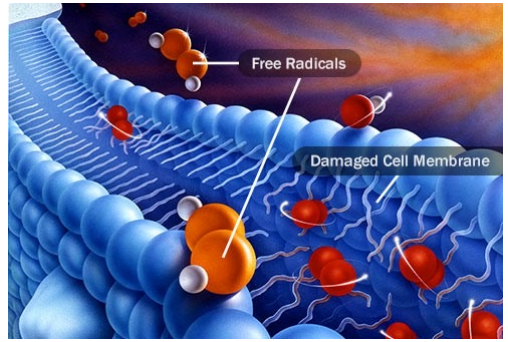
Oxidative Stress



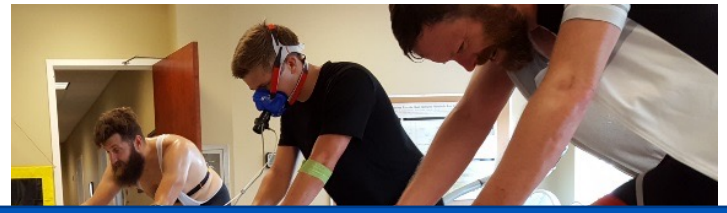
Monocyte



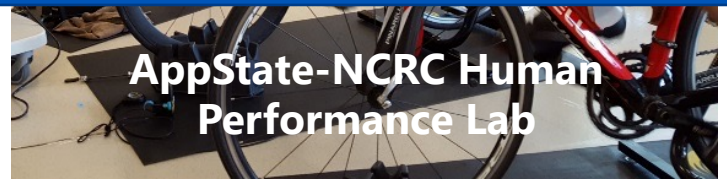
Neutrophil



Exercise and Nutrition Immunology, Multiomics Approach



Plant foods support athletic endeavor and health (compared to sugar water).



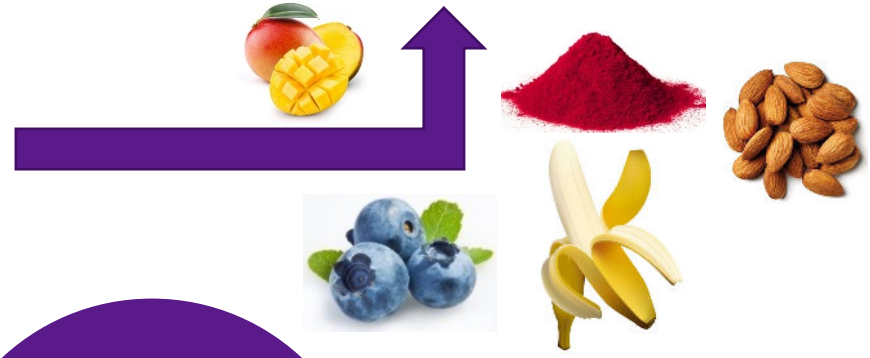
Dr. Camila Olson
Research Manager
Human Performance Lab, NCRC

Mid-1990s

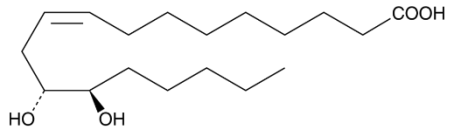
Carbohydrate ingestion

↑performance (2-6%)

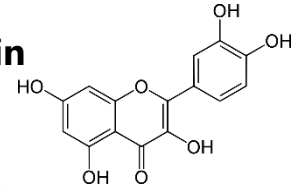
↑ serum glucose, fructose, insulin



12,13-diHOME



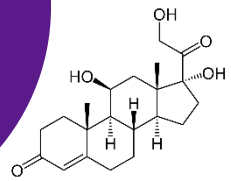
Quercetin



IL-6

↓ inflammation
(reduced neutrophilia, monocytosis, plasma IL-6, IL-1ra, IL-10, MCP-1; muscle IL-6, IL-8 mRNA)

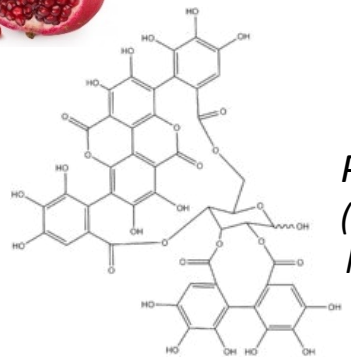
↓ cortisol, epinephrine



Cortisol

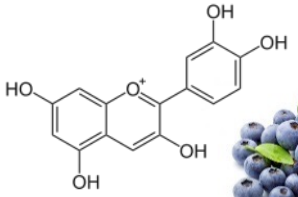
Metabolomics, Proteomics
Plant polyphenols: oxylipin and immune protein responses.

Earlier studies reported few discernable benefits of increased polyphenol intake for athletes: design shortfalls.



Punicalagin (ellagitannin, hydrolyzable tannin)

Anthocyanin

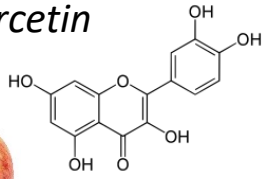


Phytochemicals

- **Phenolics (>8,000)**
  - **Flavonoids**
    - Flavan-3-ols
    - Flavonols
    - Flavones
    - Anthocyanins
    - Flavanones
    - Isoflavonoids
    - Proanthocyanins
  - Coumarins
  - Tannins
  - Stilbenes
  - Lignans
  - Phenolic Acids
- Alkaloids
- Nitrogen-Containing Compounds
- Organosulfur Compounds
- Phytosterols
- Carotenoids

Flavonoid Ingestion

Quercetin



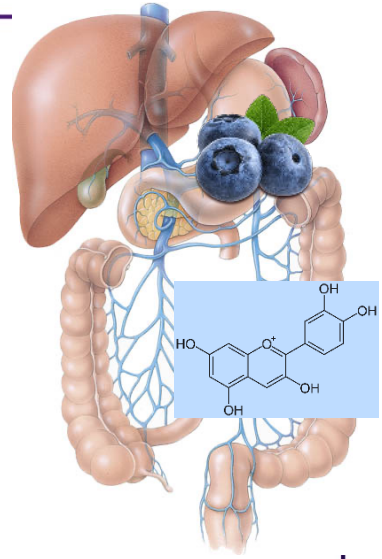
Flavonoid Metabolism

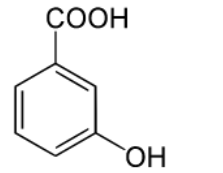
Small Intestine
(little absorption)

Large Intestine
(microbial degradation to smaller phenolics; translocation to liver & blood accelerated by exercise)

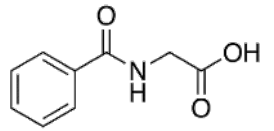
Liver

Circulation, Tissues
(Gut-derived phenolics exert bioactive effects)





Hydroxybenzoic Acid



Hippuric Acid

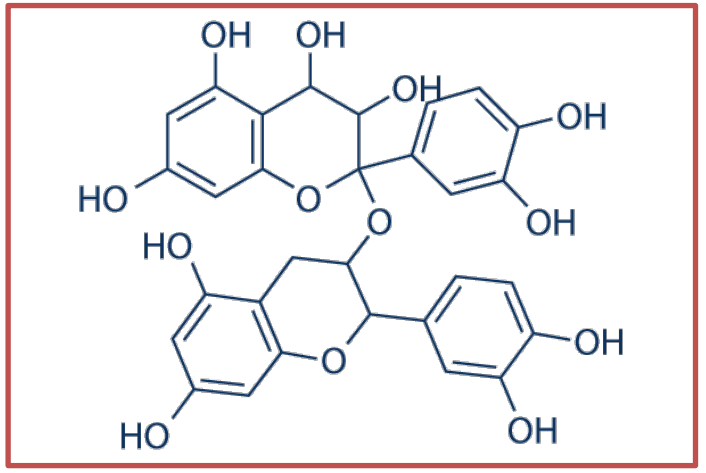
anti-inflammatory, antioxidant, anti-pathogenic, immune regulatory effects

Nutrients ***2017****, 9, 513; Eur. J. Nutr. 2016, 55, 1359–1375; Annu Rev Food Sci Technol. 2019 Mar 25;10:341-363*

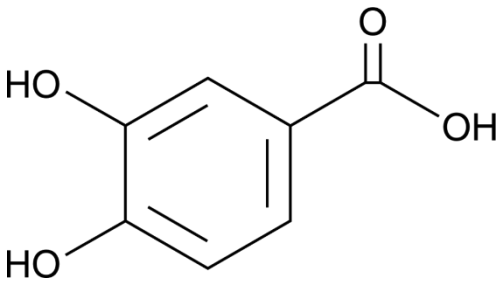
HYPOTHESIS: Almonds provide a unique and complex nutrient and polyphenol mixture that may support metabolic recovery from stressful levels of exercise. The total (poly)phenol content of almonds is **164 mg /** 57-g serving.

Raw almonds, key nutrients	100 g raw almonds	57 g raw almonds	Male RDA	Female RDA	%RDA Males	% RDA Female
Kilocalories	579	324				
Protein (g)	21.2	11.9				
Arginine (g)	2.47	1.4				
Total fat (g)	49.9	28.0				
Monounsaturated fat (g)	31.6	17.7				
Carbohydrate (g)	21.6	12.1				
Fiber (g)	12.5	7.0				
Sugars (g)	4.35	2.44				
Calcium (mg)	269	151	1000	1000	15.1	15.1
Iron (mg)	3.71	2.1	8	18	26.3	11.7
Magnesium (mg)	270	151	400	320	37.8	47.2
Zinc (mg)	3.12	1.76	11	8	16.0	22.0
Copper (mg)	1.031	0.58	0.9	0.9	64.0	64.0
Manganese (mg)	2.18	1.23	2.3	1.8	53.5	68.3
Vitamin E (ug alpha-TE)	25.6	14.4	15	15	96.0	96.0
Leucine (g)	1.47	0.834				

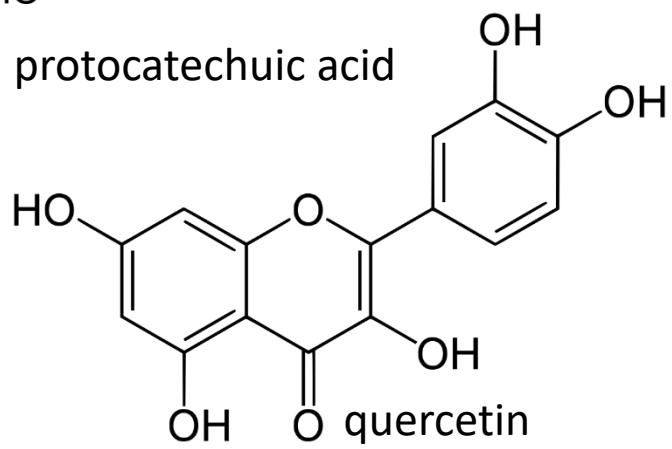
- (Poly)phenols in almonds include
  - flavonols (kaempferol, isorhamnetin, and quercetin),
  - flavanols (several types of catechins),
  - flavanones (eriodictyol and naringenin),
  - simple phenolic acids such as protocatechuic acid and vanillic acid.
- **Proanthocyanidins (PACs)** are the most abundant class of (poly)phenols in almonds.



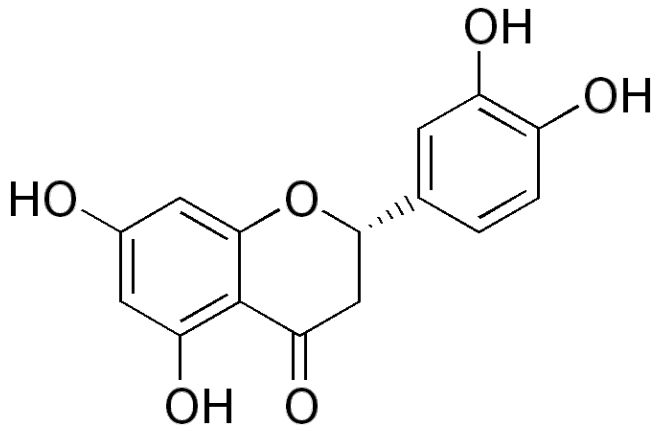
Proanthocyanidins are oligo- or polymers of monomeric flavan-3-ols



protocatechuic acid



quercetin



naringenin

Previous Studies

Metabolic Signatures Induced by Heavy Exertion and Pistachios

Nieman et al. PLoS One. 2014 Nov 19;9(11):e113725.

Funding: American Pistachio Growers

Research Design

2-wks supplementation
3 oz/d pistachio nuts or NONE, randomized assignment.

