Final Report:
Bone and Muscle Support in Ageing Women with LifeWave X49 Patch

Connor, MH[1,2]., Connor, CA.[2], Horzempa, D[2,3]. Yue, D.[4], Eickhoff, J.[5]

1 Director of Research, Akamai University, Hilo HI,
2 Earthsongs Holistic Consulting, Tucson, AZ
3 Mind-Body MD, Tucson, AZ
4 AxisPharm Laboratory, San Diego, CA
5 Senior Scientist, University of Wisconsin Madison, Madison, WI

Keywords: LifeWave, X49, Women's Health, Bone Health

ABSTRACT

Purpose:

To determine if the LifeWave X49 patch supports bone and muscle health in women ages 40 -80.

Materials:

Urine test kit, lavender top blood tubes, BD Vacutainer with Pre-attached holder, cryo tubes, racking, freezer, gloves sterile, band aids, hand sanitizer, clorox wipes, masks, UVC sterlizing wands, sterile eye droppers, sterile cotton balls, tourniquets, dry ice, shipping containers, binders, sheet protectors, computer, printer, printer cartridges, paper towels.

Method:

This proof of concept study seeks to explore the metabolic implications of wearing the Lifewave X49 patch vs the X49 in combination with X39 patch over the period of eight weeks. Measures will be taken at baseline, 24 hours, at 7 days, and 30 days and 60 days of wearing the patch. A sample of 24 subjects made up of women aged 40-80 with the goal of 20 subjects completing the study, was selected to participate in this study. Once 20 subjects completed the study recruiting and consenting was stopped. This study focused on the metabolic and physiologic impact of patch usage, with the participants using X49 at the GB34 point and X39 using the CV6 point or GV14 point as the person prefers. Acupuncture points were used to make it easier for participants to correctly place the patches. This study explores changes in AHK-Cu peptide production, changes in NTx production and to see if the X49 patch supports improved bone density.

Questionnaires:

Food diaries were maintained throughout the study by participants. Participants were asked to have a minimum of 6oz of Leucine based foods each day. Food diaries were reviewed on a weekly basis to confirm participant adherence.

Metabolic Analysis:

Sabre Sciences laboratory did the metabolic analysis (amino acid panel). Metabolic testing will consist of one 10am urine taken at baseline/day one, day two, day 7, 30 and 60. Samples will be kept in the freezer at -20F and will be shipped with ice by UPS to the Sabre Science lab in Carlsbad, CA.

AxisPharm laboratory in San Diego, CA did the blood analysis for both AHK and NTx. Two lavender top tubes will be drawn from each participant at each data point. The blood samples will go through centrifuge processing into plasma. Plasma will be seperated and placed in cryo tubes and flash frozen. Samples will be kept in the freezer at -20F and will be shipped with ice by UPS to Axis Pharm in San Diego for analysis.
Materials:

Method:

Measures were taken at baseline, 24 hours, at 7 days, and 30 days and 60 days of wearing the patch. A sample of 24 subjects made up of women aged 40-80 with the goal of 20 subjects completing the study, were selected to participate in this study. Two lavender top tubes of blood, as well as saliva and urine, were taken at each data point. Food diaries were also collected, to check for dietary impacts on changes in amino acid production.

Results:

Significant decreases from baseline were observed for AHK-Cu. The percentages of subjects with a >30% decrease in creatine levels from baseline (NTx response) to the post intervention time points. Secondary to NTx, we also saw a significant decreases in Hydroxyproline at the post intervention assessment time points. The combination of these three points strongly suggests that X49 decreases the breakdown of bones during the cycle of bone repair. In addition, we saw 14 amino acids change production levels at significance over the 60 days. The amino acids which changed were also spread between the chatecholamine, serotinergic, glutaminergic, transulferation, and histidine pathways, giving them a very broad impact.

Conclusion:

There was a significant change in both AHK-Cu and NTx. *A decrease in NTx levels has been show to correlate to a decrease in osteoclast activity, and thus a decrease in bone breakdown (Mercatali, L., et al., 2013) which means there is less bone for the osteoblastic bone formation to replace.* Secondary to NTx, we also saw a significant decrease in Hydroxyproline at 5 datapoints. "Hydroxyproline is mostly used as a diagnostic marker of bone turnover" (National Center for Biotechnology Information, 2022). The combination of these three points strongly suggests that X49 decreases the breakdown of bones during the cycle of bone repair. In addition, we saw 14 amino acids change production levels at significance over the 60 days. The amino acids which changed were also spread between the chatecholamine, seratinergic, glutaminergic, transulferation, and histidine pathways, giving them a very broad impact.

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INTRODUCTION

L-Alanine-L-Histidyl-L-Lysine Copper (AHK) has a documented impact on a variety of areas in the body, specifically skin, hair, and bone (Patt, L., 2009, Lee, W. J., et al., 2016, Jung, J. I., et al., 2018). Osteoporosis has become a "major public health problem" (Lane, N. E., 2006). As the average population age is increasing it is beginning to effect a greater proportion of the population (Lane, N. E., 2006). Unfortunately at this point in time the medications available to treat osteoporosis also have some unfortunate side effects (Mayo Clinic). These side effects are preventing some individuals from seeking treatment (Sheltawy, A. A., et al., 2015). Since AHK has a documented impact on bone it was decided to test if the X49 patches would have an effect on bone density, starting with healthy women in the age range at risk to develop osteoporosis. A dexascan was determined to be impractical due to scheduling and frequency of testing needed, so an alternative method to test for changes in bone density was chosen with serum NTx. AHK, amino acids, and hormones were also checked to develop as clear an understanding of the effects as possible.

BACKGROUND

NTx in urine has been used to index changes in bone density since at least 1994 (Garnero, P., et al., 1994). Unfortunately its utility is dependent on dilution levels, which are matched using creatinine. Fortunately NTx is also found in blood (Clemens, J. D., et al., 1997), and has been shown to reflect changes in bone density equally well (Woitge, H. W., et al., 1999).

Bones go through a cycle of repair to both microdamage and old bone "through sequential osteoclastic resorption and osteoblastic bone formation" (Eriksen E. F., 2010). NTx is released during "proteolytic cleavage of bone collagen by osteoclasts" (Clemens, J. D., et al., 1997). This means that NTx is released during the osteoclastic resorption phase of bone repair. It has specifically been found to correlate with changes in the rate of bone loss (Dresner-Pollak, R., et al., 1996). *Most importantly, a decrease in NTx levels has been show to correlate to a decrease in osteoclast activity, and thus a decrease in bone breakdown (Mercatali, L., et al., 2013) which means there is less bone for the osteoblastic bone formation to replace.*

Amino Acids are used to create proteins and peptides in the human body. Some amino acids can be produced out of smaller component pieces, but nine of them must be obtained through diet (Cleveland Clinic). Two of these amino acids which cannot be created by the body, called essential amino acids, are utilized in AHK. Because they can be used to create a broad variety of important components they also have a broad variety of functions. L-Alanine-L-Histidyl-L-Lysine Copper (AHK) is a copper polypeptide with a variety of different functions. It "increases dermal cell proliferation and viability while increasing the deposition of collagen to renew the extracellular matrix" (Patt, L., 2009). It also "promotes" the growth of human hair follicles, as is caused by stimulation of the proliferation and the preclusion of the apoptosis of dermal papilla cells" (Lee, W. J., et al., 2016). AHK linked to Vitamin C has also been shown to have "an enhancing effect on osteoblast proliferation and differentiation through activation of Smad1/5/8 and MAPK ERK1/2 and p38 signaling and without significant cytotoxicity" (Jung, J. I., et al., 2018). These functions make a fair amount of sense given the individual actions and effects of the amino acid components. Alanine has been shown to "reduce lactate concentrations during exercise and thus can improve exercise performance in endurance athletes" (Ghiasvand, R., et al., 2012). Alanine also has a stabilizing impact on glucose levels in the human body (Chiasson, J. L., et al., 1975).

Histidine is used as a "component of solutions used for organ preservation and myocardial protection in cardiac surgery" (Holeček M., 2020). It also seems to have an impact on "neurological disorders, atopic dermatitis, metabolic syndrome, diabetes, uraemic anaemia, ulcers, inflammatory bowel diseases, malignancies, and muscle performance during strenuous exercise" (Holeček M., 2020), though unfortunately more research is needed in those areas to clarify the impact. Lysine is "crucial for collagen fibre crosslinking" (Pastel, E., et al., 2018). All of these areas are likely to have an impact on bone development.

PURPOSE

To determine if the LifeWave X49 patch supports bone and muscle health in women ages 40 -81.

MATERIALS

Urine test kit, lavender top blood tubes, BD Vacutainer with Pre-attached holder, cryo tubes, racking, freezer, gloves sterile, band aids, hand sanitizer, clorox wipes, masks, UVC sterilizing wands, sterile eye droppers, sterile cotton balls, tourniquets, dry ice, shipping containers, binders, sheet protectors, computer, printer, printer cartridges, paper towels.

METHOD

This proof of concept study seeks to explore the metabolic implications of wearing the LifeWave X49 patch vs. the X49 in combination with X39 patch over the period of eight weeks. Measures were taken at baseline, 24 hours, at 7 days, and 30 days and 60 days of wearing the patch. A random sample of 24 subjects made up of women aged 40-80 with the goal of 20 subjects completing the study, were selected to participate in this study. Once 20 subjects completed the study stopped recruiting and consenting. This study focused on the metabolic and physiologic impact of patch usage, with the X49 participants using the GB34 point and X39 using the CV6 point or GV14 point as the person prefers. This study explores changes in AHK peptide production, changes in NTx production and improved bone density.

Questionnaires:

Food diaries were maintained throughout the study by participants. Participants will be asked to have a minimum of 6oz of Leucine based foods each day. Food diaries will be reviewed on a weekly basis to confirm participant adherence.

Metabolic Analysis One:

AxisPharm laboratory in San Diego, CA will do blood analysis for both AHK and NTx. Two lavender top tubes will be drawn from each participant at each data point. The blood samples will go through centrifuge processing into plasma. Plasma will be seperated and placed in cryo tubes and flash frozen. Samples will be kept in the freezer at -20F and will be shipped with ice by UPS to Axis Pharm in San Diego for analysis.

Metabolic Analysis Two:

Sabre Sciences laboratory will do metabolic analysis (amino acid panel). Metabolic testing will consist of one 10am urine taken at baseline/day one, day two, day 7, 30 and 60. Samples will be kept in the freezer at -20F and will be shipped with ice by UPS to the Sabre Science lab in Carlesbad, CA.