75-km cycling trial
(with 3 oz pistachio nuts or water only)

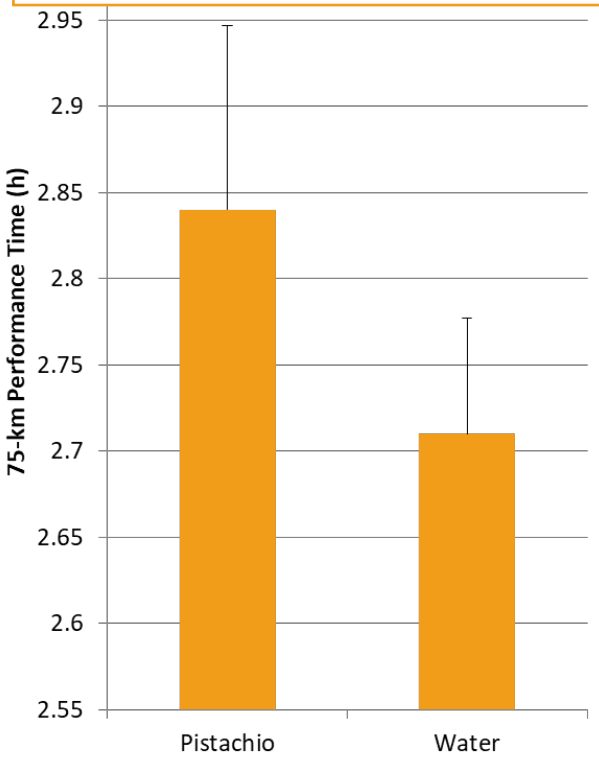
2-wks washout, then crossover

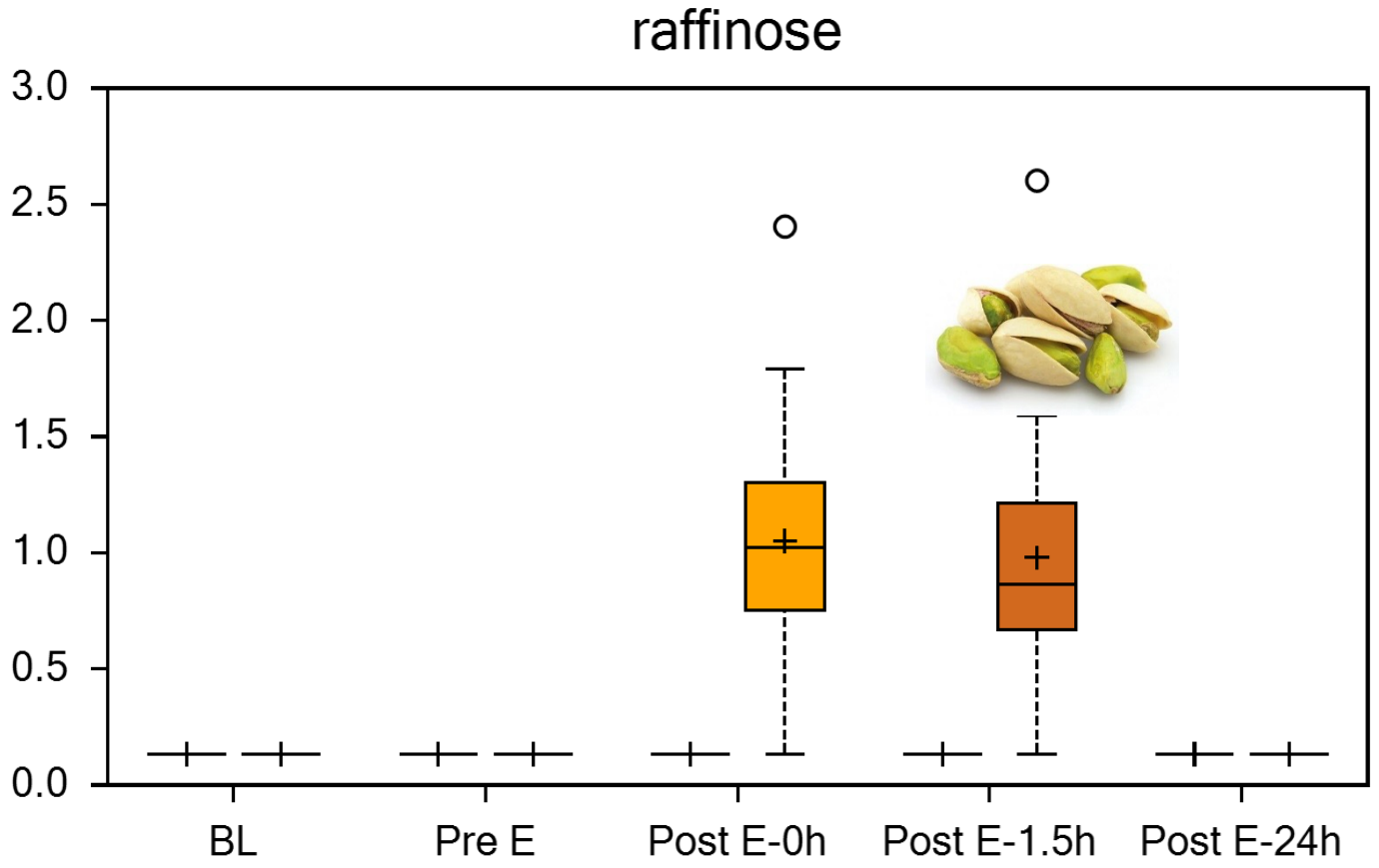
2-wks supplementation
(opposite treatment)

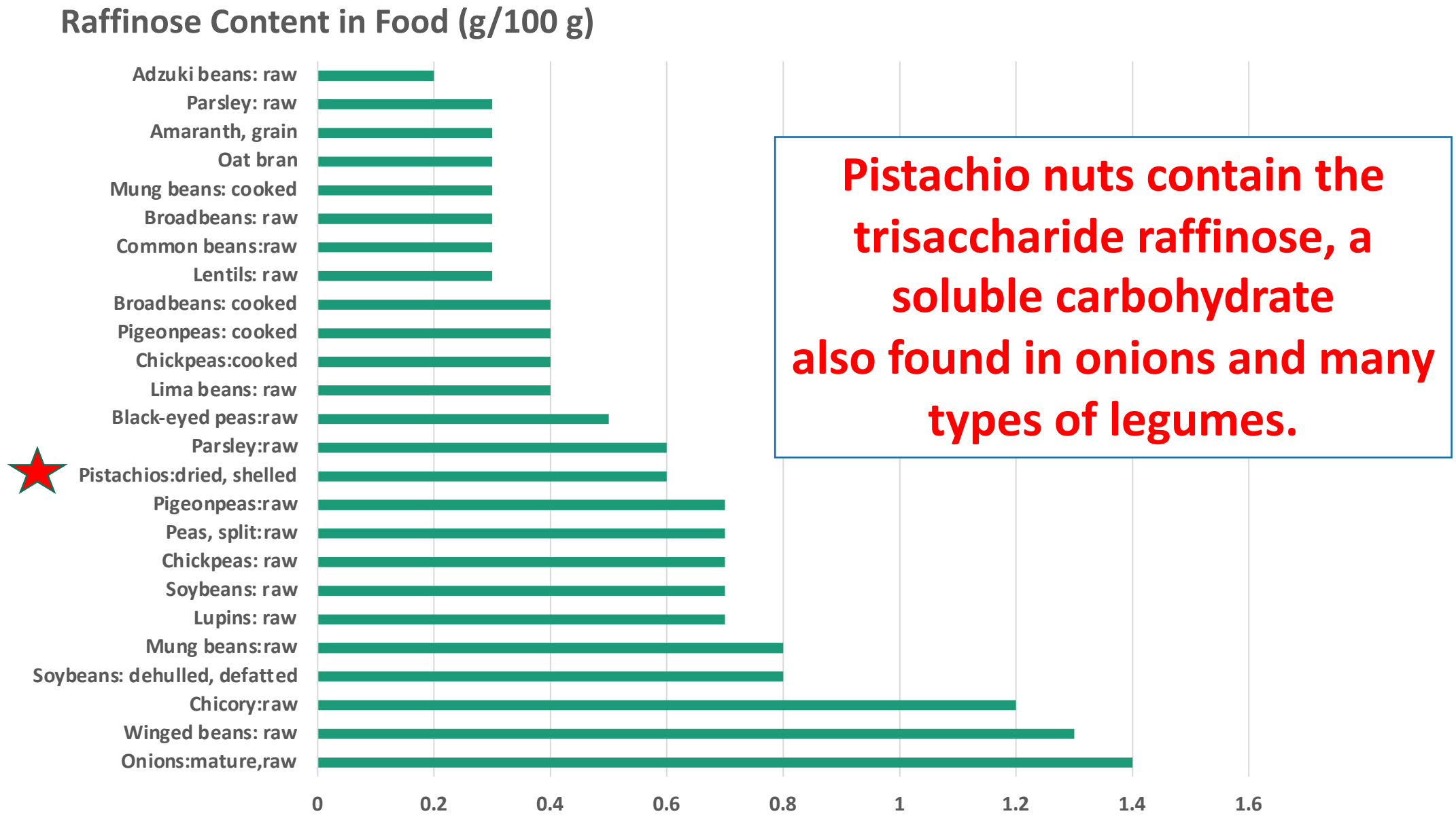
75-km cycling trial
(with 3 oz pistachio nuts or water only)

=blood draw

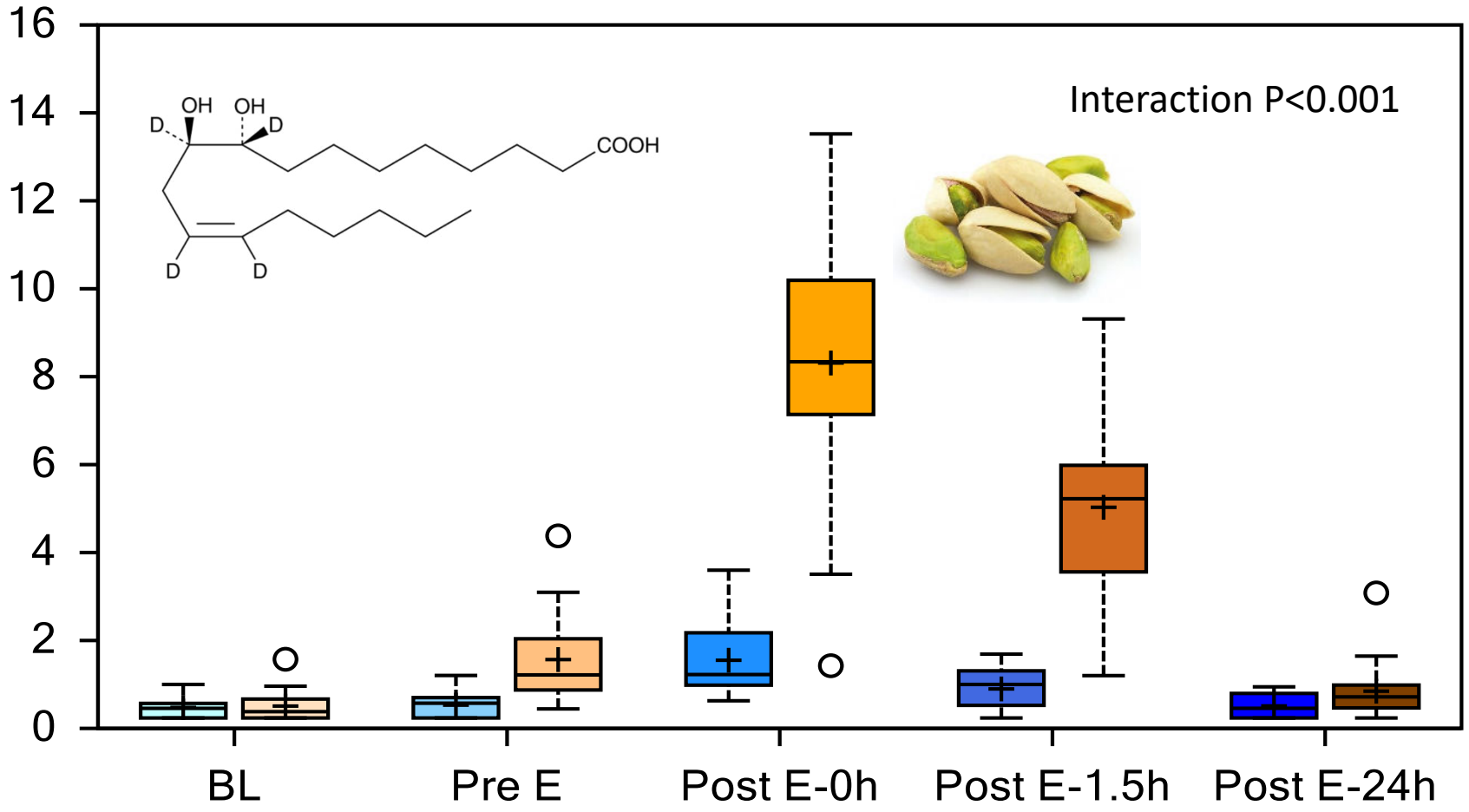
75-km Performance
time (hours) 4.8% slower





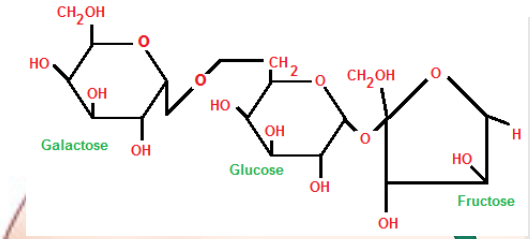

Raffinose Content in Food (g/100 g)
Adzuki beans: raw
Parsley: raw
Amaranth, grain
Oat bran
Mung beans: cooked
Broadbeans: raw
Common beans:raw
Lentils: raw
Broadbeans: cooked
Pigeonpeas: cooked
Chickpeas:cooked
Lima beans: raw
Black-eyed peas:raw
Parsley:raw
Pistachios:dried, shelled
Pigeonpeas:raw
Peas, split:raw
Chickpeas: raw
Soybeans: raw
Lupins: raw
Mung beans:raw
Soybeans: dehulled, defatted
Chicory:raw
Winged beans: raw
Onions:mature,raw
0
0.2
0.4
0.6
0.8
1
1.2
1.4
1.6
Pistachio nuts contain the trisaccharide raffinose, a soluble carbohydrate also found in onions and many types of legumes.

9,10-DiHOME

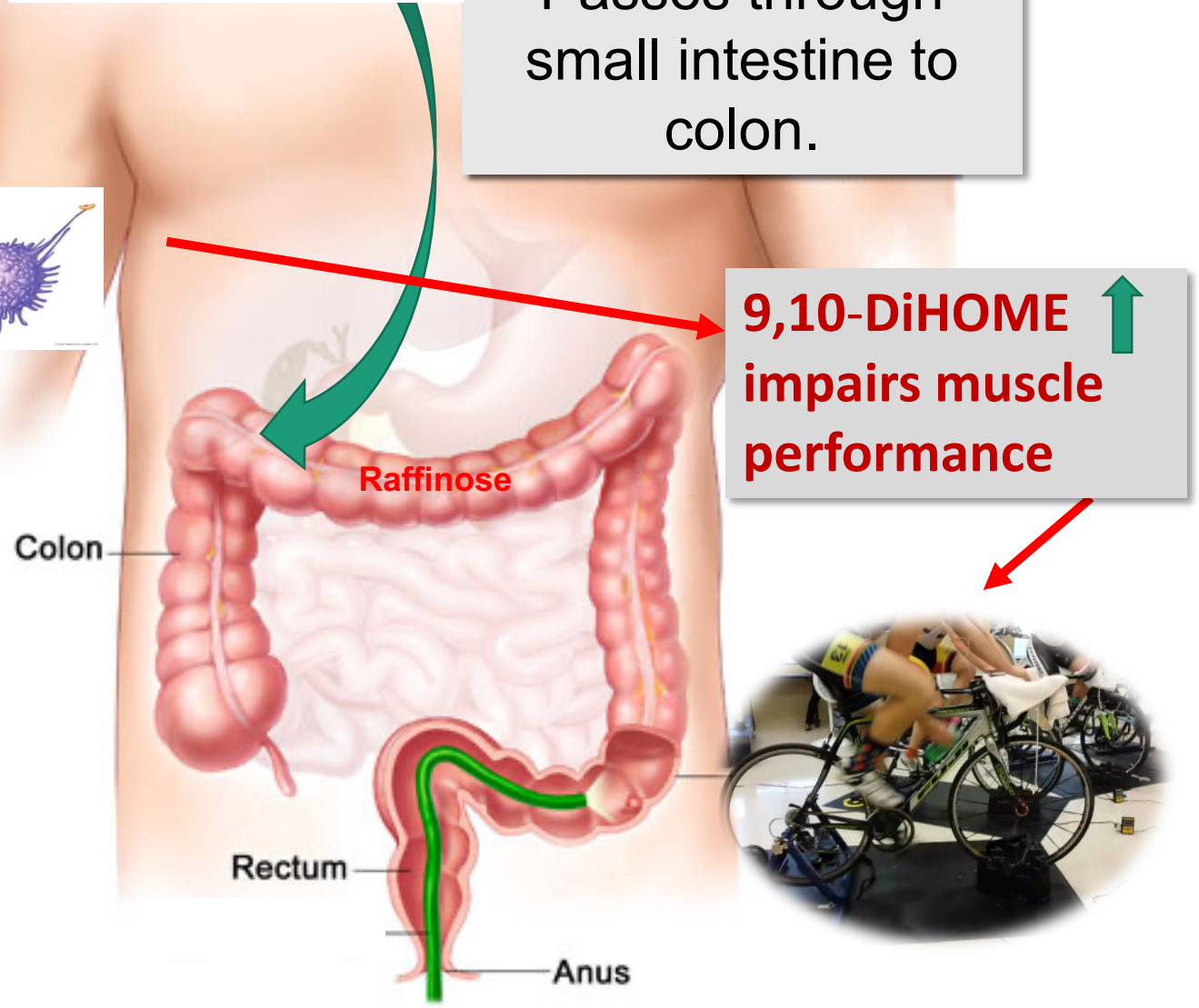


- 9,10-DiHOME is a leukotoxin derivative of linoleic acid and is produced by neutrophils and macrophages.
- Disrupts mitochondrial function by altering inner membrane integrity and increasing cytochrome C release.

Raffinose Blood Signature:
Enhanced by the “leaky gut” effect of exercise.



Raffinose Ingestion:
Passes through small intestine to colon.



9,10-DiHOME impairs muscle performance

N=66 males & females
(randomized treatment)

- Almonds (57 g/d) (n=33)
- Snack bar (324 calorie matched) n=33)

Outcome Measures

Energy, vitality: Profile of Mood States POMS), performance
Soreness: Delayed onset of muscle soreness (DOMS)

Inflammation: 70 oxylipins, lipid mediators
Inflammation: 6 cytokines and C-reactive protein (CRP)
Muscle damage: creatine kinase, myoglobin, LDH

Urine metabolites from 4-weeks almond ingestion;
3-day food record (flavonoids, macro- and micro-nutrients)

Visit #1 Pre-Study Consent, Orientation

Visit #2
Body composition and Muscle Function: Vertical jump, bench press, leg-back strength, shuttle run, Wingate
POMS; DOMS; Blood and 24h urine samples

4-Weeks Supplementation *(57 g almonds/d or 324 kcal/d snack bars; split dose, am, pm)*

3-d food record

Visit #3: Monday: Eccentric Exercise Bout

POMS; DOMS; Blood & 24h urine samples

Half snack dose

Muscle Function: Vertical jump, bench press, leg-back strength, shuttle run, Wingate

90-min eccentric exercise

POMS; DOMS; Blood

Muscle Function: Vertical jump, bench press, leg-back strength, shuttle run, Wingate

Half snack dose, PM

Visit #4: Tuesday Recovery
(7 am POMS, DOMS, blood sample, half snack dose before muscle function testing, and then afternoon)

Visit #5: Wednesday Recovery
(7 am POMS, DOMS, blood sample, half snack dose before muscle function testing, and then afternoon)

Visit #6: Thursday Recovery
(7 am POMS, DOMS, blood sample, half snack dose before muscle function testing, and then afternoon)

Visit #7: Friday Recovery
(Repeat body composition; 7 am POMS, DOMS, blood sample, half snack dose, muscle function testing)

Final Group Subject Statistics

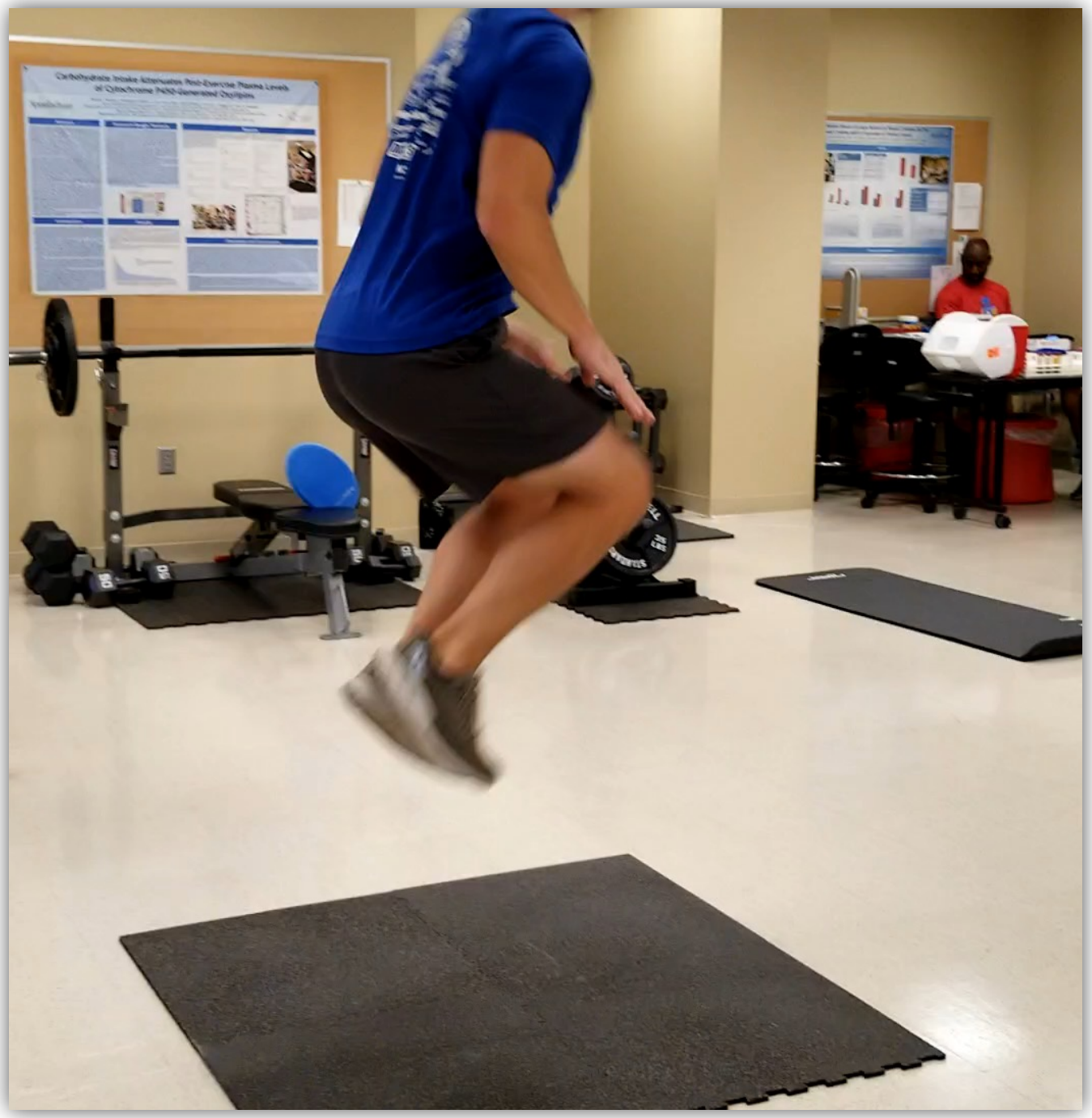
[Almond n=33 (20 males, 13 females)]

[Snack Bar n=31 (18 males, 13 females)]

	Supplement Group	Mean	Std. Deviation	Std. Error Mean
Age (yr)	Almond	46.06	8.39	1.46
	Snack Bar	46.77	9.04	1.62
Weight (kg)	Almond	77.40	11.24	1.96
	Snack Bar	79.74	10.11	1.82
Height (cm)	Almond	173.63	9.75	1.70
	Snack Bar	171.50	7.73	1.39
BMI	Almond	25.56	2.12	0.37
	Snack Bar	27.08	2.70	0.49
Body Fat (%)	Almond	27.43	7.75	1.35
	Snack Bar	28.84	9.98	1.82

- **Healthy, non-obese, non-athletes and not engaged in regular resistance training.**
- **NOTE: A limitation of the study is that it only included non-smoking participants without obesity who exercised occasionally; therefore, we cannot generalize the findings to other demographic and health status groups**

N=64 untrained subjects.
90 min eccentric exercise bout: 17 different exercises, multiple sets




Appalachian
STATE UNIVERSITY

Performance Testing

- 1. Vertical jump: Countermovement vertical jump, repeated three times, with the best score recorded as the difference between the jump and standing reach heights.
- 2. Leg-back strength: Grasp a bar attached via a chain to a force measuring device with straight arms, and then lift up with maximal effort for several seconds (best of 3 attempts).
- 3. Bench press to exhaustion: Bench press a weighted bar equal to 50% (female) 75% (male) body weight as many times as possible (to a metronome set at 60 beats/min or 30 lifts/min).
- 4. 30-second Wingate cycling test: Sprint cycle at maximal speed for 30 seconds on Lode cycle ergometer adjusted to weight (7 watts per kilogram) (peak power, mean power, adjusted to body mass).
- 5. 60 yard shuttle run test: Participants run to 5 yards, 10 yards, 15 yards, there and back, for a total of 60 yards.

Post-90-minutes exercise blood draw

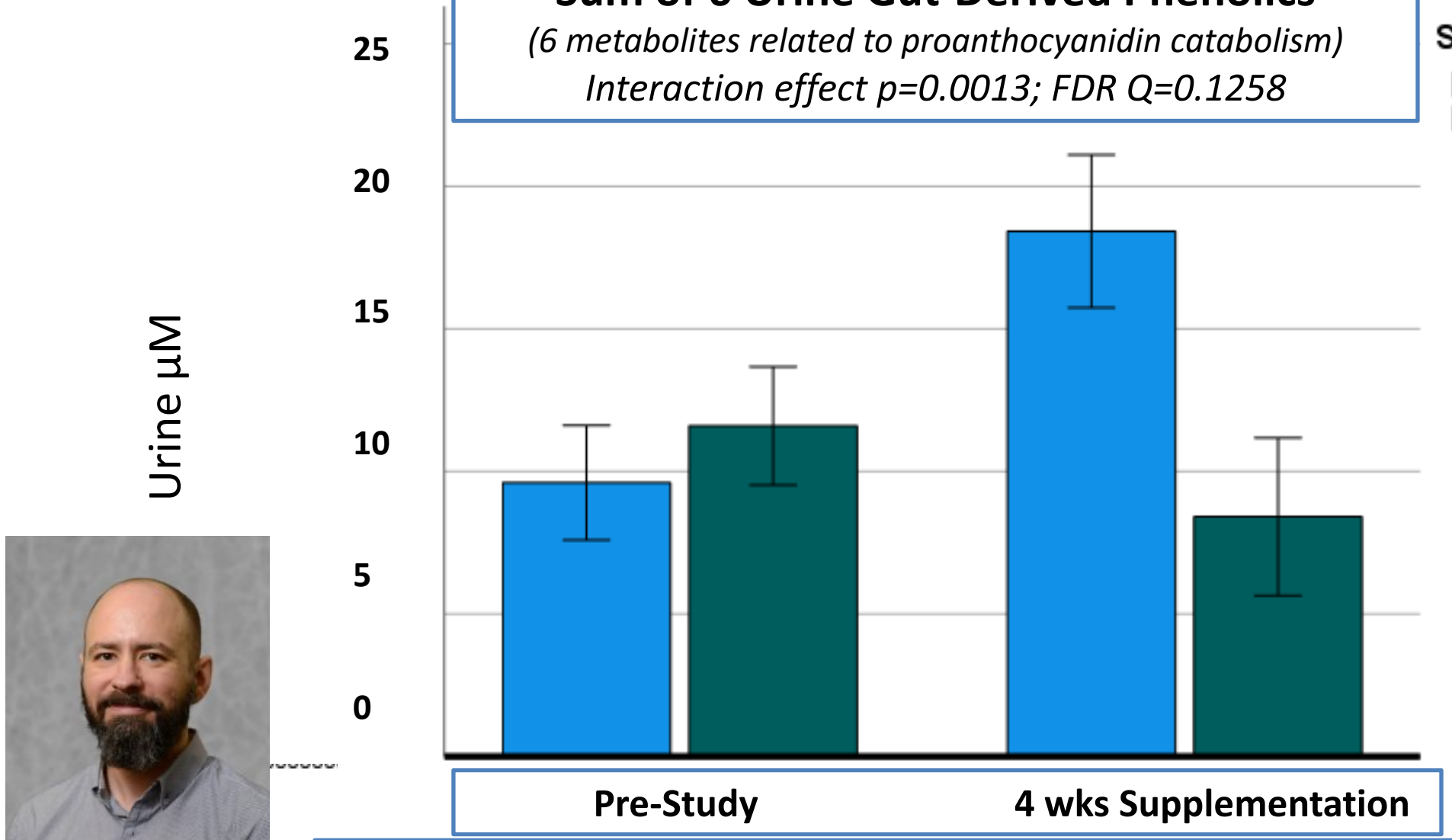
7 Blood Samples and 2 urine samples: Outcomes