Statistical Analysis:

NTx - Creatinine levels and changes in creatinine levels were not normally distributed and therefore summarized in terms of medians and ranges. Changes from baseline (day 1) to day 2, day 7, day 14, day 30 and day 60 were analyzed using a nonparametric Wilcoxon Signed Rank test. Creatine responses were defined as a decrease of at least 30% from baseline. The frequencies and percentages of creatine responses were summarized in tabular format. Creatine response rates were reported along with the corresponding two-sided 95% confidence intervals (CI) which were constructed using the Wilson score method. AHK outcomes were normally distributed and summarized in terms of means and standard deviations and changes from baseline were evaluated using a paired t-test. Amino Acid outcome parameters were summarized in terms of means, standard deviations, and medians. Changes between consecutive time points (day 1 vs. day 2, day 2 vs. day 7, day 7 vs. day 14 etc.) were evaluate using a nonparametric Wilcoxon signed rank test (Table 2). Absolute changes from baseline (day 1) to day 2, day 7, day 14, day 30 and day 60 were also summarized in terms of means, standard deviations, medians, and interquartile ranges. All reported P-values are two-sided and $P<0.05$ was used to define statistical significance. Statistical analyses were conducted using R software, version 4.1.0.

RESULTS

NTx:

Table 1: *NTx: Creatinine levels at each assessment time point*

Day	N	Median	Range
1 (baseline)	18	2.45	0.03-24.88
2	18	4.78	0.03-12.74
7	18	2.58	0.03-22.34

14	18	2.73	0.03-36.77
30	18	2.45	0.00-34.90
60	17	2.70	0.32-75.44

Table 2: NTX response (>30% decrease in creatine levels from baseline)

Day	N	**Number of Responses**	**Response Rate (95% CI)**
2	18	5	28% (12-51%)
7	18	8	44% (25-66%)
14	18	7	39% (20-61%)
30	18	8	44% (25-66%)
60	17	7	41% (22-64%)

Because this study was only 60 days and the bone development cycle is effected by 150 days we did not see significance across all parameters. *We did see in all data points (2-6) the necessary 30% decrease in NTx-creatine levels showing that* osteoclastic resorption *was taking place.* This test should be repeated in a way that covers all the cycles involved and specific analysis on a subject by subject basis should be done to confirm what % of participants see calcium re-absorption into the bone and changes in the osteoclastic resorption phase.

AHK-Cu:

Table 3: Changes in AHK-Cu concentration from baseline to day 30.

AHK Outcome	**Day**	**Mean**	**SD**	**p-value**
AHK-Cu Peak Area	30	-4626	9252	0.0489
Amount of AHK-CU injected (ng)	30	-0.19	0.38	0.0488
Total AHK-Cu in Sample (ng)	30	-18.97	37.95	0.0489
AHK-Cu Concentration in Sample (ng/ml)	30	-12.65	25.30	0.0489

A significant decrease in AHK-Cu concentration (-12.65 ng/ml, p=0.0489) was observed from baseline to Day 30, indicating that AHK-Cu was being used to build bone, muscle, tendons, skin, and hair.

Table 4: AHK: AHK outcomes at each assessment time point

Outcome Parameter	Day	N	Mean	SD
AHK-Cu Peak Area	1 (baseline)	18	11708	8104
	2	18	11669	13313
	7	18	9731	5507
	14	18	10564	6526
	30	18	7082	4441
	60	17	7737	5417
Total AHK-Cu in Sample (ng)	1 (baseline)	18	48.02	33.24
	2	18	47.86	54.61
	7	18	39.91	22.59
	14	18	43.33	26.77
	30	18	29.05	18.22
	60	17	31.74	22.22
AHK-Cu Concentration in Sample (ng/ml)	1 (baseline)	18	32.01	22.16
	2	18	31.91	36.4
	7	18	26.61	15.06
	14	18	35.02	36.59
	30	18	19.36	12.14
	60	17	26.85	33.17

Specific Amino Acids of Significance:

Table 1: Absolute decreases from baseline (Day 1)

Parameter	Day	N	Mean	SD	Median	Lower 25ht Percentile	Upper 75th Percentile	p-value[1]
DA	7	17	-14.9	27.9	-11.4	-23.0	-1.6	0.0395
Hist	30	16	-2.1	5.4	-3.9	-6.0	0.5	0.0739
LDOPA	2	17	-4.4	8.0	-1.2	-10.3	2.8	0.0963
LDOPA	30	16	-4.7	8.7	-2.8	-10.5	1.0	0.0676
Cystathionine	2	17	-1.8	2.8	-1.8	-3.6	-0.7	0.0224
HYP	7	17	-2.9	2.3	-2.9	-4.8	-1.4	0.0003
HYP	14	17	-2.1	3.0	-2.5	-4.6	0.5	0.0202
HYP	30	16	-2.2	2.8	-2.6	-4.0	0.1	0.0092
HYP	60	14	-2.5	3.0	-1.8	-5.4	-0.3	0.0061
Sar	60	14	-2.0	3.5	-1.4	-3.4	0.7	0.0500
Leu	60	14	-1.8	3.2	-2.0	-5.1	0.7	0.0676

His	2	17	-29.1	75.8	-7.7	-23.8	1.0	0.0569
Ser	60	14	-10.8	20.5	-5.9	-13.4	-0.2	0.0756
Ala	2	17	-4.2	33.3	-0.7	-6.3	1.3	0.7467
Ala	7	17	-2.2	27.2	2.3	-10.9	8.5	1.0000
Ala	14	16	-1.3	29.1	3.2	-2.5	6.1	0.3484
Cys	60	14	-7.4	12.3	-3.6	-7.8	0.0	0.0068

1:p-value for evaluating changes from baseline (day 1)

Table 2: Absolute increases from baseline (Day 1)

Param	Day	N	Mean	SD	Median	Lower 25ht Percentile	Upper 75th Percentile	p-value1
GSH	7	17	0.2	0.5	0.3	0.0	0.5	0.0627
Gln	7	17	36.9	48.9	22.8	6.6	59.6	0.0040
Gly	2	17	39.9	101.0	13.6	0.4	24.5	0.0038
Asn	14	16	21.3	38.8	11.4	-4.1	42.5	0.0654
Ala	30	15	0.8	27.0	1.2	-4.8	15.9	0.4543
Ala	60	14	3.4	21.2	2.1	-5.4	9.5	0.6257

Table 3: *Summary statistics of outcome parameters, stratified by time point*

Parameter	**Day**	**N**	**Mean**	**SD**	**Median**	**p-value[1]**	**p-value[2]**	**p-value[3]**	**p-value[4]**	**p-value[5]**
5-HT	1	17	109.7	24.7	110.0	0.6441	0.3061	0.0695	0.0250	0.9999
5-HT	2	17	107.0	25.2	102.4					
5-HT	7	17	101.8	24.1	101.7					
5-HT	14	17	115.9	24.4	121.3					
5-HT	30	16	101.8	28.6	103.3					
5-HT	60	14	103.0	26.8	105.8					
NE	1	17	37.6	9.3	37.3	0.4307	0.0448	0.1740	0.5966	0.5416
NE	2	17	35.4	8.5	36.2					
NE	7	17	39.7	9.0	38.0					
NE	14	17	35.8	8.5	35.9					
NE	30	16	34.9	8.6	36.6					
NE	60	14	32.8	8.9	32.5					
ME	1	17	27.3	6.9	27.9	0.4874	0.3348	0.0459	0.0021	0.6698
ME	2	17	26.3	6.7	26.5					
ME	7	17	25.2	8.0	22.1					
ME	14	17	29.1	6.0	29.4					
ME	30	16	25.1	7.3	25.1					

ME	60	14	24.5	8.1	25.3					
HCys2	1	17	1.4	0.6	1.2	0.1328	0.9522	0.5896	0.6278	0.0308
HCys2	2	17	1.6	0.6	1.3					
HCys2	7	17	1.5	0.7	1.3					
HCys2	14	17	1.3	0.5	1.3					
HCys2	30	16	1.3	0.5	1.1					
HCys2	60	14	1.1	0.4	1.1					
Hcys/HCys2	1	17	0.9	0.3	0.9	0.1626	0.5871	0.4304	0.5336	0.0513
Hcys/HCys2	2	17	0.8	0.3	0.7					
Hcys/HCys2	7	17	0.8	0.2	0.8					
Hcys/HCys2	14	17	0.9	0.4	0.8					
Hcys/HCys2	30	16	0.9	0.3	0.9					
Hcys/HCys2	60	14	1.0	0.4	1.1					
Cystathionine	1	17	14.9	6.8	13.9	0.0224	0.4874	0.4800	0.6412	0.8198
Cystathionine	2	17	13.1	6.6	11.4					
Cystathionine	7	17	14.9	7.2	13.9					
Cystathionine	14	17	13.3	6.7	13.9					
Cystathionine	30	16	12.4	6.8	12.6					
Cystathionine	60	14	14.7	8.4	14.4					
AAA	1	17	13.4	9.0	11.4	0.2744	0.2293	0.4657	0.0258	0.4917
AAA	2	17	15.2	12.1	12.1					
AAA	7	17	14.3	12.4	9.6					
AAA	14	17	14.2	11.1	7.9					
AAA	30	16	12.3	10.7	6.6					
AAA	60	14	10.9	10.6	5.6					
ABA	1	17	7.5	3.1	7.1	0.9357	0.4515	0.0910	0.4716	0.4854
ABA	2	17	7.4	2.2	6.9					
ABA	7	17	7.1	2.6	6.3					
ABA	14	17	8.2	2.9	6.9					
ABA	30	16	7.8	3.2	6.5					
ABA	60	14	7.3	2.6	7.6					
HYP	1	17	14.0	2.8	15.3	0.1231	0.0348	0.2633	0.9800	0.7728
HYP	2	17	12.9	3.2	13.7					
HYP	7	17	11.2	2.8	11.3					
HYP	14	17	12.0	3.1	11.6					
HYP	30	16	11.7	3.8	12.0					
HYP	60	14	11.4	3.8	10.5					
GSH	1	17	1.4	0.5	1.3	0.4262	0.0857	0.9222	0.2217	0.5085
GSH	2	17	1.4	0.4	1.3					
GSH	7	17	1.6	0.5	1.5					
GSH	14	17	1.6	0.5	1.8					
GSH	30	16	1.4	0.4	1.4					

GSH	60	14	1.4	0.5	1.3					
Lys	1	17	26.2	23.5	16.4	0.2633	0.0032	0.1973	0.9341	0.9460
Lys	2	17	20.2	12.7	15.9					
Lys	7	17	30.0	23.3	15.2					
Lys	14	16	25.4	30.5	15.2					
Lys	30	15	18.5	8.9	16.0					
Lys	60	14	16.8	12.3	12.8					
Gln	1	17	169.2	70.9	193.7	0.6356	0.0004	0.1754	0.5336	0.2163
Gln	2	17	178.8	87.0	197.0					
Gln	7	17	206.0	82.8	231.0					
Gln	14	16	188.4	89.2	194.5					
Gln	30	15	190.3	96.2	194.7					
Gln	60	14	165.0	96.7	154.4					
His	1	17	102.8	88.6	69.8	0.0569	0.0013	0.9100	0.1354	0.4548
His	2	17	73.7	53.2	58.9					
His	7	17	93.8	65.7	73.8					
His	14	16	93.7	68.4	75.9					
His	30	15	125.3	108.2	80.2					
His	60	14	87.9	84.8	57.1					
Trp	1	17	14.8	14.2	13.6	0.6526	0.2078	0.1826	0.0859	0.6909
Trp	2	17	13.7	13.0	16.1					
Trp	7	17	17.5	17.9	19.2					
Trp	14	16	11.3	9.6	11.7					
Trp	30	15	15.0	16.4	12.5					
Trp	60	14	10.1	10.5	8.5					

p-value[1]: p-value for comparing Day 1 vs. Day 2
p-value[2]: p-value for comparing Day 2 vs. Day 7
p-value[3]: p-value for comparing Day 7 vs. Day 14
p-value[4]: p-value for comparing Day 14 vs. Day 30
p-value[5]: p-value for comparing Day 30 vs. Day 60

Food Diaries:

Food diary summaries showed that most vegetarians and vegans involved in the study failed to eat sufficient foods which contained lycine.