1. Creatine kinase, myoglobin, LDH (muscle damage)
2. C-reactive protein and six cytokines (IL-6, IL-8, IL-10, IL-1ra, MCP-1, GCSF) (inflammation)
3. Comprehensive diagnostic chemistry panel
4. Urine almond gut-derived metabolites
5. Plasma oxylipins (n=70) (inflammation)

Composite Variable
Sum of 6 Urine Gut-Derived Phenolics

(6 metabolites related to proanthocyanidin catabolism)

Interaction effect p=0.0013; FDR Q=0.1258

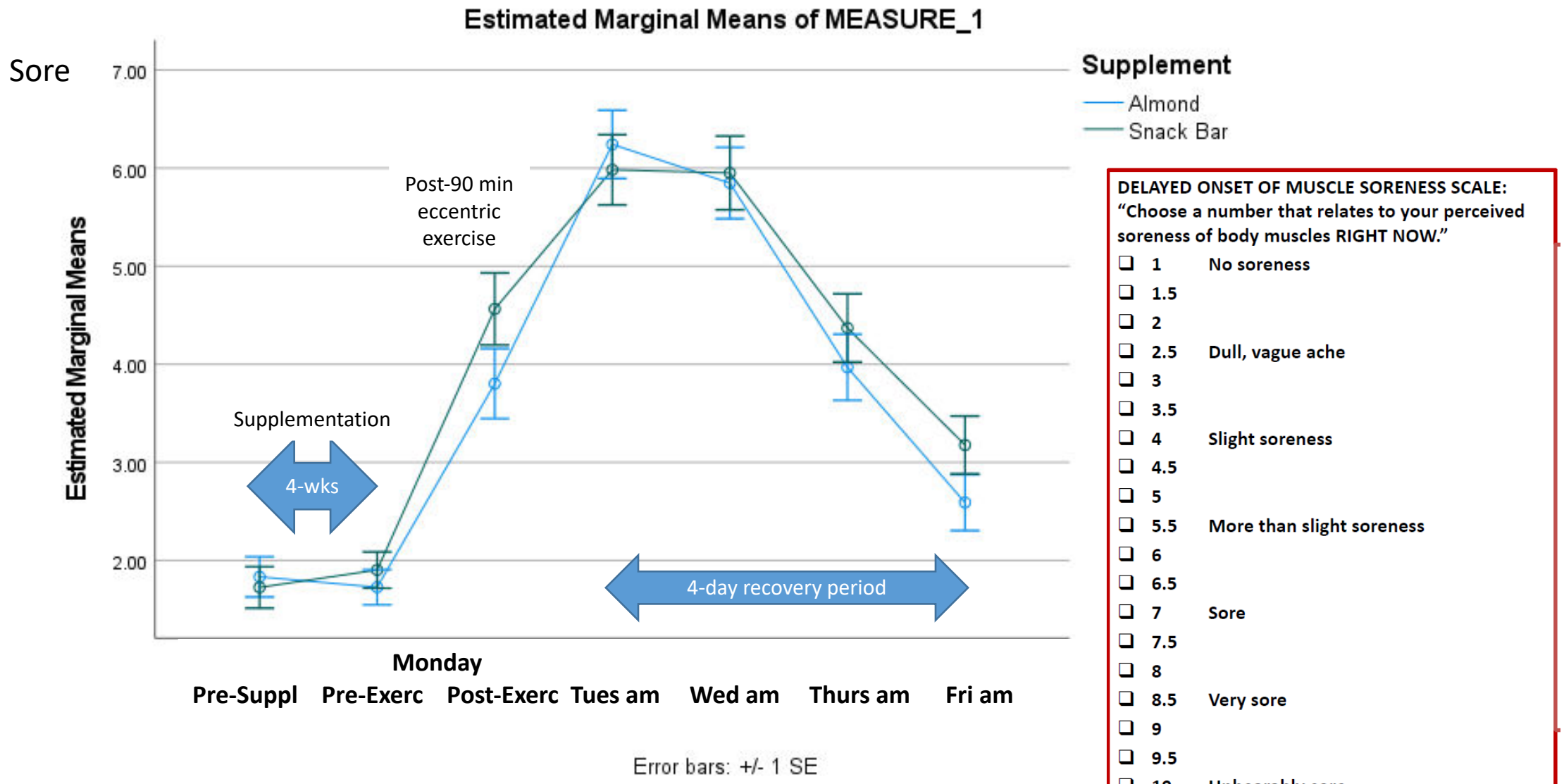


Dr. Colin Kay

5-(3',4'-Dihydroxyphenyl)-γ-valerolactone; hydroxybenzoic acid-sulfate* (2 isomers of unknown structural configuration); 5-hydroxyphenylpropanoic acid-3-sulfate; benzoic acid-4-sulfate; 5-hydroxyphenylpropanoic acid-3-O-glucuronide; 3-(4-methoxyphenyl)propanoic acid-sulfate

Delayed Onset of Muscle Soreness (DOMS) (1-10 scale)

Time effect, p<0.001; Time x treatment effect, p=0.187; Interpretation: The 90-minute eccentric exercise protocol had a significant effect in increasing muscle soreness, with no treatment effect (almonds vs snack bar).

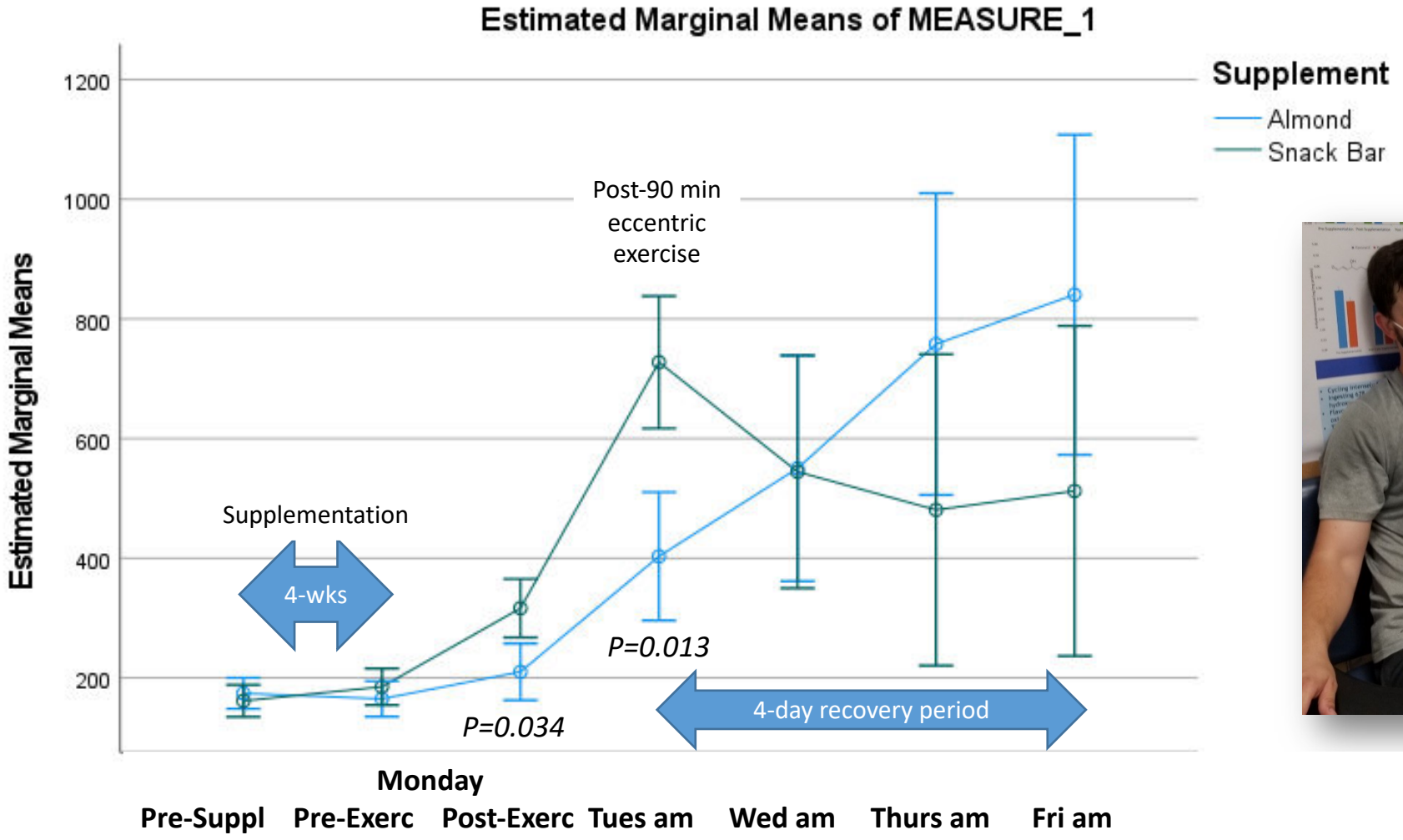


DELAYED ONSET OF MUSCLE SORENESS SCALE:
"Choose a number that relates to your perceived soreness of body muscles RIGHT NOW."

❑	1	No soreness
❑	1.5	
❑	2	
❑	2.5	Dull, vague ache
❑	3	
❑	3.5	
❑	4	Slight soreness
❑	4.5	
❑	5	
❑	5.5	More than slight soreness
❑	6	
❑	6.5	
❑	7	Sore
❑	7.5	
❑	8	
❑	8.5	Very sore
❑	9	
❑	9.5	
❑	10	Unbearably sore

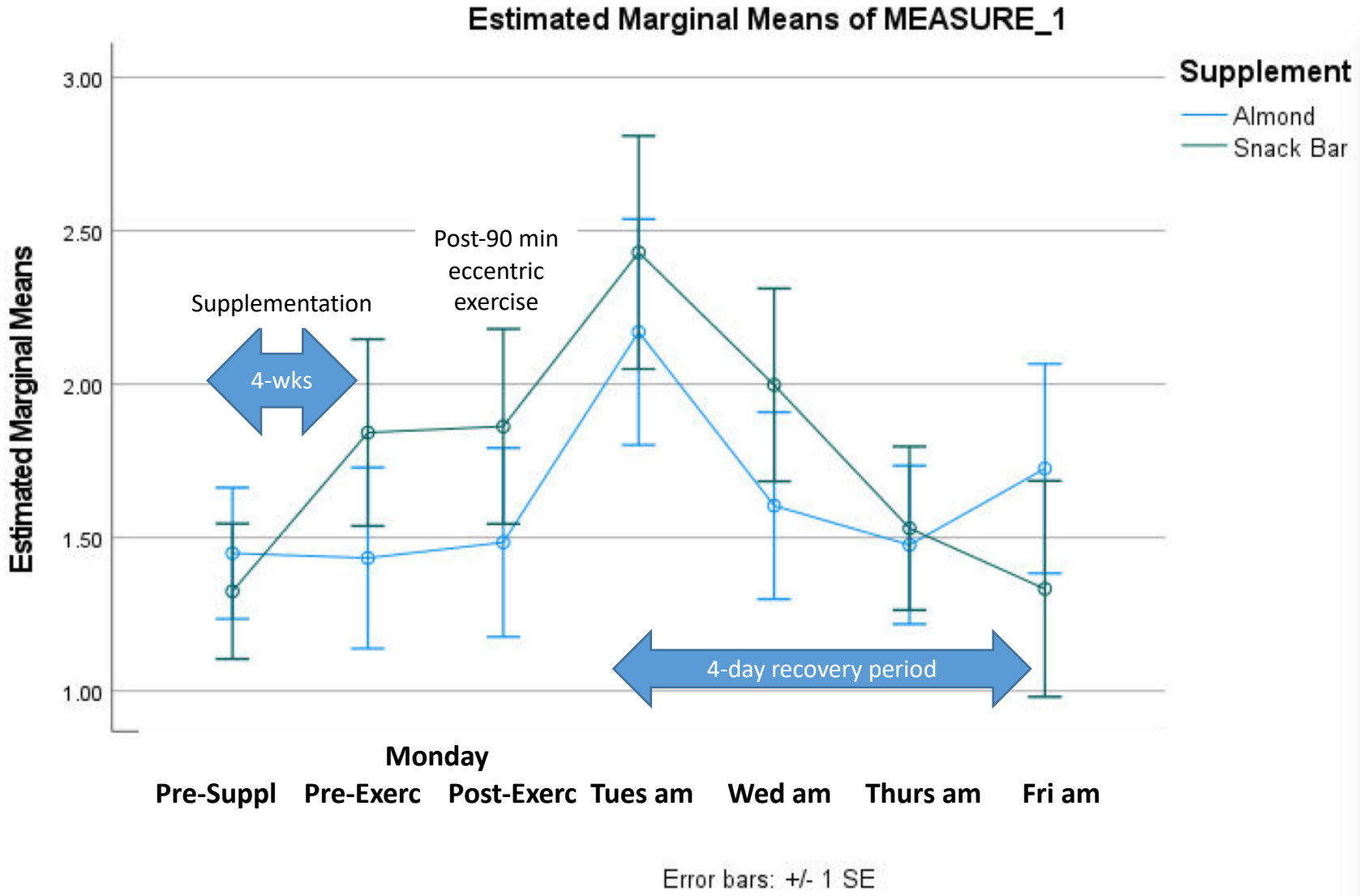
Serum Creatine Kinase

Time effect, p<0.001; Time x treatment effect, p=0.151; Interpretation: The 90-minute eccentric exercise protocol had a significant effect i increasing muscle damage (to a large and sustained level). The overall interaction effect was not significant, but change in CK differed betw groups immediately post-exercise and after 1-day of recovery (i.e., lower muscle damage at those time points for the almond group)..