DISCUSSION

This data demonstrates that the LifeWave X49 patch triggers the use of more AHK-Cu. AHK-Cu is involved in the creation of bone, tendon, skin, hair etc. We can see that at data point 5 (30 days). X49 does effect calcium re-absorbtion in a cycle 30-40 days as shown by the NTX and Hydroxyproline data. (*The decrease in osteoclast activity means that less of the bone is being broken down, (Mercatali, L., et al., 2013) which means there is less bone for the osteoblastic bone formation to replace.*) The cycle involved means that X49 does not make the changes in a regularly trackable form for bone formation in less than 150 days. That is why only part of the participants showed an increase in second step bone absorption and not every participant showed an increase.

NTx, while not statistically significant across all participants at all data points, is still an exciting finding. It is also supported by the significant decrease in Hydroxyproline at 5 datapoints. Most medications for bone support have significant side effects. We had no side effects reported. In addition, we had two individuals in the study who had standard bone density scans during the study. Both individuals showed a slowing of bone loss and an increase in bone density in areas that had been previously compromised. This is a wellness product and not a medical product, however, consideration should be given to this patch which could potentially be used to mitigate early bone loss.

There were some surprises in the data. X49 triggered support of the dopamine pathway. We got significance on day 2 and day 30. X49 triggered support for 5HT in the catecholamine pathway supporting serotonin production. Women in the study got less depressed and probably slept better. This is a very interesting finding as many women at the pari-menopausal and menopausal stage suffer from chronic imbalances between serotonin and dopamine and can have fairly significant mood swings. It is also logical that the trans-sulferation pathway is stimulated and X49 triggers more glutathione production. In addition, we saw 14 amino acids change production levels at significance over the 60 days. In general we would expect to see an ocassional spike in one or two amino acids but like the X39 product we again saw significance in a much larger than expected number. The amino acids which changed were also spread between the chatecholamine, seratinergic, glutaminergic, transulferation, and histidine pathways, giving them a very broad impact. In addition to the amino acid changes we saw a significant increase in 2-aminoadipic acid, which is produced by lysine degradation. This lends support to the theory that the amino acids were being used.

What we expected and did not see was an individual increase of Alanine as a direct amino acid measured by urine but the amino acid data did show Leucine went to almost significance at .06 on day 60 and it is a pre-cursor of the Alanine. That may be because we were not checking blood and AHK-Cu was being used and was not available free in the urine. It is also possible we did not check with sufficient frequency or for a long enough period given the 150 day cycle involved. We did see that AHK-Cu dropped significantly at 30 days and we would only see that effect if the tri-peptide was being used more effectively. We did see significance on the Histodine on day 2 at .05 and almost significance on day 30 at .07. So we got much of the expected pathway information. We simply did not find a direct amino acid increase in Alanine link except by the measure of the tri-peptide.

It should be noted that this was an open label study. It might be a good idea to repeat this study as both a double-blind and with a longer duration, so a complete bone re-absorbtion and formation cycle can be monitored. It might also be a good idea to include lycine supplements in follow-up studies, as it was found that multiple participants were not consuming enough in their diet.

CONCLUSION:

There was a significant change in both AHK-Cu and NTx. *A decrease in NTx levels has been show to correlate to a decrease in osteoclast activity, and thus a decrease in bone breakdown (Mercatali, L., et al., 2013) which means there is less bone for the osteoblastic bone formation to replace.* Secondary to NTx, we also saw a significant decrease in Hydroxyproline at 5 datapoints. "Hydroxyproline is mostly used as a diagnostic marker of bone turnover" (National Center for Biotechnology Information, 2022). The combination of these three points strongly suggests that X49 decreases the breakdown of bones during the cycle of bone repair. In addition, we saw 14 amino acids change production levels at significance over the 60 days. The amino acids which changed were also spread between the chatecholamine, seratinergic, glutaminergic, transulferation, and histidine pathways, giving them a very broad impact.

REFERENCES

Chiasson, J. L., Liljenquist, J. E., Sinclair-Smith, B. C., & Lacy, W. W. (1975). Gluconeogenesis from alanine in normal postabsorptive man. Intrahepatic stimulatory effect of glucagon. *Diabetes*, *24*(6), 574–584. https://doi.org/10.2337/diab.24.6.574

Clemens, J. D., Herrick, M. V., Singer, F. R., & Eyre, D. R. (1997). Evidence that serum NTx (collagen-type I N-telopeptides) can act as an immunochemical marker of bone resorption. *Clinical chemistry*, *43*(11), 2058–2063.

Cleveland Clinic Staff. (12/22/2021) Amino Acids. Pulled 6/15/2022. https://my.clevelandclinic.org/health/articles/22243-amino-acids

Dresner-Pollak, R., Parker, R. A., Poku, M., Thompson, J., Seibel, M. J., & Greenspan, S. L. (1996). Biochemical markers of bone turnover reflect femoral bone loss in elderly women. *Calcified tissue international*, *59*(5), 328–333. https://doi.org/10.1007/s002239900135

Eriksen E. F. (2010). Cellular mechanisms of bone remodeling. *Reviews in endocrine & metabolic disorders*, *11*(4), 219–227. https://doi.org/10.1007/s11154-010-9153-1

Garnero, P., Shih, W. J., Gineyts, E., Karpf, D. B., & Delmas, P. D. (1994). Comparison of new biochemical markers of bone turnover in late postmenopausal osteoporotic women in response to alendronate treatment. *The Journal of clinical endocrinology and metabolism*, *79*(6), 1693–1700. https://doi.org/10.1210/jcem.79.6.7989477

Ghiasvand, R., Askari, G., Malekzadeh, J., Hajishafiee, M., Daneshvar, P., Akbari, F., & Bahreynian, M. (2012). Effects of Six Weeks of β-alanine Administration on VO(2) max, Time to Exhaustion and Lactate Concentrations in Physical Education Students. *International journal of preventive medicine*, *3*(8), 559–563.

Holeček M. (2020). Histidine in Health and Disease: Metabolism, Physiological Importance, and Use as a Supplement. *Nutrients*, *12*(3), 848. https://doi.org/10.3390/nu12030848

Lane N. E. (2006). Epidemiology, etiology, and diagnosis of osteoporosis. *American journal of obstetrics and gynecology*, *194*(2 Suppl), S3–S11. https://doi.org/10.1016/j.ajog.2005.08.047

Lee, W. J., Sim, H. B., Jang, Y. H., Lee, S. J., Kim, d., & Yim, S. H. (2016). Efficacy of a Complex of 5-Aminolevulinic Acid and Glycyl-Histidyl-Lysine Peptide on Hair Growth. *Annals of dermatology*, *28*(4), 438–443. https://doi.org/10.5021/ad.2016.28.4.438

Mayo Clinic Staff. (updated: 2/1/2022) Drugs and Supplements Ibandronate (Oral Route) Side Effects. Pulled 6/14/2022. https://www.mayoclinic.org/drugs-supplements/ibandronate-oral-route/side-effects/drg-20068079

Mercatali, L., Ricci, M., Scarpi, E., Serra, P., Fabbri, F., Ricci, R., Liverani, C., Zanoni, M., Zoli, W., Maltoni, R., Gunelli, E., Amadori, D., & Ibrahim, T. (2013). RANK/RANK-L/OPG in Patients with Bone Metastases Treated with Anticancer Agents and Zoledronic Acid: A Prospective Study. International journal of molecular sciences. 14. 10683-10693. 10.3390/ijms140610683.

National Center for Biotechnology Information (2022). PubChem Compound Summary for CID 5810, Hydroxyproline. Retrieved June 16, 2022 from https://pubchem.ncbi.nlm.nih.gov/compound/Hydroxyproline.

Pastel, E., Price, E., Sjöholm, K., McCulloch, L. J., Rittig, N., Liversedge, N., Knight, B., Møller, N., Svensson, P. A., & Kos, K. (2018). Lysyl oxidase and adipose tissue dysfunction. *Metabolism: clinical and experimental*, *78*, 118–127. https://doi.org/10.1016/j.metabol.2017.10.002

Patt, L. (2009) Neova® DNA Repair Factor Nourishing Lotion Stimulates Collagen and Speeds Natural Repair Process. Procyte, A PhotoMedex Company.

Sheltawy, A. A., Criseno, S., Gittoes, N. J., & Crowley, R. K. (2015). Fear of medication side effects is a barrier to optimal osteoporosis care. *Osteoporosis international : a journal established as result of cooperation between the European Foundation for Osteoporosis and the National Osteoporosis Foundation of the USA*, *26*(2), 843–844. https://doi.org/10.1007/s00198-014-2922-z

Woitge, H. W., Pecherstorfer, M., Li, Y., Keck, A. V., Horn, E., Ziegler, R., & Seibel, M. J. (1999). Novel serum markers of bone resorption: clinical assessment and comparison with established urinary indices. *Journal of bone and mineral research : the official journal of the American Society for Bone and Mineral Research*, *14*(5), 792–801. https://doi.org/10.1359/jbmr.1999.14.5.792

APPENDIX ONE : Complete Statistical Analysis of NTX and AHK

November 16, 2021

Statistical Methods: Creatinine levels and changes in creatinine levels were not normally distributed and therefore summarized in terms of means and ranges. Changes from baseline were analyzed using a nonparametric Wilcoxon Signed Rank test. Creatine responses were defined as a decrease of at least 30% from baseline. The number of frequencies of responses were summarized in tabular format. Creatine response rates were reported along with the corresponding two-sided 95% confidence intervals which were constructed using the Wilson score method. AHK outcomes were summarized in terms of means and standard deviations and changes from baseline were evaluated using a pared t-test. All reported p-values are two-sided and $p<0.05$ was used to define statistical significance.

N=18 study participants with median age of 65 years (range 48-77 years).

Table 1: *NTx: Creatinine levels at each assessment time point*

	N	Median	Range
R1	18	2.45	0.03-24.88
R2	18	4.78	0.03-12.74
R3	18	2.58	0.03-22.34
R4	18	2.73	0.03-36.77
R5	18	2.45	0.00-34.90
R6	17	2.70	0.32-75.44

Table 2: NTX: Change in Creatinine levels from baseline

		Change in creatinine from baseline		
	N	**Median**	**Range**	**p-value**
R2	18	0.68	-20.55-8.40	0.8536
R3	18	-0.69	-13.16-10.48	0.3692
R4	18	-0.03	-23.04-19.67	0.9999
R5	18	-0.05	-23.40-23.55	0.9999
R6	17	0.37	-23.54-72.73	0.5477

There were no significant changes in creatinine levels observed.

Table 3: NTX response (>30% decrease in creatine levels from baseline)

	N	**Number of Responses**	**Response Rate (95% CI)**
R2	18	5	28% (12-51%)
R3	18	8	44% (25-66%)
R4	18	7	39% (20-61%)
R5	18	8	44% (25-66%)
R6	17	7	41% (22-64%)

The NTX response rates range from 28% at R2 to 44% at R3 and R4. There is no significant trend in response rates from R2-R6 (p=0.46).

Table 4: *AHK: AHK outcomes at each assessment time point*

Outcome Parameter	**Time**	**N**	**Mean**	**SD**
AHK-Cu Peak Area	R1	18	11708	8104
	R2	18	11669	13313
	R3	18	9731	5507
	R4	18	10564	6526
	R5	18	7082	4441
	R6	17	7737	5417
Amount of AHK-CU injected (ng)	R1	18	0.48	0.33
	R2	18	0.48	0.55
	R3	18	0.4	0.23
	R4	18	0.43	0.27
	R5	18	0.29	0.18
	R6	17	0.32	0.22
Total AHK-Cu in Sample (ng)	R1	18	48.02	33.24
	R2	18	47.86	54.61
	R3	18	39.91	22.59
	R4	18	43.33	26.77
	R5	18	29.05	18.22
	R6	17	31.74	22.22
AHK-Cu Concentration in Sample (ng/ml)	R1	18	32.01	22.16
	R2	18	31.91	36.4
	R3	18	26.61	15.06
	R4	18	35.02	36.59
	R5	18	19.36	12.14
	R6	17	26.85	33.17

Figure 2: *AHK-Cu Concentration in sample over time*

Table 4: *AHK Change in AHK outcomes from baseline*

AHK Outcome	**Time**	**Mean**	**SD**	**p-value**
AHK-Cu Peak Area	R2	-39	15862	0.9919
	R3	-1977	6478	0.2128
	R4	-1144	7795	0.5418
	R5	-4626	9252	0.0489
	R6	-4018	9540	0.1017

Amount of AHK-CU injected (ng)	R2	0.00	0.65	0.9917
	R3	-0.08	0.27	0.2120
	R4	-0.05	0.32	0.5413
	R5	-0.19	0.38	0.0488
	R6	-0.16	0.39	0.1014
Total AHK-Cu in Sample (ng)	R2	-0.16	65.06	0.9919
	R3	-8.11	26.57	0.2127
	R4	-4.69	31.97	0.5418
	R5	-18.97	37.95	0.0489
	R6	-16.48	39.13	0.1017
AHK-Cu Concentration in Sample (ng/ml)	R2	-0.11	43.37	0.9918
	R3	-5.41	17.71	0.2127
	R4	3.01	35.37	0.7226
	R5	-12.65	25.30	0.0489
	R6	-5.29	40.85	0.6006

A significant decrease in AHK-Cu concentration (-12.65 ng/ml, p=0.0489) was observed from baseline to R5.

APPENDIX TWO: Complete Statistical Analysis of Amino Acids

June 10, 2022

Statistical Analysis: Outcome parameters were summarized in terms of means, standard deviations, and medians. Changes between consecutive time points (day 1 vs. day 2, day 2 vs. day 7, day 7 vs. day 14 etc.) were evaluate using a nonparametric Wilcoxon signed rank test (Table 2). Absolute changes from baseline (day 1) to day 2, day 7, day 14, day 30 and day 60 were summarized in terms of means, standard deviations, medians, lower 25th percentiles and 75th percentiles, and evaluated using a nonparametric Wilcoxon signed rank test. All reported P-values are two-sided and P<0.05 was used to define statistical significance. Statistical analyses were conducted using R software, version 4.1.0.

Table 1: Absolute changes from baseline (Day 1)

Parameter	**Day**	**N**	**Mean**	**SD**	**Median**	**Lower 25ht Percentile**	**Upper 75th Percentile**	**p-value[1]**
Cre	2	17	-6.7	61.0	6.6	-22.4	23.2	0.7819
Cre	7	17	-0.2	79.4	14.3	-52.2	50.7	0.8536
Cre	14	17	-1.4	75.3	9.0	-48.7	28.9	0.6533
Cre	30	16	16.2	78.6	6.6	-32.3	40.8	0.4954
Cre	60	14	32.0	91.5	11.5	-29.7	118.0	0.2958
5-HT	2	17	-2.7	24.5	-2.2	-8.4	6.3	0.6441
5-HT	7	17	-7.9	22.0	-3.1	-14.3	4.2	0.2633
5-HT	14	17	6.2	28.4	10.9	-6.7	21.8	0.3778
5-HT	30	16	-7.2	34.8	-4.2	-18.0	11.4	0.5619
5-HT	60	14	-8.3	30.2	-5.8	-20.8	21.6	0.4631
DA	2	17	-9.9	29.0	-12.3	-25.7	10.6	0.1454
DA	7	17	-14.9	27.9	-11.4	-23.0	-1.6	0.0395
DA	14	17	-2.1	41.2	-9.6	-19.2	0.0	0.2114
DA	30	16	-7.9	36.6	-7.9	-37.6	8.5	0.2689
DA	60	14	-11.6	29.9	-7.4	-18.5	-1.0	0.1531
5-HT/DA	2	17	0.0	0.2	0.0	0.0	0.1	0.3231
5-HT/DA	7	17	0.0	0.2	0.0	-0.1	0.1	0.7556
5-HT/DA	14	17	0.1	0.2	0.1	-0.1	0.2	0.1485
5-HT/DA	30	16	0.0	0.3	0.0	-0.1	0.1	0.8898
5-HT/DA	60	14	0.0	0.2	0.0	0.0	0.1	0.8188
NE	2	17	-2.2	9.0	-1.0	-7.7	3.5	0.4307
NE	7	17	2.1	7.5	1.7	-3.5	9.6	0.4038
NE	14	17	-1.8	7.8	-1.4	-5.5	1.3	0.2386
NE	30	16	-2.2	10.6	-2.0	-6.9	3.9	0.4637

NE	60	14	-3.0	10.3	-2.1	-8.9	4.2	0.3258
E	2	17	-0.3	1.3	-0.4	-1.3	0.4	0.3230
E	7	17	0.3	1.8	0.3	-1.3	1.8	0.5966
E	14	17	-0.1	2.1	0.0	-1.5	1.1	0.6975
E	30	16	-0.4	1.9	-0.1	-1.7	1.2	0.6233
E	60	14	0.5	1.7	0.6	-0.8	1.3	0.4357
NE/E	2	17	-0.1	1.4	0.2	-0.8	1.0	0.9632
NE/E	7	17	0.0	1.4	0.1	-0.4	0.8	0.6777
NE/E	14	17	-0.2	1.6	-0.1	-0.8	0.9	0.8111
NE/E	30	16	0.0	1.2	0.3	-0.5	0.9	0.5037
NE/E	60	14	-0.8	1.7	-0.3	-2.3	0.3	0.1353
GABA	2	17	-0.2	0.7	-0.1	-0.7	0.4	0.4111
GABA	7	17	-0.2	1.5	-0.3	-0.7	0.5	0.3712
GABA	14	17	-0.3	0.9	-0.4	-0.8	0.2	0.2576
GABA	30	16	-0.3	1.0	-0.4	-1.2	0.3	0.1715
GABA	60	14	-0.2	1.1	0.1	-1.1	0.7	0.5085
Glu	2	17	0.2	4.2	0.5	-2.3	2.5	0.8626
Glu	7	17	-0.1	4.5	-0.6	-1.5	1.5	0.7554
Glu	14	17	0.4	4.1	1.0	-1.7	3.8	0.6197
Glu	30	16	-0.4	3.5	-0.7	-1.5	2.0	0.8999
Glu	60	14	0.0	4.3	0.6	-2.5	1.7	0.8425
Hist	2	17	-1.7	4.2	-1.3	-2.8	0.8	0.1059
Hist	7	17	-0.2	6.2	-1.1	-3.5	4.0	0.9724
Hist	14	17	0.4	5.1	0.6	-3.7	2.5	0.8267
Hist	30	16	-2.1	5.4	-3.9	-6.0	0.5	0.0739
Hist	60	14	-1.4	6.3	-1.2	-5.1	2.7	0.4631
TA	2	17	-1.7	4.9	-1.6	-3.0	1.5	0.1901
TA	7	17	-1.8	7.8	-2.2	-5.9	0.3	0.2069
TA	14	17	-0.9	5.5	-0.8	-2.1	1.8	0.6685
TA	30	16	-1.5	7.7	-2.1	-4.0	4.2	0.4637
TA	60	14	-1.4	8.4	-0.7	-3.5	3.3	0.8552
LDOPA	2	17	-4.4	8.0	-1.2	-10.3	2.8	0.0963
LDOPA	7	17	-3.0	9.8	-1.5	-8.8	1.2	0.1743
LDOPA	14	17	-3.7	9.5	-4.4	-7.8	1.9	0.1454
LDOPA	30	16	-4.7	8.7	-2.8	-10.5	1.0	0.0676
LDOPA	60	14	2.3	8.6	2.5	-6.4	7.2	0.3998
ME	2	17	-0.9	8.3	-1.1	-4.7	4.5	0.4874
ME	7	17	-2.1	10.5	-2.4	-5.9	8.4	0.5791
ME	14	17	1.8	8.7	1.7	-3.7	8.6	0.4038
ME	30	16	-2.1	8.8	-2.0	-6.4	3.3	0.3824
ME	60	14	-2.7	8.7	0.4	-7.3	3.1	0.4729
NorM	2	17	-4.9	32.7	-3.6	-22.2	16.4	0.6441