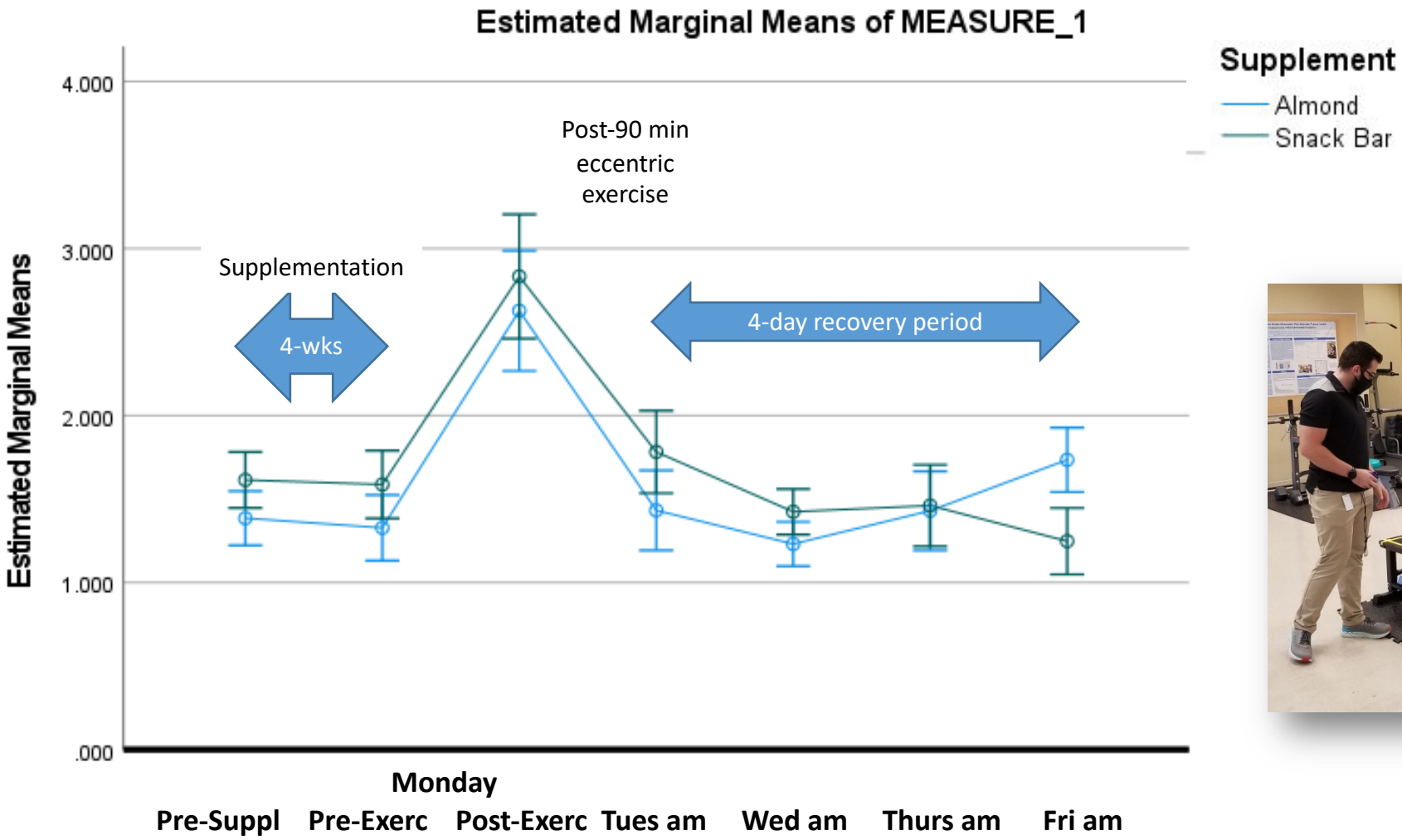
Serum C-Reactive Protein (CRP)

Time effect, p<0.001; Time x treatment effect, p=0.145; Interpretation: The 90-minute eccentric exercise protocol had a significant effect in increasing inflammation, with no treatment effect (almonds vs snack bar). (Weak trend for an almond anti-inflammatory effect).



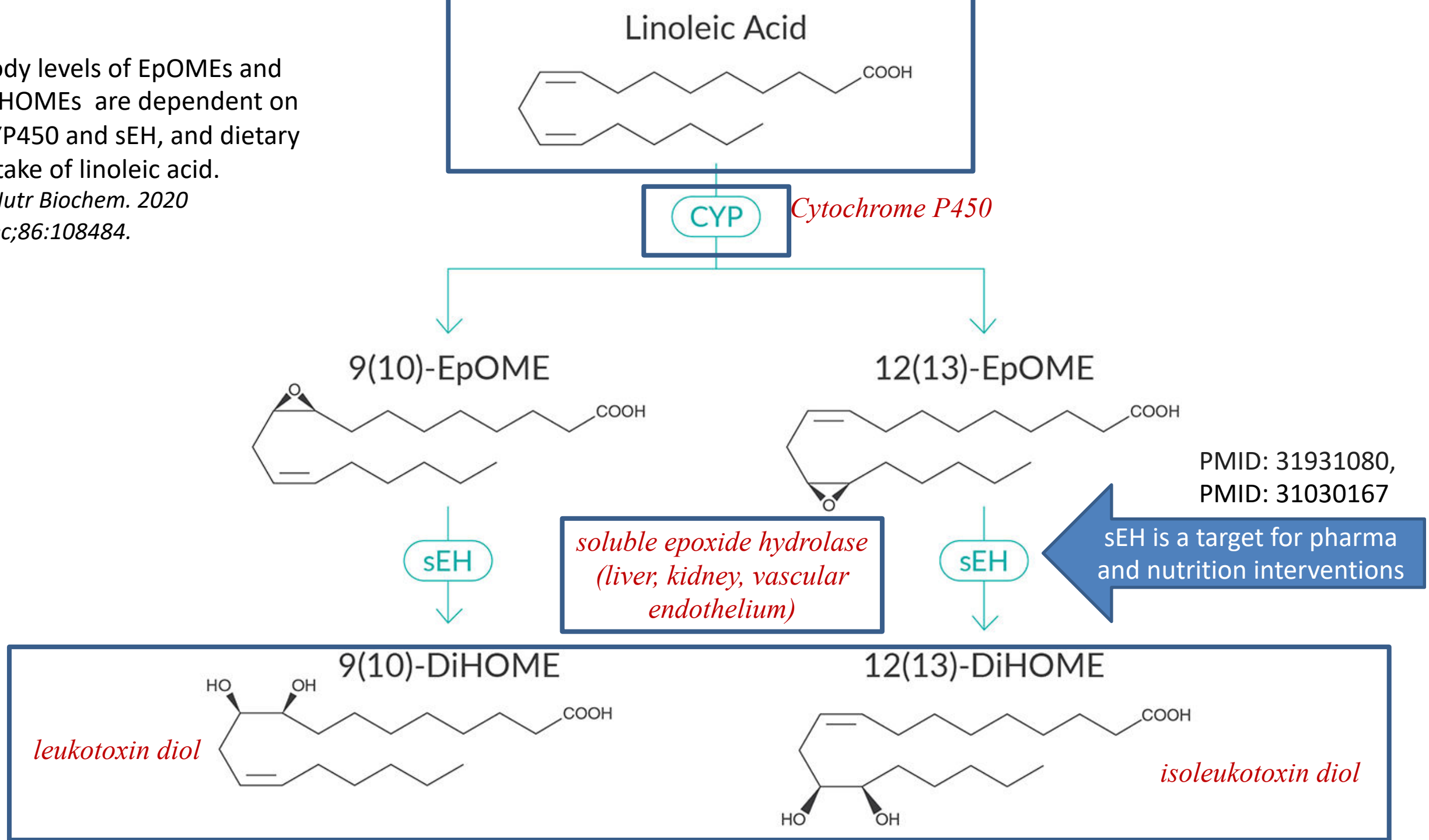
RESULTS: Plasma Cytokine IL-6

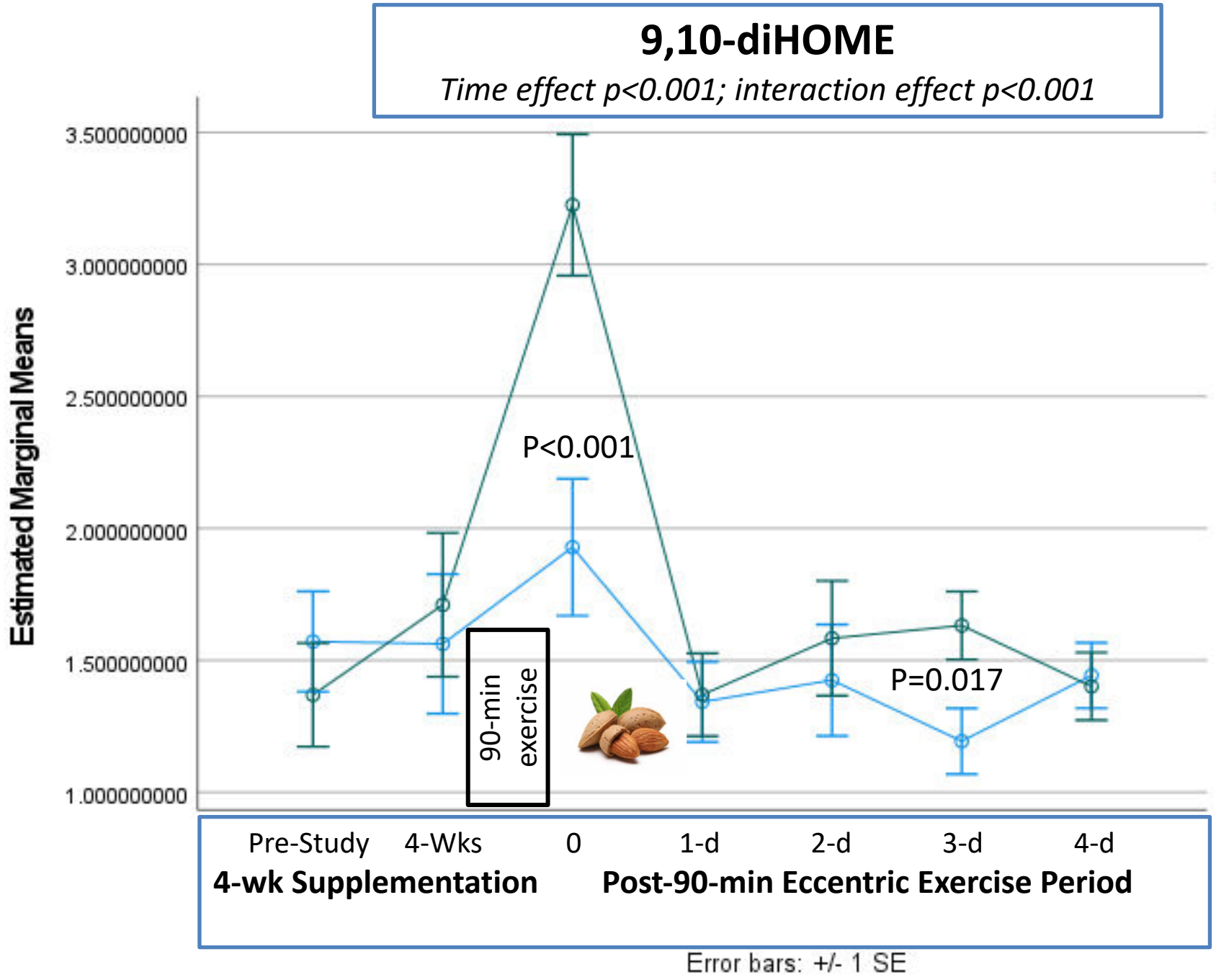
Time effect, p<0.001; Time x treatment effect, p=0.323; Interpretation: The 90-minute eccentric exercise protocol had a significant effect in increasing IL-6 (a measure of inflammation), with no treatment effect (almonds vs snack bar)



Body levels of EpOMEs and DiHOMEs are dependent on CYP450 and sEH, and dietary intake of linoleic acid.

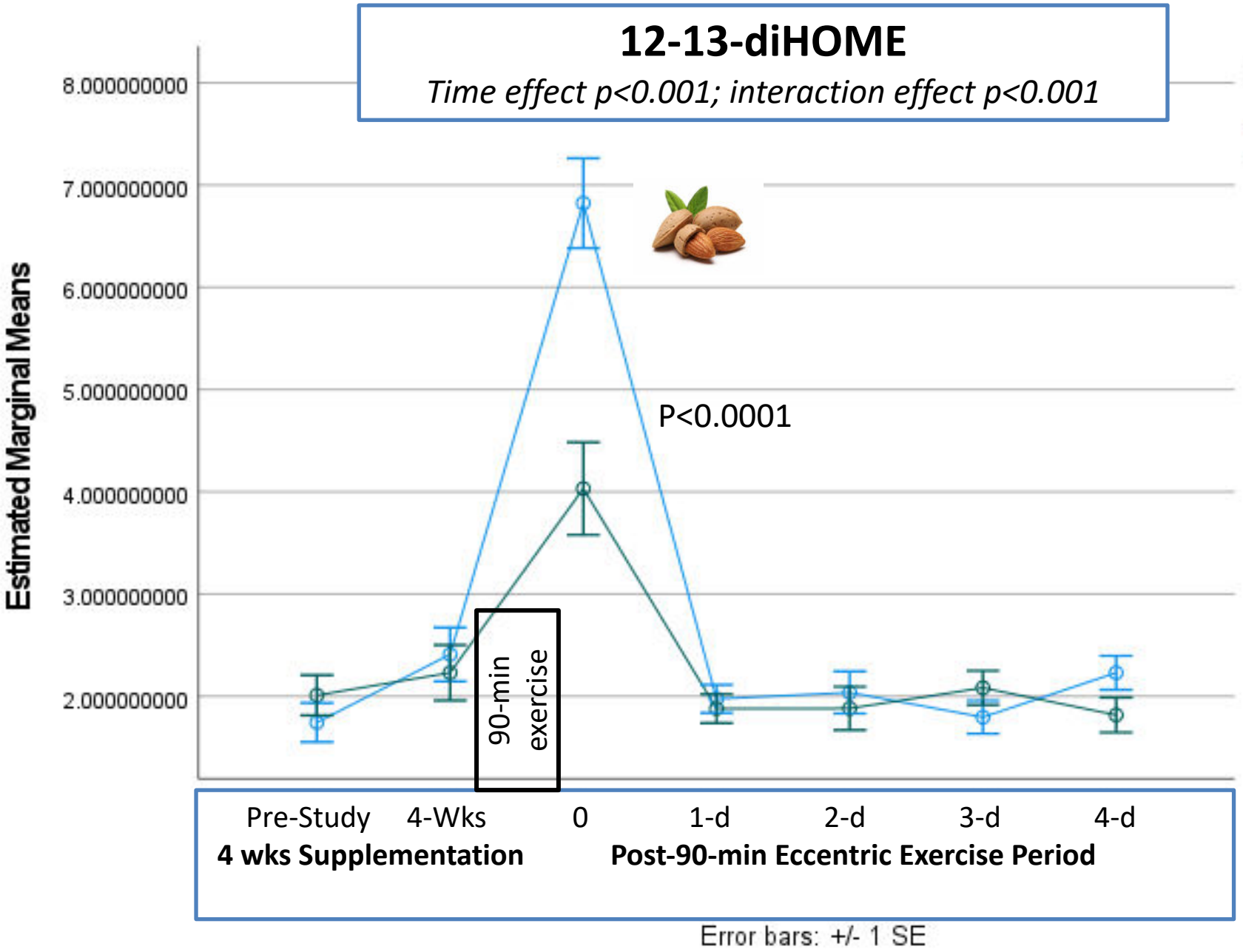
J Nutr Biochem. 2020 Dec;86:108484.