NorM	7	17	-0.6	55.0	8.8	-35.1	31.6	0.9265
NorM	14	17	10.8	48.4	19.9	-22.2	39.9	0.2435
NorM	30	16	-3.3	58.5	-12.6	-38.0	26.4	0.6322
NorM	60	14	7.0	54.7	10.2	-21.1	45.1	0.6257
3-MT	2	17	-0.5	9.4	-0.9	-5.2	6.4	1.0000
3-MT	7	17	-3.1	19.5	-1.7	-12.5	6.5	0.5791
3-MT	14	17	-1.8	14.2	0.1	-14.1	8.4	0.7467
3-MT	30	16	-5.0	13.0	-7.8	-15.8	5.2	0.1628
3-MT	60	14	-1.6	20.0	-2.3	-14.4	9.9	0.6698
Hcys	2	17	0.1	0.2	0.1	0.0	0.2	0.2749
Hcys	7	17	0.1	0.4	0.0	-0.2	0.3	0.3997
Hcys	14	17	0.0	0.3	0.1	-0.2	0.2	0.6405
Hcys	30	16	0.0	0.3	-0.1	-0.3	0.3	0.8396
Hcys	60	14	0.0	0.3	-0.1	-0.2	0.2	0.9204
HCys2	2	17	0.2	0.5	0.1	-0.1	0.4	0.1328
HCys2	7	17	0.1	0.6	0.1	-0.3	0.5	0.5701
HCys2	14	17	0.0	0.6	0.0	-0.4	0.4	0.9895
HCys2	30	16	-0.1	0.8	0.0	-0.5	0.4	0.7529
HCys2	60	14	-0.3	0.8	-0.2	-0.4	0.2	0.3497
Hcys/HCys2	2	17	-0.1	0.3	-0.1	-0.2	0.1	0.1626
Hcys/HCys2	7	17	-0.1	0.3	-0.1	-0.2	0.2	0.3694
Hcys/HCys2	14	17	0.0	0.5	0.1	-0.3	0.4	0.8085
Hcys/HCys2	30	16	0.0	0.4	0.0	-0.3	0.3	0.7157
Hcys/HCys2	60	14	0.2	0.5	0.2	-0.2	0.4	0.3027
Cystathionine	2	17	-1.8	2.8	-1.8	-3.6	-0.7	0.0224
Cystathionine	7	17	0.0	7.4	-1.6	-3.4	0.9	0.3003
Cystathionine	14	17	-1.6	5.7	-0.8	-2.0	1.1	0.4037
Cystathionine	30	16	-2.1	6.1	-1.1	-2.4	1.3	0.3484
Cystathionine	60	14	0.0	9.0	-1.7	-4.3	0.0	0.2439
AAA	2	17	1.8	4.3	0.2	-0.9	4.0	0.2744
AAA	7	17	0.9	5.7	-0.7	-3.2	2.1	0.9724
AAA	14	17	0.8	3.8	0.0	-1.9	2.8	0.5619
AAA	30	16	-0.6	3.0	-0.8	-2.7	1.5	0.3550
AAA	60	14	-0.6	3.7	-0.5	-3.0	1.3	0.4631
ABA	2	17	-0.1	2.6	0.2	-0.6	0.8	0.9357
ABA	7	17	-0.4	3.7	0.0	-2.0	1.4	0.6473
ABA	14	17	0.7	2.8	1.3	-0.1	2.0	0.2633
ABA	30	16	0.1	3.7	0.5	-3.1	2.8	0.8999
ABA	60	14	-0.8	3.4	-0.2	-4.0	1.2	0.4725
BAIBA	2	17	-2.1	12.0	-3.4	-7.5	6.4	0.7032
BAIBA	7	17	-2.7	21.7	-2.1	-7.9	6.7	0.5870
BAIBA	14	17	-8.0	22.8	-4.1	-9.7	2.4	0.2842

BAIBA	30	16	3.3	30.7	-0.1	-7.8	8.4	0.9799
BAIBA	60	14	9.9	62.6	-0.1	-14.9	17.4	1.0000
HYP	2	17	-1.1	3.1	-1.1	-3.5	1.0	0.1231
HYP	7	17	-2.9	2.3	-2.9	-4.8	-1.4	0.0003
HYP	14	17	-2.1	3.0	-2.5	-4.6	0.5	0.0202
HYP	30	16	-2.2	2.8	-2.6	-4.0	0.1	0.0092
HYP	60	14	-2.5	3.0	-1.8	-5.4	-0.3	0.0061
Hlys	2	17	0.1	1.9	-0.6	-1.0	1.1	0.9171
Hlys	7	17	-0.5	1.9	-0.3	-1.6	0.4	0.3972
Hlys	14	17	-0.4	2.0	-0.5	-1.1	0.4	0.4104
Hlys	30	16	-0.1	1.9	0.6	-1.4	1.3	1.0000
Hlys	60	14	0.1	2.1	0.3	-1.0	0.9	0.8552
PEA	2	17	0.1	0.3	0.1	-0.2	0.3	0.3221
PEA	7	17	0.1	0.7	0.1	-0.4	0.5	0.8993
PEA	14	17	0.1	0.5	-0.1	-0.2	0.4	0.7717
PEA	30	16	0.0	0.7	0.1	-0.6	0.6	0.7921
PEA	60	14	0.1	0.7	0.2	-0.6	0.5	0.9883
Hser	2	17	0.2	1.6	0.2	-0.1	0.8	0.2107
Hser	7	17	-0.1	2.4	-0.5	-0.9	0.7	0.7029
Hser	14	17	-0.1	2.1	0.0	-1.0	0.7	0.7436
Hser	30	16	-0.2	1.5	-0.1	-0.8	0.3	0.6284
Hser	60	14	-0.2	1.7	-0.1	-0.7	0.9	0.8926
GSH	2	17	0.0	0.4	-0.1	-0.2	0.1	0.4261
GSH	7	17	0.2	0.5	0.3	0.0	0.5	0.0627
GSH	14	17	0.2	0.7	0.3	-0.2	0.8	0.1035
GSH	30	16	0.0	0.6	0.1	-0.2	0.4	0.7327
GSH	60	14	0.0	0.6	-0.1	-0.3	0.3	0.9883
Sar	2	17	-1.7	4.1	-0.6	-3.1	0.5	0.1114
Sar	7	17	-0.8	3.8	0.2	-0.7	1.0	0.7727
Sar	14	17	-1.1	3.8	-0.1	-1.1	0.8	0.4557
Sar	30	16	-1.0	3.2	-0.7	-1.4	0.7	0.3412
Sar	60	14	-2.0	3.5	-1.4	-3.4	0.7	0.0500
Arg	2	17	-0.3	3.6	0.6	-0.8	1.3	0.6527
Arg	7	17	0.4	5.4	0.8	-3.3	2.3	0.9632
Arg	14	16	0.1	2.1	-0.2	-0.9	1.5	0.7300
Arg	30	15	1.8	5.3	1.0	-1.9	4.9	0.2769
Arg	60	14	0.4	4.1	-0.1	-2.3	3.5	0.9392
Pro	2	17	-0.4	14.4	-0.3	-2.7	2.4	0.9632
Pro	7	17	-1.1	11.1	0.9	-3.1	2.3	1.0000
Pro	14	16	0.7	9.8	-0.2	-2.0	4.4	0.8707
Pro	30	15	-2.3	10.9	0.1	-4.9	1.7	0.3894
Pro	60	14	-2.3	10.8	-1.4	-5.9	1.9	0.4631

Lys	2	17	-6.0	14.6	-0.8	-8.3	2.1	0.2633
Lys	7	17	3.8	13.5	2.8	-0.2	9.2	0.1901
Lys	14	16	-1.5	35.6	-0.6	-4.2	2.5	0.6229
Lys	30	15	-5.9	19.6	0.3	-4.9	6.0	0.9780
Lys	60	14	-4.1	15.3	-2.1	-5.5	3.5	0.4169
Leu	2	17	0.1	4.8	-0.9	-2.1	1.3	0.5711
Leu	7	17	-0.1	3.6	-0.4	-2.6	2.3	0.7208
Leu	14	16	0.3	4.1	0.1	-2.2	3.9	0.7917
Leu	30	15	-0.8	3.3	-1.3	-3.8	0.1	0.2078
Leu	60	14	-1.8	3.2	-2.0	-5.1	0.7	0.0676
Ile	2	17	-12.7	56.4	-0.3	-22.0	1.8	0.6355
Ile	7	17	-2.4	61.1	1.7	-0.7	3.0	0.4377
Ile	14	16	-10.3	53.7	1.6	-0.8	5.7	0.2078
Ile	30	15	-17.3	67.2	0.4	-4.0	2.2	0.6788
Ile	60	14	-13.0	71.9	1.2	-1.3	6.7	0.3910
Gln	2	17	9.7	48.0	6.4	-28.1	33.3	0.6356
Gln	7	17	36.9	48.9	22.8	6.6	59.6	0.0040
Gln	14	16	21.0	53.6	1.1	-17.2	58.1	0.2744
Gln	30	15	28.8	55.0	1.8	-15.4	64.4	0.1688
Gln	60	14	4.0	57.9	-5.3	-42.3	37.6	1.0000
His	2	17	-29.1	75.8	-7.7	-23.8	1.0	0.0569
His	7	17	-9.0	72.4	1.2	-4.4	6.1	0.5791
His	14	16	-10.8	78.8	-2.0	-13.8	31.6	0.7436
His	30	15	19.8	39.8	8.3	-4.9	43.7	0.1170
His	60	14	-14.3	106.1	-5.1	-15.3	20.7	0.8077
Trp	2	17	-1.1	4.2	0.1	-1.0	0.9	0.6526
Trp	7	17	2.7	9.2	0.7	-0.3	9.1	0.1487
Trp	14	16	-3.1	10.8	-0.2	-3.6	1.7	0.5036
Trp	30	15	0.2	8.2	0.3	-3.2	3.3	0.7148
Trp	60	14	-3.1	10.4	-0.2	-8.1	1.5	0.5516
Tryp	2	17	1.1	5.1	0.0	-0.4	0.4	0.9899
Tryp	7	17	-2.9	15.9	-0.5	-3.4	-0.1	0.1061
Tryp	14	16	1.5	14.9	-0.2	-0.6	0.4	0.6091
Tryp	30	15	1.5	22.4	-0.1	-1.0	0.6	0.8440
Tryp	60	14	0.9	22.1	-0.2	-1.1	0.5	0.5936
5HTP	2	17	2.2	16.5	1.6	-1.0	10.3	0.3060
5HTP	7	17	0.9	16.9	-2.9	-7.7	14.5	1.0000
5HTP	14	16	5.8	24.3	5.6	-11.4	13.0	0.5282
5HTP	30	15	4.1	26.8	4.3	-8.5	15.0	0.5708
5HTP	60	14	3.9	20.3	1.3	-16.0	20.3	0.5830
Tyr	2	17	-21.3	151.3	-11.6	-27.0	12.0	0.5477
Tyr	7	17	8.8	176.6	6.6	-17.8	31.7	0.5171

Tyr	14	16	-14.0	155.7	-1.2	-26.0	25.1	0.8603
Tyr	30	15	-29.0	142.0	-2.5	-19.2	36.5	0.8904
Tyr	60	14	-10.0	164.8	1.0	-48.6	17.8	0.9515
Gly	2	17	39.9	101.0	13.6	0.4	24.5	0.0038
Gly	7	17	-3.6	74.4	-0.2	-18.9	18.5	0.7819
Gly	14	16	-7.5	32.3	0.1	-11.5	7.1	0.6685
Gly	30	15	-0.1	44.8	0.1	-3.0	29.5	0.9014
Gly	60	14	11.4	55.2	1.2	-1.9	55.8	0.4263
Bala	2	17	0.9	9.1	-0.5	-4.5	0.5	0.5703
Bala	7	17	2.2	12.3	1.4	-0.6	3.2	0.4038
Bala	14	16	-3.0	7.6	0.5	-3.7	1.6	0.4567
Bala	30	15	-2.6	8.3	0.3	-10.7	3.1	0.7197
Bala	60	14	2.8	8.2	2.0	-3.9	10.8	0.3054
Phe	2	17	-4.5	30.7	-2.3	-22.5	3.6	0.4332
Phe	7	17	-10.1	34.6	-11.7	-33.2	17.5	0.3289
Phe	14	16	-5.6	22.5	-2.7	-23.0	7.9	0.3484
Phe	30	15	-18.1	36.7	-14.9	-32.2	6.9	0.1070
Phe	60	14	-9.4	28.4	-2.7	-15.3	8.5	0.4631
Asn	2	17	12.8	50.9	2.0	-8.8	8.9	0.9265
Asn	7	17	1.4	29.4	0.0	-9.4	15.6	0.8999
Asn	14	16	21.3	38.8	11.4	-4.1	42.5	0.0654
Asn	30	15	11.1	54.2	0.8	-24.4	24.9	0.8469
Asn	60	14	8.0	33.9	2.8	-7.2	16.2	0.5830
Thr	2	17	-4.8	16.9	-1.0	-15.0	1.3	0.3842
Thr	7	17	2.9	27.1	0.6	-8.4	5.0	0.6441
Thr	14	16	-3.7	17.1	-0.5	-22.6	3.4	0.7820
Thr	30	15	-5.0	15.7	-1.8	-14.8	1.1	0.2026
Thr	60	14	-5.8	19.3	-0.1	-14.9	1.8	0.4725
Met	2	17	0.0	1.1	0.1	-0.7	0.6	0.9894
Met	7	17	0.5	1.9	0.4	-0.4	0.7	0.3972
Met	14	16	0.4	1.9	0.6	-0.1	1.4	0.1164
Met	30	15	0.3	1.3	0.5	-0.4	1.0	0.3657
Met	60	14	0.4	1.7	0.3	-0.1	1.2	0.3105
Asp	2	17	-2.3	10.3	0.1	-0.1	0.5	0.8902
Asp	7	17	-1.3	14.7	0.4	0.0	2.1	0.2832
Asp	14	16	-0.5	5.3	0.2	-0.3	0.6	0.5309
Asp	30	15	-4.3	14.2	0.0	-0.5	0.9	0.7233
Asp	60	14	-6.3	14.9	0.1	-2.0	0.5	0.6226
Ser	2	17	-2.7	16.5	2.4	-7.9	4.5	1.0000
Ser	7	17	0.8	27.6	1.4	-12.6	4.9	0.9632
Ser	14	16	2.9	38.3	-2.3	-13.7	4.3	0.4954
Ser	30	15	-8.7	23.6	-2.6	-22.1	3.6	0.3970

Ser	60	14	-10.8	20.5	-5.9	-13.4	-0.2	0.0756
Ala	2	17	-4.2	33.3	-0.7	-6.3	1.3	0.7467
Ala	7	17	-2.2	27.2	2.3	-10.9	8.5	1.0000
Ala	14	16	-1.3	29.1	3.2	-2.5	6.1	0.3484
Ala	30	15	0.8	27.0	1.2	-4.8	15.9	0.4543
Ala	60	14	3.4	21.2	2.1	-5.4	9.5	0.6257
Val	2	17	-1.3	4.9	-0.3	-4.1	2.7	0.4948
Val	7	17	-0.9	7.9	0.4	-4.6	2.1	0.7206
Val	14	16	-2.2	6.1	-1.5	-4.0	1.3	0.1734
Val	30	15	-0.9	6.7	0.3	-6.0	3.4	0.9677
Val	60	14	-0.8	18.6	-3.9	-9.7	0.2	0.1099
Cys	2	17	-0.9	15.0	2.0	-4.9	5.0	0.7563
Cys	7	17	-1.3	5.8	-2.0	-3.8	2.0	0.2789
Cys	14	16	-3.4	12.4	-1.1	-3.1	3.5	0.5245
Cys	30	15	-4.6	14.6	-1.9	-6.1	3.5	0.2222
Cys	60	14	-7.4	12.3	-3.6	-7.8	0.0	0.0068
C?C	2	17	-1.5	5.0	0.0	-4.0	1.1	0.3910
C?C	7	17	-1.5	4.7	-0.2	-2.9	0.2	0.2605
C?C	14	16	-2.3	6.8	-0.1	-2.4	0.0	0.1140
C?C	30	15	-3.0	8.4	0.0	-4.0	0.1	0.2500
C?C	60	14	-1.2	4.9	-0.1	-3.0	0.2	0.2500
Cys/C-C	2	17	0.0	1.2	0.0	0.0	0.2	0.8350
Cys/C-C	7	17	0.0	1.2	0.1	0.0	0.2	0.4819
Cys/C-C	14	16	0.2	2.0	0.0	-0.2	0.3	0.8467
Cys/C-C	30	15	0.5	3.4	0.0	-0.7	0.1	0.6768
Cys/C-C	60	14	-0.2	1.3	0.0	-0.6	0.1	0.6406
Cit	2	17	-1.2	5.3	-0.5	-1.3	1.2	0.6523
Cit	7	17	-2.1	7.8	-0.7	-3.4	0.9	0.4800
Cit	14	16	0.1	4.8	-0.8	-2.7	1.4	0.5886
Cit	30	15	-2.5	8.9	-0.8	-2.3	2.2	0.5711
Cit	60	14	-2.9	9.4	-0.8	-2.9	1.2	0.2468
Car	2	17	-1.6	5.7	-0.3	-2.3	0.3	0.2918
Car	7	17	-1.9	5.5	-0.9	-2.1	1.3	0.4103
Car	14	16	-1.0	5.9	-0.4	-1.4	0.8	0.4712
Car	30	15	-1.8	5.5	-0.8	-2.9	0.7	0.2129
Car	60	14	-1.8	6.0	0.1	-2.8	1.2	0.5090
Orn	2	11	-1.6	4.9	-0.5	-5.1	1.6	0.4258
Orn	7	11	0.9	4.9	-0.1	-3.0	6.3	0.7002
Orn	14	10	-1.4	6.5	-0.3	-3.2	2.1	0.6953
Orn	30	9	-1.8	7.5	0.1	-4.6	2.2	0.8438
Orn	60	8	-2.1	5.3	-0.8	-4.9	1.8	0.6406

1:p-value for evaluating changes from baseline (day 1)

Table 2: *Summary statistics of outcome parameters, stratified by time point*

Parameter	Day	N	Mean	SD	Median	p-value[1]	p-value[2]	p-value[3]	p-value[4]	p-value[5]
Cre	1	17	75.5	54.5	64.8	0.7819	0.3005	0.8176	0.1754	0.5830
Cre	2	17	68.8	43.5	62.3	.	.	.	.	.
Cre	7	17	75.3	54.0	59.2	.	.	.	.	.
Cre	14	17	74.1	52.6	66.9	.	.	.	.	.
Cre	30	16	94.9	78.2	70.2	.	.	.	.	.
Cre	60	14	113.7	91.2	93.1	.	.	.	.	.
5-HT	1	17	109.7	24.7	110.0	0.6441	0.3061	0.0695	0.0250	0.9999
5-HT	2	17	107.0	25.2	102.4	.	.	.	.	.
5-HT	7	17	101.8	24.1	101.7	.	.	.	.	.
5-HT	14	17	115.9	24.4	121.3	.	.	.	.	.
5-HT	30	16	101.8	28.6	103.3	.	.	.	.	.
5-HT	60	14	103.0	26.8	105.8	.	.	.	.	.
DA	1	17	152.7	32.7	149.0	0.1454	0.3061	0.2069	0.8603	0.4631
DA	2	17	142.8	30.5	139.6	.	.	.	.	.
DA	7	17	137.8	23.2	133.7	.	.	.	.	.
DA	14	17	150.6	33.3	141.3	.	.	.	.	.
DA	30	16	145.3	35.3	141.3	.	.	.	.	.
DA	60	14	143.9	31.1	138.2	.	.	.	.	.
5-HT/DA	1	17	0.7	0.2	0.7	0.3231	0.7379	0.5034	0.1330	0.7354
5-HT/DA	2	17	0.8	0.2	0.9	.	.	.	.	.
5-HT/DA	7	17	0.8	0.2	0.7	.	.	.	.	.
5-HT/DA	14	17	0.8	0.2	0.8	.	.	.	.	.
5-HT/DA	30	16	0.7	0.3	0.6	.	.	.	.	.
5-HT/DA	60	14	0.7	0.2	0.6	.	.	.	.	.
NE	1	17	37.6	9.3	37.3	0.4307	0.0448	0.1740	0.5966	0.5416
NE	2	17	35.4	8.5	36.2	.	.	.	.	.
NE	7	17	39.7	9.0	38.0	.	.	.	.	.
NE	14	17	35.8	8.5	35.9	.	.	.	.	.
NE	30	16	34.9	8.6	36.6	.	.	.	.	.
NE	60	14	32.8	8.9	32.5	.	.	.	.	.
E	1	17	6.9	1.5	6.8	0.3230	0.1260	0.3554	0.5352	0.3182
E	2	17	6.7	1.5	7.2	.	.	.	.	.

E	7	17	7.3	1.3	7.3	.	.	.	.	.
E	14	17	6.8	1.6	6.8	.	.	.	.	.
E	30	16	6.5	1.5	6.4	.	.	.	.	.
E	60	14	7.2	1.2	7.1	.	.	.	.	.
NE/E	1	17	5.5	1.2	5.4	0.9632	0.7057	0.9365	0.9780	0.1189
NE/E	2	17	5.4	1.2	5.6	.	.	.	.	.
NE/E	7	17	5.5	1.2	5.0	.	.	.	.	.
NE/E	14	17	5.3	0.8	5.3	.	.	.	.	.
NE/E	30	16	5.5	1.3	5.3	.	.	.	.	.
NE/E	60	14	4.7	1.4	4.2	.	.	.	.	.
GABA	1	17	2.3	1.3	2.3	0.4111	0.8991	0.6524	0.9999	0.8508
GABA	2	17	2.1	1.4	1.8	.	.	.	.	.
GABA	7	17	2.1	1.3	1.8	.	.	.	.	.
GABA	14	17	2.0	1.2	1.9	.	.	.	.	.
GABA	30	16	2.0	1.1	1.9	.	.	.	.	.
GABA	60	14	2.3	1.1	2.4	.	.	.	.	.
Glu	1	17	9.3	3.9	9.5	0.8626	0.6112	0.4947	0.7721	0.9595
Glu	2	17	9.5	2.9	9.6	.	.	.	.	.
Glu	7	17	9.1	3.2	9.6	.	.	.	.	.
Glu	14	17	9.7	3.1	10.1	.	.	.	.	.
Glu	30	16	9.2	3.4	9.8	.	.	.	.	.
Glu	60	14	9.8	3.9	10.3	.	.	.	.	.
Hist	1	17	17.0	5.4	17.3	0.1059	0.3969	0.7119	0.1266	0.7966
Hist	2	17	15.4	4.7	15.0	.	.	.	.	.
Hist	7	17	16.8	5.6	15.1	.	.	.	.	.
Hist	14	17	17.4	6.0	17.2	.	.	.	.	.
Hist	30	16	14.7	4.1	13.4	.	.	.	.	.
Hist	60	14	15.0	3.7	15.8	.	.	.	.	.
TA	1	17	12.0	6.9	10.4	0.1901	0.9174	0.3348	0.5035	0.8196
TA	2	17	10.4	5.8	8.1	.	.	.	.	.
TA	7	17	10.2	6.0	8.3	.	.	.	.	.
TA	14	17	11.1	6.1	12.0	.	.	.	.	.
TA	30	16	10.3	6.5	9.1	.	.	.	.	.
TA	60	14	10.4	6.3	10.4	.	.	.	.	.
LDOPA	1	17	31.4	8.4	31.0	0.0963	1.0000	0.8176	0.3484	0.1040
LDOPA	2	17	27.0	6.9	28.5	.	.	.	.	.
LDOPA	7	17	28.4	10.2	28.4	.	.	.	.	.
LDOPA	14	17	27.7	9.4	24.9	.	.	.	.	.
LDOPA	30	16	26.7	7.3	25.6	.	.	.	.	.
LDOPA	60	14	34.6	11.7	33.4	.	.	.	.	.
ME	1	17	27.3	6.9	27.9	0.4874	0.3348	0.0459	0.0021	0.6698
ME	2	17	26.3	6.7	26.5	.	.	.	.	.