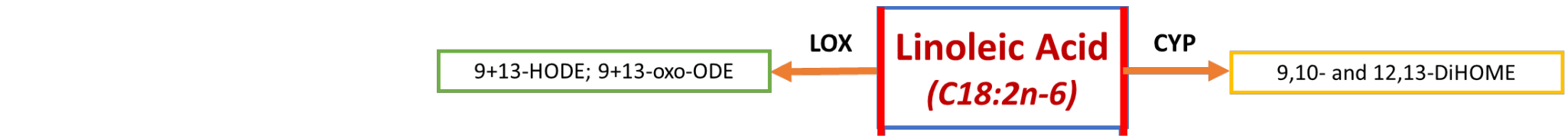


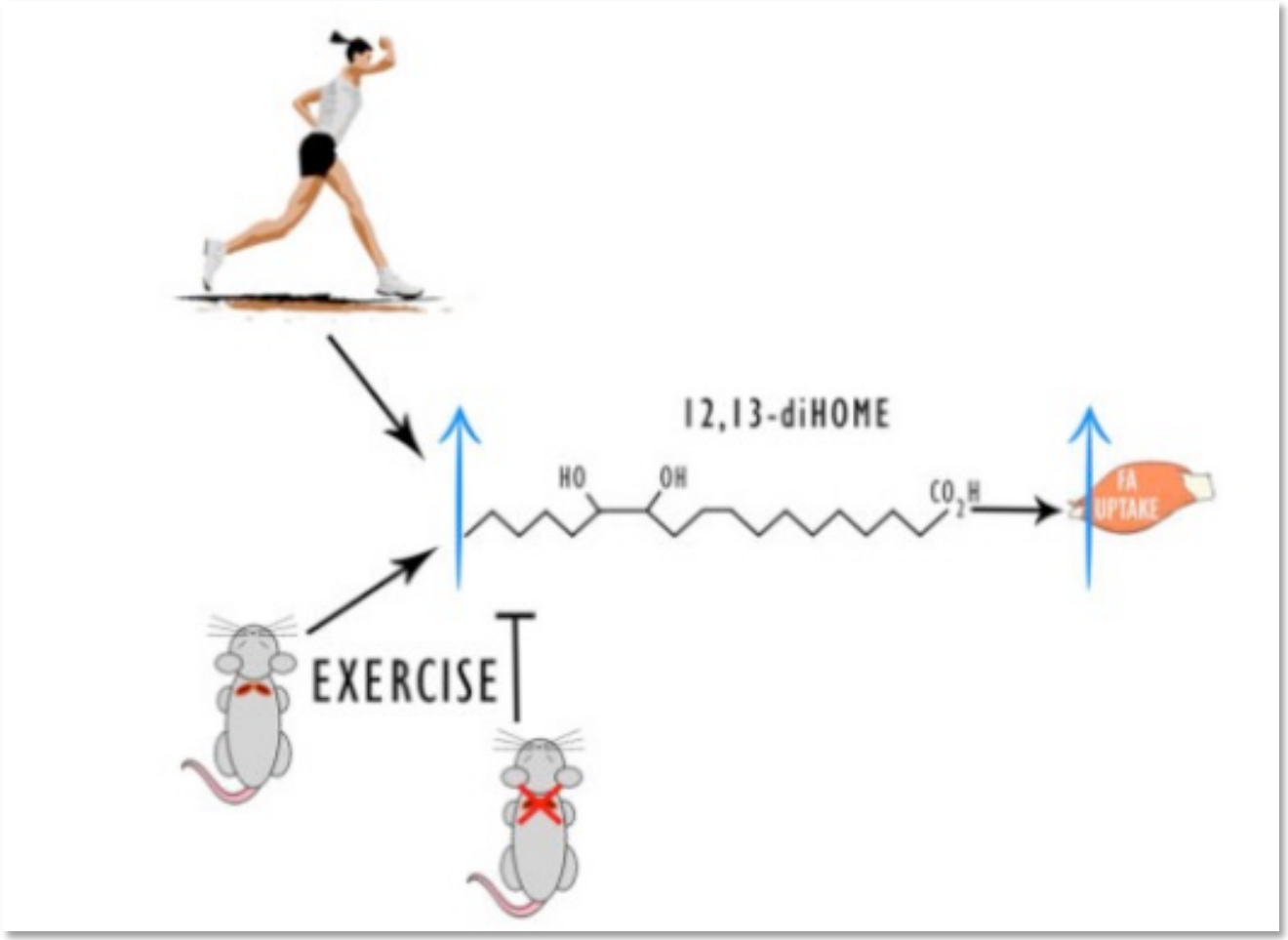
Almond intake (4 weeks) countered the 9,10-diHOME increase immediately following the 90-min eccentric exercise protocol.

9,10-DiHOME can DISRUPT mitochondrial function (Sisemore et al. Arch Biochem Biophys. 2001 Aug 1;392(1):32-7).



Almond intake (4 weeks) augmented the 12-13-diHOME increase immediately following the 90-min eccentric exercise protocol.

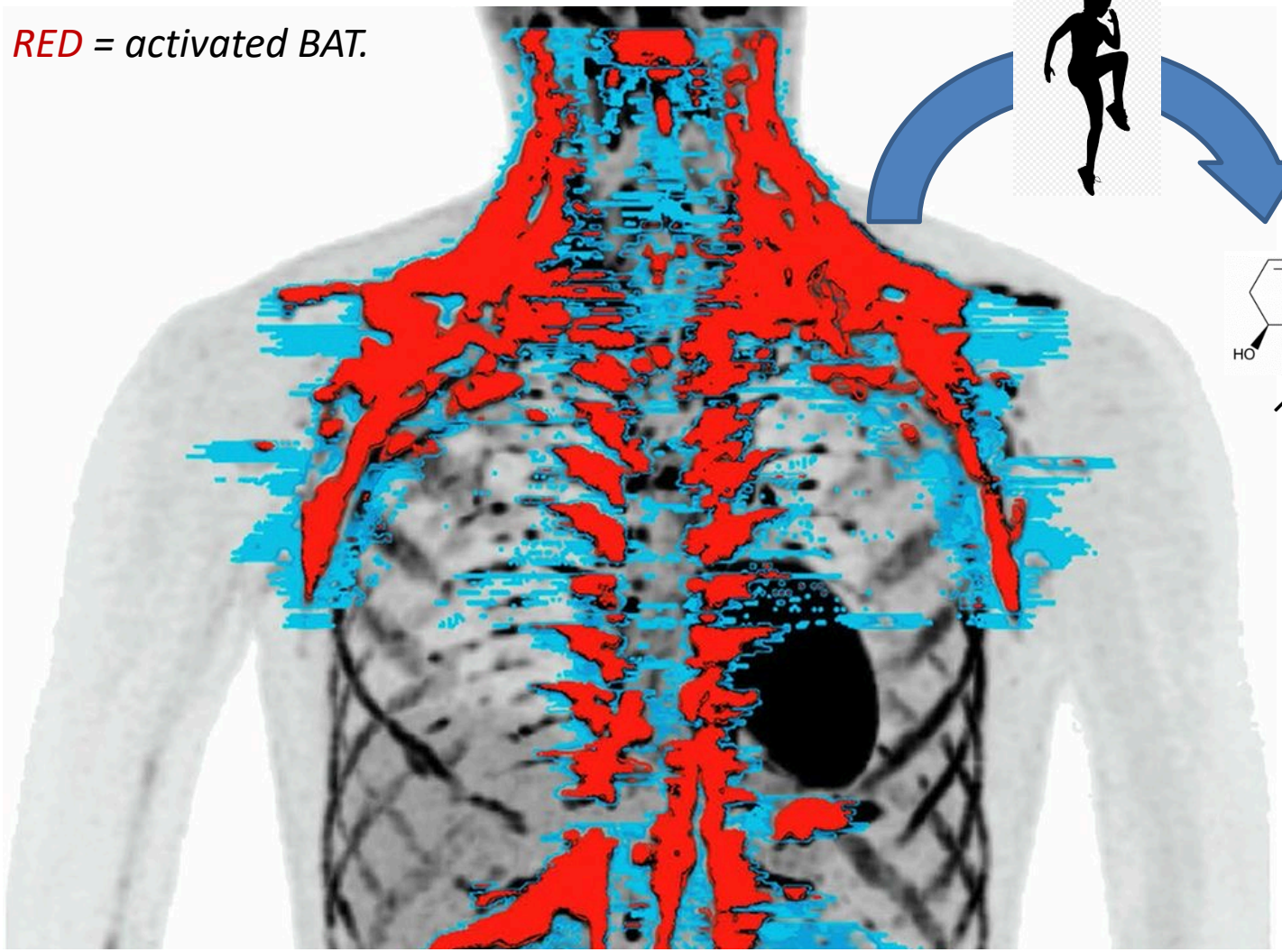


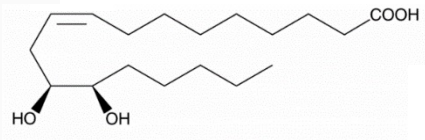


Prior research indicated that 12,13-diHOME is secreted from brown adipose tissue **(BAT)** during exercise and increases fatty acid uptake and mitochondrial respiration in skeletal muscle. This lipokine is positively correlated with aerobic fitness and negatively related to BMI and fat mass.

Source: Stanford KI, et al. 12,13-diHOME: An Exercise-Induced Lipokine that Increases Skeletal Muscle Fatty Acid Uptake. Cell Metab. 2018 Jun 5;27(6):1357.

Human brown adipose tissue (1.5% of body mass)

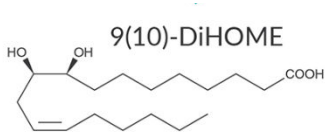




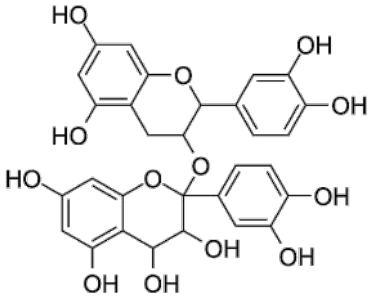
↑12,13-diHOME,
↓9,10-diHOME

Increased fatty acid uptake
and mitochondrial respiration
and improved recovery.

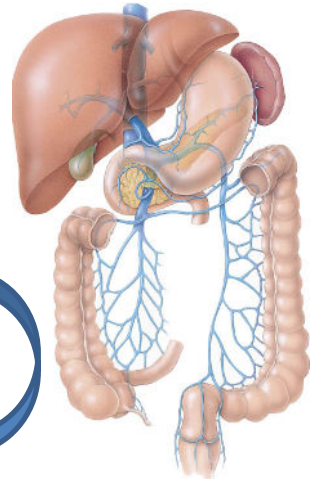
Proc Natl Acad Sci U S A. 2017 Aug 8;114(32):8649-8654.