ME	7	17	25.2	8.0	22.1	.	.	.	.	.
ME	14	17	29.1	6.0	29.4	.	.	.	.	.
ME	30	16	25.1	7.3	25.1	.	.	.	.	.
ME	60	14	24.5	8.1	25.3	.	.	.	.	.
NorM	1	17	112.0	43.8	116.7	0.6441	0.6777	0.3290	0.1439	1.0000
NorM	2	17	107.0	39.2	108.9	.	.	.	.	.
NorM	7	17	111.4	36.8	106.0	.	.	.	.	.
NorM	14	17	122.7	32.1	121.6	.	.	.	.	.
NorM	30	16	111.0	37.7	103.0	.	.	.	.	.
NorM	60	14	114.7	37.8	115.0	.	.	.	.	.
3-MT	1	17	32.2	11.3	30.5	1.0000	0.2633	0.7467	0.2576	1.0000
3-MT	2	17	31.7	9.9	30.1	.	.	.	.	.
3-MT	7	17	29.2	16.1	24.3	.	.	.	.	.
3-MT	14	17	30.4	13.6	29.7	.	.	.	.	.
3-MT	30	16	26.8	11.9	25.0	.	.	.	.	.
3-MT	60	14	30.1	16.5	23.4	.	.	.	.	.
Hcys	1	17	1.0	0.3	1.1	0.2749	0.9100	0.8781	0.8027	0.5745
Hcys	2	17	1.1	0.3	1.1	.	.	.	.	.
Hcys	7	17	1.1	0.5	1.0	.	.	.	.	.
Hcys	14	17	1.1	0.4	1.1	.	.	.	.	.
Hcys	30	16	1.0	0.4	1.0	.	.	.	.	.
Hcys	60	14	1.0	0.3	1.1	.	.	.	.	.
HCys2	1	17	1.4	0.6	1.2	0.1328	0.9522	0.5896	0.6278	0.0308
HCys2	2	17	1.6	0.6	1.3	.	.	.	.	.
HCys2	7	17	1.5	0.7	1.3	.	.	.	.	.
HCys2	14	17	1.3	0.5	1.3	.	.	.	.	.
HCys2	30	16	1.3	0.5	1.1	.	.	.	.	.
HCys2	60	14	1.1	0.4	1.1	.	.	.	.	.
Hcys/HCys2	1	17	0.9	0.3	0.9	0.1626	0.5871	0.4304	0.5336	0.0513
Hcys/HCys2	2	17	0.8	0.3	0.7	.	.	.	.	.
Hcys/HCys2	7	17	0.8	0.2	0.8	.	.	.	.	.
Hcys/HCys2	14	17	0.9	0.4	0.8	.	.	.	.	.
Hcys/HCys2	30	16	0.9	0.3	0.9	.	.	.	.	.
Hcys/HCys2	60	14	1.0	0.4	1.1	.	.	.	.	.
Cystathionine	1	17	14.9	6.8	13.9	0.0224	0.4874	0.4800	0.6412	0.8198
Cystathionine	2	17	13.1	6.6	11.4	.	.	.	.	.
Cystathionine	7	17	14.9	7.2	13.9	.	.	.	.	.
Cystathionine	14	17	13.3	6.7	13.9	.	.	.	.	.
Cystathionine	30	16	12.4	6.8	12.6	.	.	.	.	.
Cystathionine	60	14	14.7	8.4	14.4	.	.	.	.	.
AAA	1	17	13.4	9.0	11.4	0.2744	0.2293	0.4657	0.0258	0.4917
AAA	2	17	15.2	12.1	12.1	.	.	.	.	.

AAA	7	17	14.3	12.4	9.6	.	.	.	.	.
AAA	14	17	14.2	11.1	7.9	.	.	.	.	.
AAA	30	16	12.3	10.7	6.6	.	.	.	.	.
AAA	60	14	10.9	10.6	5.6	.	.	.	.	.
ABA	1	17	7.5	3.1	7.1	0.9357	0.4515	0.0910	0.4716	0.4854
ABA	2	17	7.4	2.2	6.9	.	.	.	.	.
ABA	7	17	7.1	2.6	6.3	.	.	.	.	.
ABA	14	17	8.2	2.9	6.9	.	.	.	.	.
ABA	30	16	7.8	3.2	6.5	.	.	.	.	.
ABA	60	14	7.3	2.6	7.6	.	.	.	.	.
BAIBA	1	17	106.6	45.4	92.9	0.7033	0.6197	0.3529	0.7339	0.2349
BAIBA	2	17	104.5	48.8	97.3	.	.	.	.	.
BAIBA	7	17	103.9	46.2	99.6	.	.	.	.	.
BAIBA	14	17	98.5	44.5	94.4	.	.	.	.	.
BAIBA	30	16	110.7	43.0	114.8	.	.	.	.	.
BAIBA	60	14	120.8	43.5	112.8	.	.	.	.	.
HYP	1	17	14.0	2.8	15.3	0.1231	0.0348	0.2633	0.9800	0.7728
HYP	2	17	12.9	3.2	13.7	.	.	.	.	.
HYP	7	17	11.2	2.8	11.3	.	.	.	.	.
HYP	14	17	12.0	3.1	11.6	.	.	.	.	.
HYP	30	16	11.7	3.8	12.0	.	.	.	.	.
HYP	60	14	11.4	3.8	10.5	.	.	.	.	.
Hlys	1	17	5.3	2.1	5.9	0.9171	0.1486	0.5097	0.7153	0.5919
Hlys	2	17	5.4	1.9	5.1	.	.	.	.	.
Hlys	7	17	4.7	1.4	5.1	.	.	.	.	.
Hlys	14	17	4.9	1.8	5.2	.	.	.	.	.
Hlys	30	16	5.1	1.6	5.3	.	.	.	.	.
Hlys	60	14	5.4	1.5	5.2	.	.	.	.	.
PEA	1	17	1.5	0.5	1.4	0.3221	0.9905	0.9173	0.9271	0.3130
PEA	2	17	1.6	0.5	1.5	.	.	.	.	.
PEA	7	17	1.5	0.5	1.5	.	.	.	.	.
PEA	14	17	1.5	0.4	1.5	.	.	.	.	.
PEA	30	16	1.5	0.4	1.5	.	.	.	.	.
PEA	60	14	1.4	0.4	1.4	.	.	.	.	.
Hser	1	17	4.4	1.7	4.1	0.2107	0.5710	0.9499	0.7150	0.4537
Hser	2	17	4.6	1.6	4.3	.	.	.	.	.
Hser	7	17	4.3	2.0	3.6	.	.	.	.	.
Hser	14	17	4.3	2.0	3.7	.	.	.	.	.
Hser	30	16	4.1	1.1	4.3	.	.	.	.	.
Hser	60	14	4.3	1.5	4.2	.	.	.	.	.
GSH	1	17	1.4	0.5	1.3	0.4262	0.0857	0.9222	0.2217	0.5085
GSH	2	17	1.4	0.4	1.3	.	.	.	.	.

GSH	7	17	1.6	0.5	1.5	.	.	.	.	.
GSH	14	17	1.6	0.5	1.8	.	.	.	.	.
GSH	30	16	1.4	0.4	1.4	.	.	.	.	.
GSH	60	14	1.4	0.5	1.3	.	.	.	.	.
Sar	1	17	4.4	4.5	2.6	0.1114	0.1629	0.6773	0.6690	0.2744
Sar	2	17	2.7	1.8	2.0	.	.	.	.	.
Sar	7	17	3.6	3.0	2.1	.	.	.	.	.
Sar	14	17	3.3	3.5	2.0	.	.	.	.	.
Sar	30	16	3.5	2.4	2.6	.	.	.	.	.
Sar	60	14	3.0	2.2	2.2	.	.	.	.	.
Arg	1	17	10.9	6.0	9.1	0.6527	0.7725	0.6778	0.3815	0.3054
Arg	2	17	10.6	6.4	8.8	.	.	.	.	.
Arg	7	17	11.3	6.5	9.9	.	.	.	.	.
Arg	14	16	10.6	5.4	9.4	.	.	.	.	.
Arg	30	15	11.3	7.5	9.5	.	.	.	.	.
Arg	60	14	10.1	7.0	8.1	.	.	.	.	.
Pro	1	17	12.6	15.6	8.5	0.9632	0.4038	0.7820	0.1733	0.7483
Pro	2	17	12.1	11.9	6.8	.	.	.	.	.
Pro	7	17	11.4	8.1	9.9	.	.	.	.	.
Pro	14	16	13.6	12.6	7.1	.	.	.	.	.
Pro	30	15	11.2	8.3	7.3	.	.	.	.	.
Pro	60	14	12.1	9.6	8.6	.	.	.	.	.
Lys	1	17	26.2	23.5	16.4	0.2633	0.0032	0.1973	0.9341	0.9460
Lys	2	17	20.2	12.7	15.9	.	.	.	.	.
Lys	7	17	30.0	23.3	15.2	.	.	.	.	.
Lys	14	16	25.4	30.5	15.2	.	.	.	.	.
Lys	30	15	18.5	8.9	16.0	.	.	.	.	.
Lys	60	14	16.8	12.3	12.8	.	.	.	.	.
Leu	1	17	10.2	4.9	9.0	0.5711	0.8982	0.3551	0.3375	0.2813
Leu	2	17	10.3	8.1	8.1	.	.	.	.	.
Leu	7	17	10.2	6.5	9.6	.	.	.	.	.
Leu	14	16	10.7	3.8	10.0	.	.	.	.	.
Leu	30	15	9.7	4.7	9.3	.	.	.	.	.
Leu	60	14	8.5	3.7	7.5	.	.	.	.	.
Ile	1	17	97.8	134.0	8.4	0.6355	0.2583	0.7197	0.2349	0.5186
Ile	2	17	85.1	117.2	10.2	.	.	.	.	.
Ile	7	17	95.4	131.6	9.6	.	.	.	.	.
Ile	14	16	93.3	117.4	10.7	.	.	.	.	.
Ile	30	15	92.6	115.3	8.9	.	.	.	.	.
Ile	60	14	104.0	123.6	10.3	.	.	.	.	.
Gln	1	17	169.2	70.9	193.7	0.6356	0.0004	0.1754	0.5336	0.2163
Gln	2	17	178.8	87.0	197.0	.	.	.	.	.

Gln	7	17	206.0	82.8	231.0	.	.	.	.	.
Gln	14	16	188.4	89.2	194.5	.	.	.	.	.
Gln	30	15	190.3	96.2	194.7	.	.	.	.	.
Gln	60	14	165.0	96.7	154.4	.	.	.	.	.
His	1	17	102.8	88.6	69.8	0.0569	0.0013	0.9100	0.1354	0.4548
His	2	17	73.7	53.2	58.9	.	.	.	.	.
His	7	17	93.8	65.7	73.8	.	.	.	.	.
His	14	16	93.7	68.4	75.9	.	.	.	.	.
His	30	15	125.3	108.2	80.2	.	.	.	.	.
His	60	14	87.9	84.8	57.1	.	.	.	.	.
Trp	1	17	14.8	14.2	13.6	0.6526	0.2078	0.1826	0.0859	0.6909
Trp	2	17	13.7	13.0	16.1	.	.	.	.	.
Trp	7	17	17.5	17.9	19.2	.	.	.	.	.
Trp	14	16	11.3	9.6	11.7	.	.	.	.	.
Trp	30	15	15.0	16.4	12.5	.	.	.	.	.
Trp	60	14	10.1	10.5	8.5	.	.	.	.	.
Tryp	1	17	26.5	38.3	1.6	0.9899	0.2918	0.3026	0.6724	0.6616
Tryp	2	17	27.6	39.3	1.7	.	.	.	.	.
Tryp	7	17	23.7	32.4	1.6	.	.	.	.	.
Tryp	14	16	29.6	39.4	1.6	.	.	.	.	.
Tryp	30	15	31.4	41.9	1.6	.	.	.	.	.
Tryp	60	14	32.9	41.0	1.7	.	.	.	.	.
5HTP	1	17	62.2	26.2	55.7	0.3061	0.9632	0.3287	0.7504	0.7334
5HTP	2	17	64.3	35.0	53.7	.	.	.	.	.
5HTP	7	17	63.1	34.4	53.7	.	.	.	.	.
5HTP	14	16	67.5	34.0	65.0	.	.	.	.	.
5HTP	30	15	67.2	37.8	53.7	.	.	.	.	.
5HTP	60	14	64.7	34.8	61.0	.	.	.	.	.
Tyr	1	17	123.6	150.0	82.4	0.5477	0.1454	0.3755	0.7197	0.5418
Tyr	2	17	102.3	99.1	60.1	.	.	.	.	.
Tyr	7	17	132.4	150.6	78.0	.	.	.	.	.
Tyr	14	16	113.5	112.6	62.4	.	.	.	.	.
Tyr	30	15	104.2	83.9	65.6	.	.	.	.	.
Tyr	60	14	133.5	133.2	81.1	.	.	.	.	.
Gly	1	17	134.5	109.1	183.8	0.0039	0.3290	0.8469	0.4212	0.1272
Gly	2	17	174.4	175.7	209.3	.	.	.	.	.
Gly	7	17	130.8	111.3	153.5	.	.	.	.	.
Gly	14	16	127.3	106.3	157.2	.	.	.	.	.
Gly	30	15	130.9	117.1	144.3	.	.	.	.	.
Gly	60	14	138.6	133.0	143.0	.	.	.	.	.
Bala	1	17	21.8	21.0	7.5	0.5703	0.6528	0.3755	0.8433	0.1465
Bala	2	17	22.7	23.3	20.2	.	.	.	.	.

Bala	7	17	24.0	24.1	16.0	.	.	.	.	.
Bala	14	16	19.9	19.9	11.5	.	.	.	.	.
Bala	30	15	21.4	19.4	13.5	.	.	.	.	.
Bala	60	14	28.1	22.2	25.9	.	.	.	.	.
Phe	1	17	62.4	37.5	57.3	0.4332	0.2247	0.5282	0.1205	0.5879
Phe	2	17	57.9	26.7	63.3	.	.	.	.	.
Phe	7	17	52.3	23.1	49.2	.	.	.	.	.
Phe	14	16	57.6	35.8	45.4	.	.	.	.	.
Phe	30	15	44.7	21.2	44.2	.	.	.	.	.
Phe	60	14	55.1	36.0	49.0	.	.	.	.	.
Asn	1	17	69.2	60.9	47.3	0.9265	0.3290	0.2522	0.4887	0.9460
Asn	2	17	82.0	62.9	66.2	.	.	.	.	.
Asn	7	17	70.6	59.4	39.6	.	.	.	.	.
Asn	14	16	89.1	62.6	92.1	.	.	.	.	.
Asn	30	15	71.2	53.2	63.5	.	.	.	.	.
Asn	60	14	70.6	60.4	56.2	.	.	.	.	.
Thr	1	17	21.5	17.8	12.8	0.3842	0.2202	0.1167	0.9014	0.4658
Thr	2	17	16.7	15.7	6.5	.	.	.	.	.
Thr	7	17	24.4	33.4	9.5	.	.	.	.	.
Thr	14	16	18.1	21.4	11.0	.	.	.	.	.
Thr	30	15	15.8	12.3	9.1	.	.	.	.	.
Thr	60	14	16.1	19.2	7.8	.	.	.	.	.
Met	1	17	3.4	2.7	2.8	0.9894	0.4559	0.6773	0.6900	0.5083
Met	2	17	3.4	2.2	3.4	.	.	.	.	.
Met	7	17	3.9	2.9	3.9	.	.	.	.	.
Met	14	16	3.7	2.1	3.7	.	.	.	.	.
Met	30	15	3.5	2.3	3.1	.	.	.	.	.
Met	60	14	3.6	2.5	3.3	.	.	.	.	.
Asp	1	17	17.9	26.4	2.3	0.8902	0.2166	0.5336	0.8669	0.0244
Asp	2	17	15.6	20.8	2.8	.	.	.	.	.
Asp	7	17	16.6	21.5	4.4	.	.	.	.	.
Asp	14	16	18.5	27.0	2.7	.	.	.	.	.
Asp	30	15	15.9	19.4	2.5	.	.	.	.	.
Asp	60	14	15.3	17.6	2.6	.	.	.	.	.
Ser	1	17	48.9	28.3	41.8	1.0000	0.9265	0.5707	0.2524	0.4143
Ser	2	17	46.2	23.1	42.9	.	.	.	.	.
Ser	7	17	49.6	22.2	43.3	.	.	.	.	.
Ser	14	16	52.2	36.3	41.3	.	.	.	.	.
Ser	30	15	41.3	17.5	36.3	.	.	.	.	.
Ser	60	14	38.9	19.2	33.1	.	.	.	.	.
Ala	1	17	42.1	43.8	26.6	0.7467	0.5171	0.6686	0.3028	0.7869
Ala	2	17	37.9	31.9	27.2	.	.	.	.	.

Ala	7	17	39.9	33.0	29.2	.	.	.	.	.
Ala	14	16	40.9	39.5	24.3	.	.	.	.	.
Ala	30	15	43.4	38.3	30.3	.	.	.	.	.
Ala	60	14	41.8	40.2	25.5	.	.	.	.	.
Val	1	17	22.4	7.8	23.1	0.4948	0.6777	0.3826	0.2022	0.1272
Val	2	17	21.2	8.7	17.8	.	.	.	.	.
Val	7	17	21.6	8.1	24.8	.	.	.	.	.
Val	14	16	20.3	7.9	21.0	.	.	.	.	.
Val	30	15	22.5	7.8	24.3	.	.	.	.	.
Val	60	14	23.0	14.7	21.8	.	.	.	.	.
Cys	1	17	32.3	10.6	30.1	0.7563	0.8502	0.6230	0.5522	0.2434
Cys	2	17	31.4	11.9	28.1	.	.	.	.	.
Cys	7	17	31.0	7.0	29.1	.	.	.	.	.
Cys	14	16	29.2	4.6	29.6	.	.	.	.	.
Cys	30	15	28.5	6.8	28.1	.	.	.	.	.
Cys	60	14	24.5	5.8	25.7	.	.	.	.	.
C-C	1	17	21.4	16.9	29.1	0.3910	0.6621	0.9482	0.5723	0.2754
C-C	2	17	19.9	15.7	23.9	.	.	.	.	.
C-C	7	17	19.9	16.3	25.2	.	.	.	.	.
C-C	14	16	18.7	14.4	27.3	.	.	.	.	.
C-C	30	15	17.4	14.3	22.1	.	.	.	.	.
C-C	60	14	17.3	15.3	24.2	.	.	.	.	.
Cys/C-C	1	17	1.9	1.6	1.0	0.8350	0.7344	0.5078	0.4883	0.7500
Cys/C-C	2	17	1.9	1.3	1.1	.	.	.	.	.
Cys/C-C	7	17	1.9	1.4	1.1	.	.	.	.	.
Cys/C-C	14	16	2.2	2.1	1.1	.	.	.	.	.
Cys/C-C	30	15	2.6	3.6	1.1	.	.	.	.	.
Cys/C-C	60	14	1.9	1.4	1.1	.	.	.	.	.
Cit	1	17	7.0	8.5	5.2	0.6523	0.4583	0.3372	0.5708	0.7864
Cit	2	17	5.8	4.1	4.7	.	.	.	.	.
Cit	7	17	4.9	2.4	4.7	.	.	.	.	.
Cit	14	16	7.2	8.1	3.9	.	.	.	.	.
Cit	30	15	4.8	1.5	4.8	.	.	.	.	.
Cit	60	14	4.8	2.7	5.3	.	.	.	.	.
Car	1	17	5.8	5.4	4.8	0.2918	0.5553	0.9097	0.8469	0.8259
Car	2	17	4.2	2.7	3.4	.	.	.	.	.
Car	7	17	3.9	1.8	3.6	.	.	.	.	.
Car	14	16	4.9	4.7	3.8	.	.	.	.	.
Car	30	15	4.2	2.6	3.1	.	.	.	.	.
Car	60	14	4.6	4.1	3.4	.	.	.	.	.
Orn	1	11	8.3	6.1	5.9	0.4258	0.1816	0.1602	0.9766	0.3750

Orn	2	11	6.7	2.8	6.3	.	.	.	.	.
Orn	7	11	9.2	4.4	9.9	.	.	.	.	.
Orn	14	10	7.1	1.5	6.7	.	.	.	.	.
Orn	30	9	7.2	2.5	6.2	.	.	.	.	.
Orn	60	8	6.8	3.0	6.6	.	.	.	.	.

p-value[1]: p-value for comparing Day 1 vs. Day 2
p-value[2]: p-value for comparing Day 2 vs. Day 7
p-value[3]: p-value for comparing Day 7 vs. Day 14
p-value[4]: p-value for comparing Day 14 vs. Day 30
p-value[5]: p-value for comparing Day 30 vs. Day 60