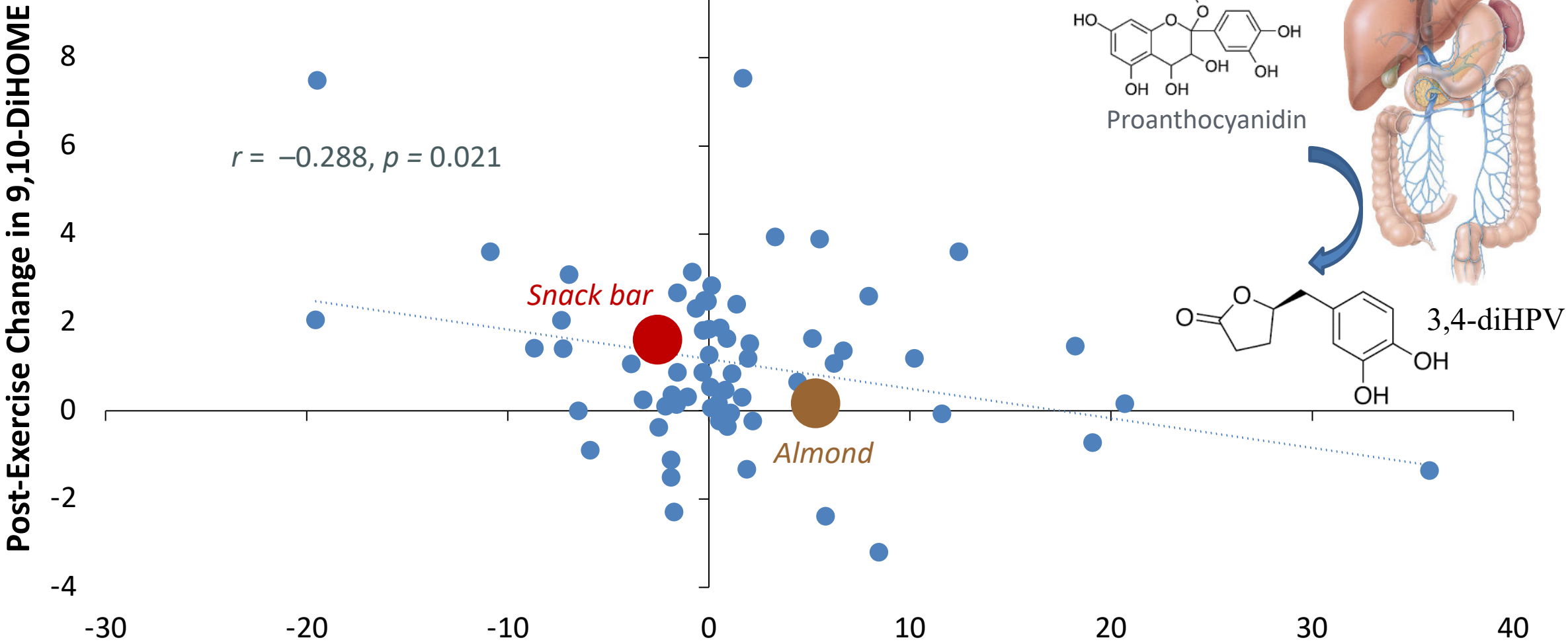
Linoleic Acid (C18:2n-6) → CYP → 9,10- and 12,13-DiHOME



Proanthocyanidin

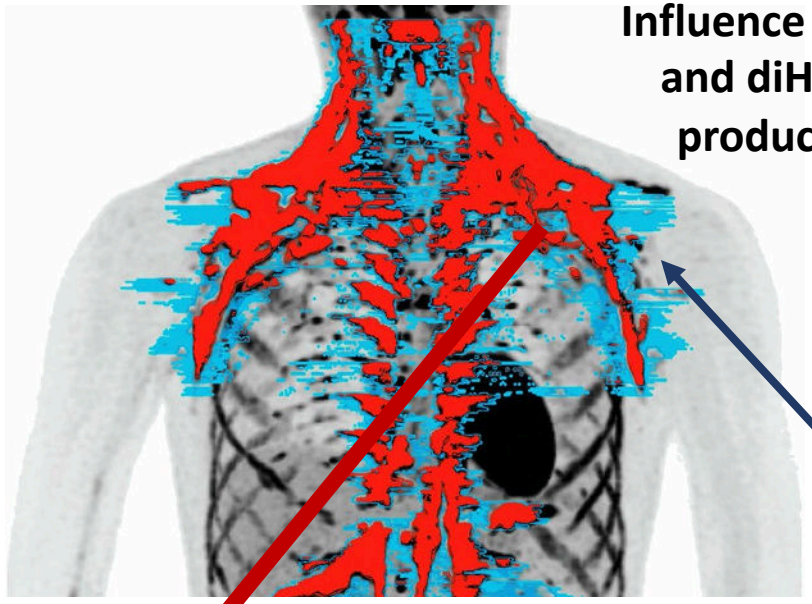


3,4-diHPV

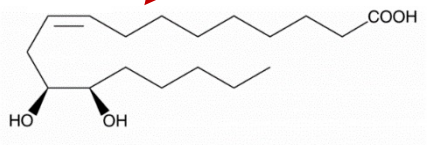


Rats treated with antibiotics to deplete gut bacteria: sEH activity was strongly altered.

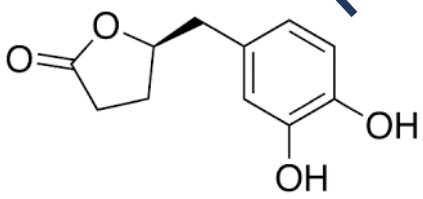
J Nutr Biochem. (2017) 49:8–14.



Influence on sEH and diHOME production

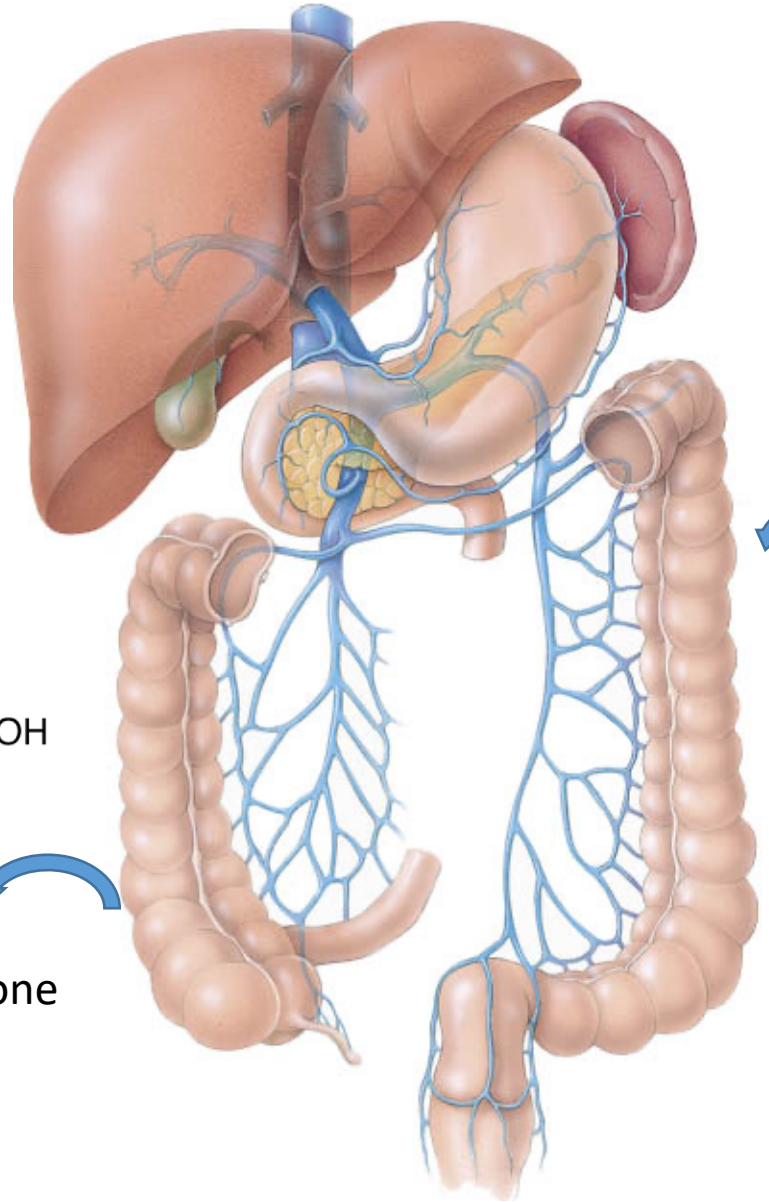


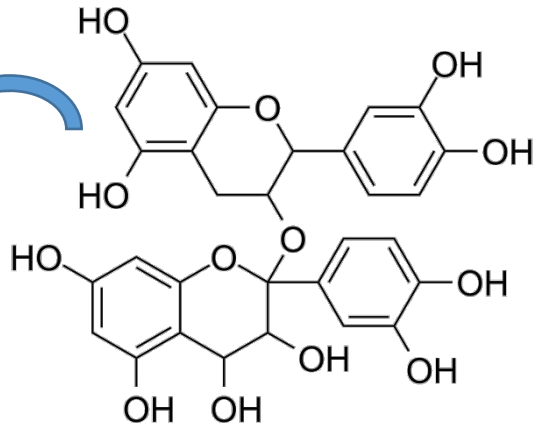
↑12,13-diHOME,
↓9,10-diHOME



3,4-diHPV

5-(3',4'-Dihydroxyphenyl)-γ-valerolactone

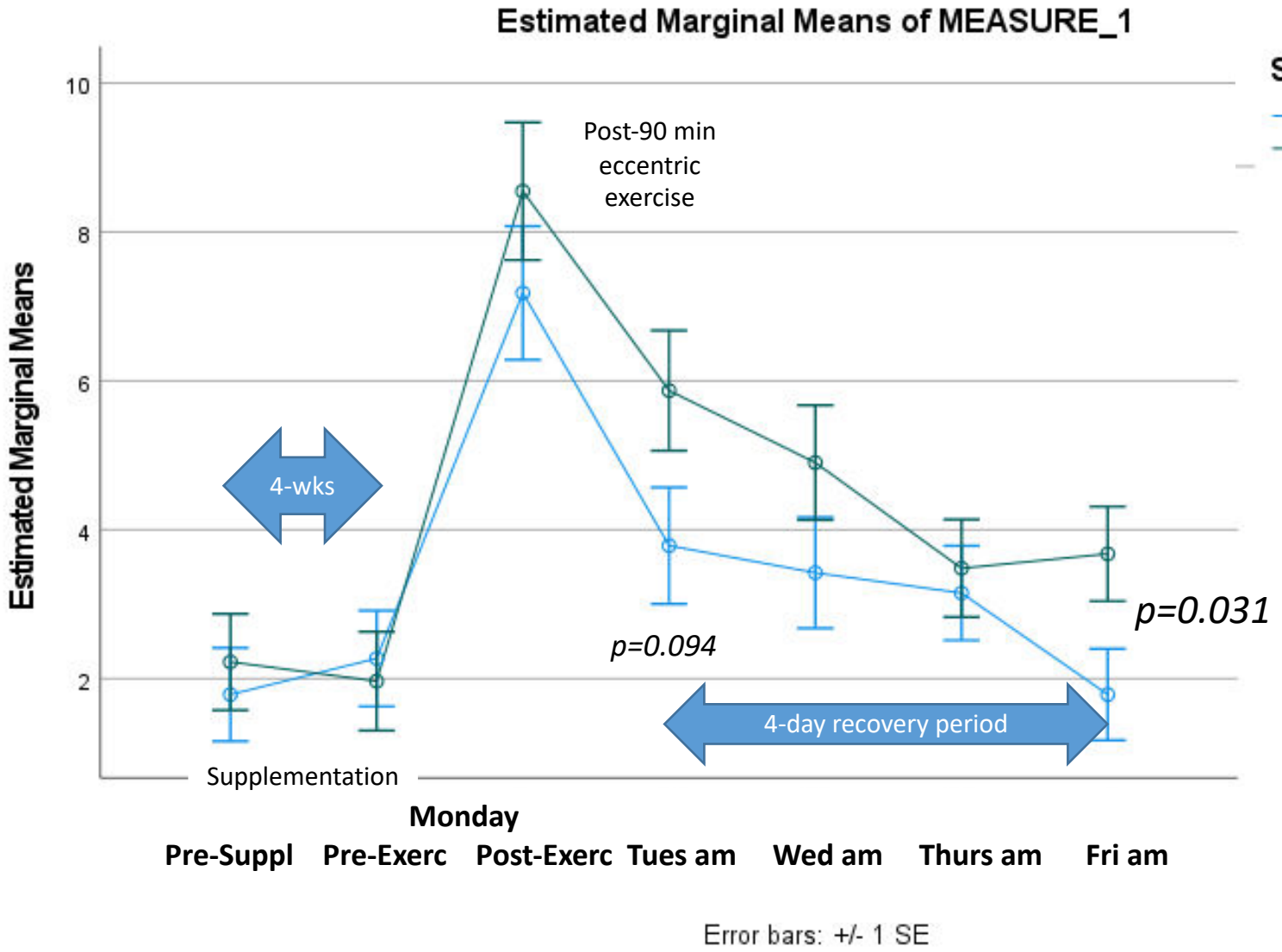




Proanthocyanidin

Profile of Mood States: Fatigue Domain

Time effect, p<0.001; Time x treatment effect, p=0.051; Interpretation: The 90-minute eccentric exercise protocol had a significant effect in increasing fatigue. The pattern of change across all time points tended to be different between groups (interaction effect, p=0.051) with lower fatigue levels in the almond group. Change contrast between groups on Friday morning, p=0.031.



Abbreviated POMS (Revised Version)

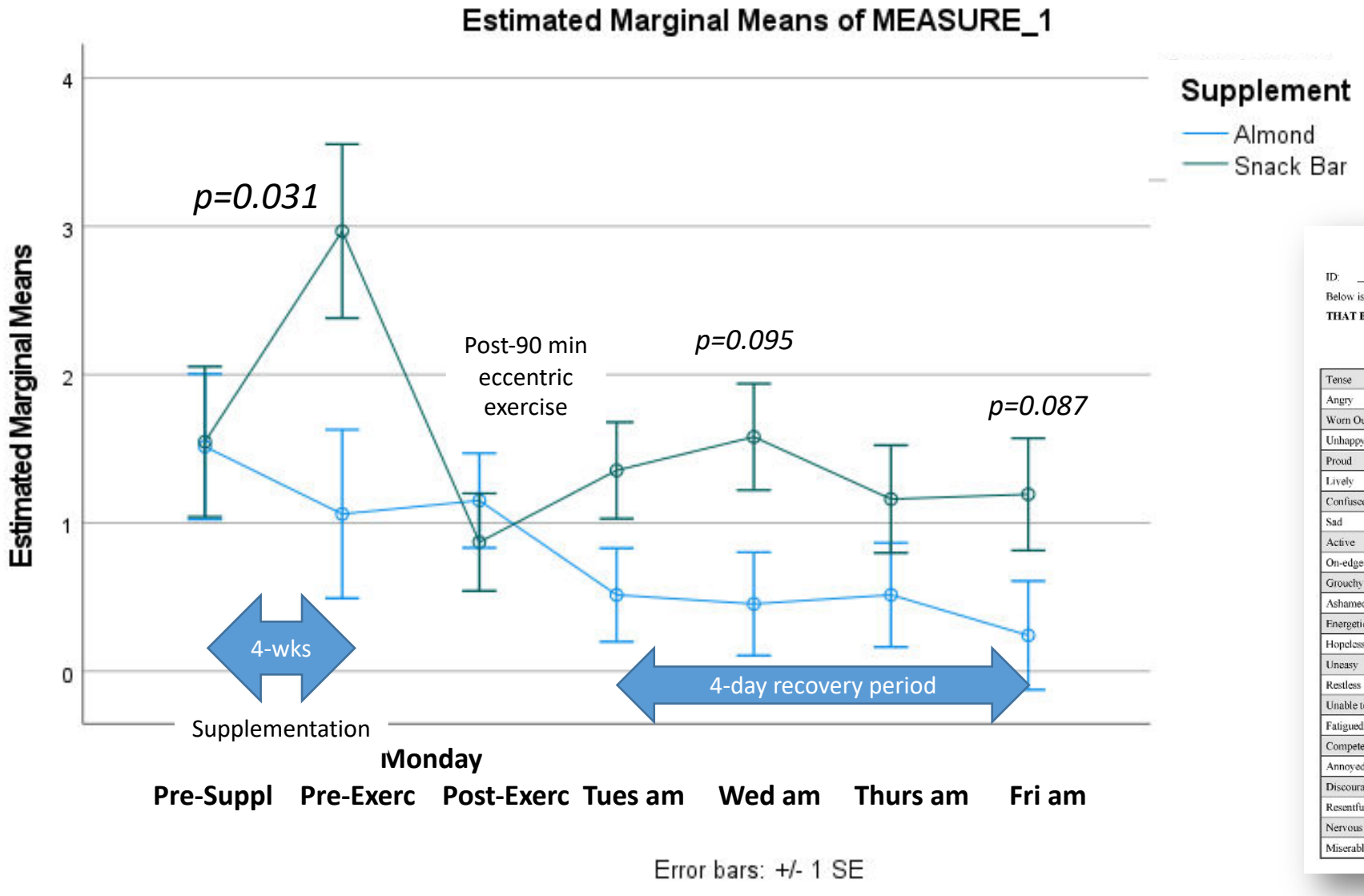
ID: _____ Date:

Below is a list of words that describe feelings people have. Please **CIRCLE THE NUMBER THAT BEST DESCRIBES HOW YOU FEEL RIGHT NOW**.

	Not At All	A Little	Moderately	Quite a lot	Extremely
Tense	0	1	2	3	4
Angry	0	1	2	3	4
Worn Out	0	1	2	3	4
Unhappy	0	1	2	3	4
Proud	0	1	2	3	4
Lively	0	1	2	3	4
Confused	0	1	2	3	4
Sad	0	1	2	3	4
Active	0	1	2	3	4
On-edge	0	1	2	3	4
Grouchy	0	1	2	3	4
Ashamed	0	1	2	3	4
Energetic	0	1	2	3	4
Hopeless	0	1	2	3	4
Uneasy	0	1	2	3	4
Restless	0	1	2	3	4
Unable to concentrate	0	1	2	3	4
Fatigued	0	1	2	3	4
Competent	0	1	2	3	4
Annoyed	0	1	2	3	4
Discouraged	0	1	2	3	4
Resentful	0	1	2	3	4
Nervous	0	1	2	3	4
Miserable	0	1	2	3	4

Profile of Mood States: Tension Domain

Time effect, p=0.006; Time x treatment effect, p=0.033; Interpretation: The pattern of change across all time points was different between groups (interaction effect, p=0.033) with lower tension levels in the almond group. P-values represent change contrast between groups.



Abbreviated POMS (Revised Version)

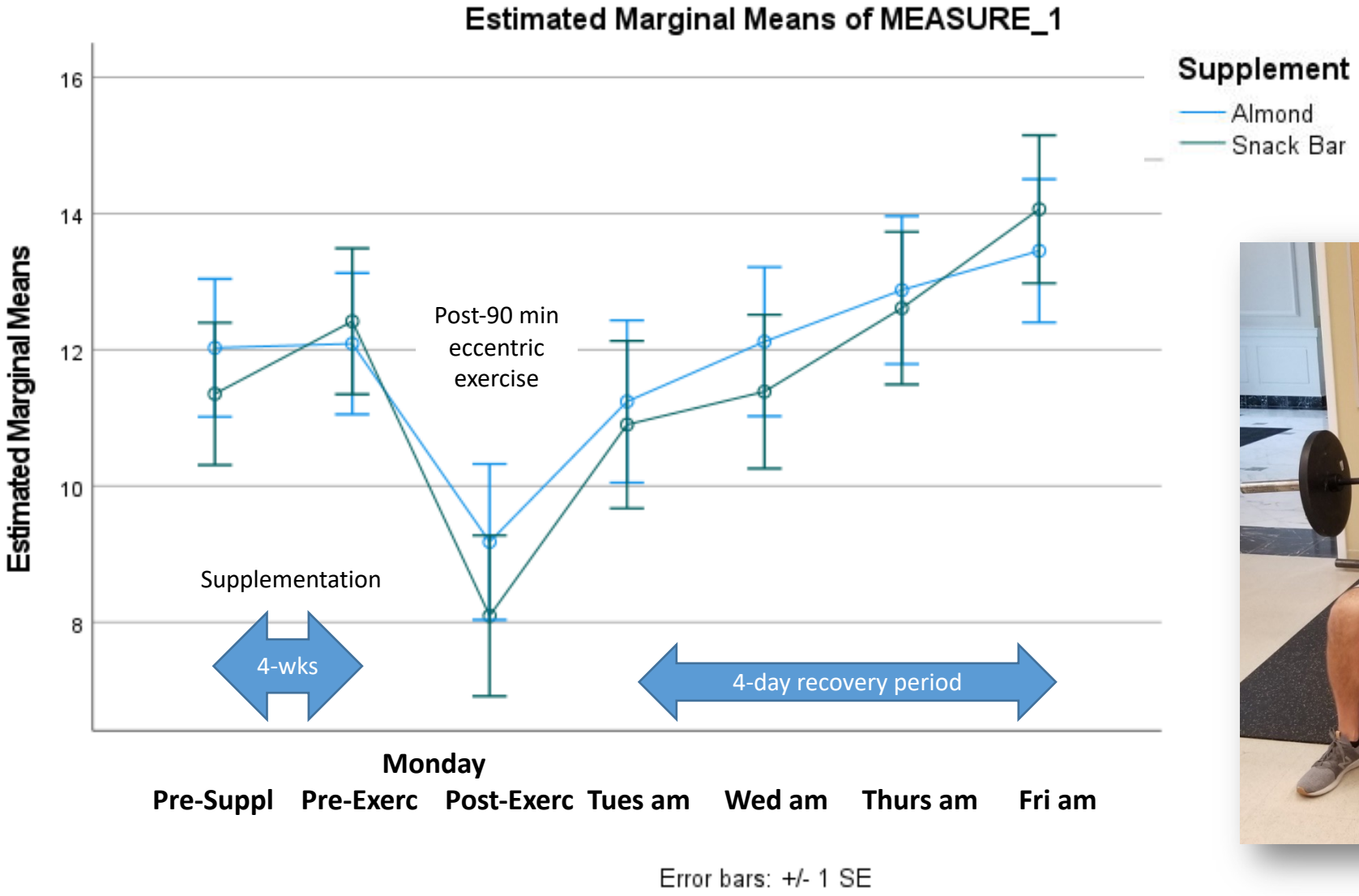
ID: _____ Date:

Below is a list of words that describe feelings people have. Please **CIRCLE THE NUMBER THAT BEST DESCRIBES HOW YOU FEEL RIGHT NOW**.

	Not At All	A Little	Moderately	Quite a lot	Extremely
Tense	0	1	2	3	4
Angry	0	1	2	3	4
Worn Out	0	1	2	3	4
Unhappy	0	1	2	3	4
Proud	0	1	2	3	4
Lively	0	1	2	3	4
Confused	0	1	2	3	4
Sad	0	1	2	3	4
Active	0	1	2	3	4
On-edge	0	1	2	3	4
Grouchy	0	1	2	3	4
Ashamed	0	1	2	3	4
Energetic	0	1	2	3	4
Hopeless	0	1	2	3	4
Uneasy	0	1	2	3	4
Restless	0	1	2	3	4
Unable to concentrate	0	1	2	3	4
Fatigued	0	1	2	3	4
Competent	0	1	2	3	4
Annoyed	0	1	2	3	4
Discouraged	0	1	2	3	4
Resentful	0	1	2	3	4
Nervous	0	1	2	3	4
Miserable	0	1	2	3	4

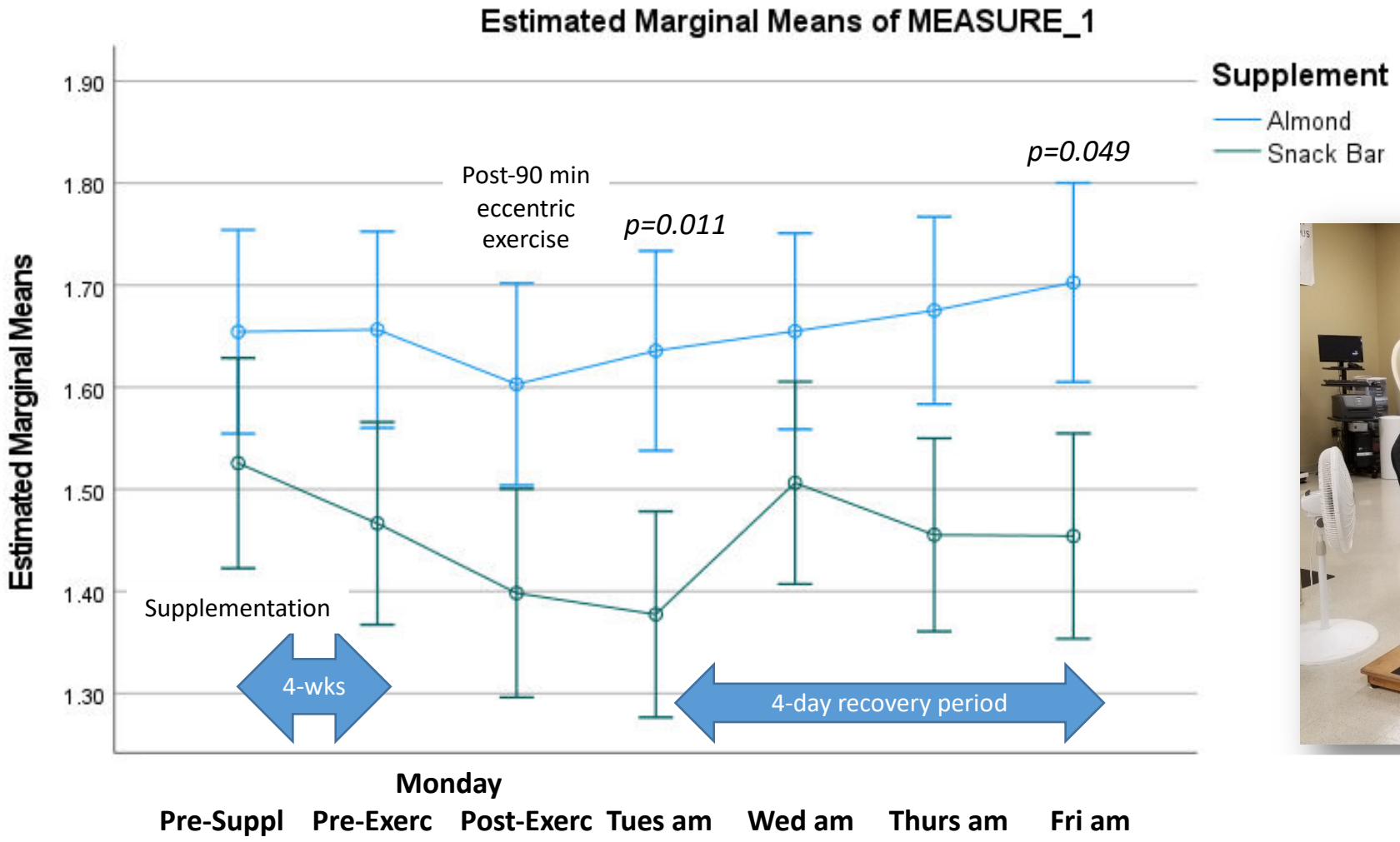
Bench Press (repetitions)

Time effect, p<0.001; Time x treatment effect, p=0.338; Interpretation: Bench press capacity (reps to fatigue) was significantly decreased after the 90-min exercise bout, but without group differences.



Leg-Back Strength (kg/kg body weight)

Time effect, p=0.004; Time x treatment effect, p=0.029; Interpretation: Leg-back strength (weight adjusted) was negatively affected in the snack bar group, but not the almond group (significant treatment effect). P-value contrasts showed group differences after 1 and 4 days recovery.



Conclusions

- Elevated post-exercise plasma levels of 12,13-DiHOME and lower 9,10-DiHOME with almond intake support positive metabolic outcomes for adults engaging in unaccustomed eccentric exercise bouts.
- Other almond related benefits for exercisers include reduced feelings of fatigue and tension, better leg-back strength during recovery, and decreased muscle damage during the first day of recovery.

Implications for Health Professionals

- Almond intake can be encouraged for physically active people to improve recovery from stressful levels of exercise.
- Supporting recovery with an accessible, whole food intervention such as almonds could increase compliance with exercise recommendations.
- Almonds can be a valuable addition to athletes' diets because of the high and varied amounts of nutrients provided, and novel findings that almond intake augmented release of 12,13-DiHOME.

Nieman et al. 2022. Frontiers in Nutrition.

Implications for Consumers

- Almonds can be added to sports nutrition strategies to help with recovery from exercise.

- Regular almond intake may improve fatigue, tension and mood state after exercise, which could help encourage future exercise sessions.

- Almonds are food for fitness offering a nutrition package of healthy fats, the antioxidant vitamin E, and polyphenols that can help explain these beneficial outcomes. One serving of almonds (28g) has 13 g of good unsaturated fat and only 1 g of saturated fat and 50% of the Daily Value of Vitamin E.

Nieman et al. 2022. Frontiers in Nutrition.

Q&A