An Open-Label Study to Evaluate the Safety and Efficacy of a Novel Oral Nitric Oxide Supplement

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Introduction:

Nitric oxide (NO) is an important signaling molecule in the body, involved in virtually every organ and system in the body. It is known primarily for its role in vasodilation, and by extension its importance in maintaining normal blood pressure and blood flow to tissues. A deficiency in NO availability is often associated with cardiovascular conditions.

The discovery of the enterosalivary pathway, through which dietary ingestion of nitrates can promote NO production in the body, generated increasing interest in the potential for dietary supplementation for NO. A promising novel approach to improving the safety and efficacy of NO supplementation is to combine inorganic nitrates with hydrogen sulfide (H2S) precursors. Another important gaseous signaling molecule in the body, H2S interacts synergistically with NO while also exerting its own cardioprotective and vasodilatory effects.

This study was commissioned to evaluate the safety and efficacy of a novel nitric oxide support supplement designed to target the metabolism of both NO and H2S in the body. The study's primary endpoint was the change in blood pressure. A secondary endpoint was the change in nitric oxide levels, as measured by saliva NO test strips.

Methods:

Population: Participants (n=12) with mildly elevated blood pressure (130-140/85-90 mmHg) were recruited. None were on concomitant supplements, and any medications were maintained with no changes for the duration of the study or the preceding six months.

Participants included 4 males aged 65-73; 8 females aged 52-73. Eleven participants had systolic blood pressure above 130 mmHg. Of the 12 participants, 6 had normotensive diastolic readings, while 6 were mildly hypertensive for diastolic (diastolic hypertensive defined as >=80; Diastolic normotensive defined as <80; this subgroup of participants ranged from 60-75 yo).

Intervention: Participants ingested 2 capsules of a proprietary nitric oxide (NO) support supplement** every morning, for four weeks. Participants were asked to abstain from mouthwash and products containing chlorhexidine. They were also provided toothpaste, saliva nitric oxide strips of a commercially available brand, and if they did not have one at home, a blood pressure monitor.

Study protocol was reviewed and approved by an IRB board, and all participants signed consent forms.

Measurements: Study participants were evaluated in the clinic by a clinician at baseline, 2 weeks, and 4 weeks for salivary nitric oxide. Saliva nitric oxide test strips were used both in-clinic and at-home. Blood pressure was measured in the brachial artery 3 times, 5 minutes apart.

The study also included 30 days of in-home measurements, performed by the participants themselves. Subjects were instructed to use the NO saliva test strip and check their blood pressure upon arising each day, one hour before eating or brushing their teeth. The NO saliva test was repeated at 2 and 6 hours post-dose and the results recorded on the study testing record form.

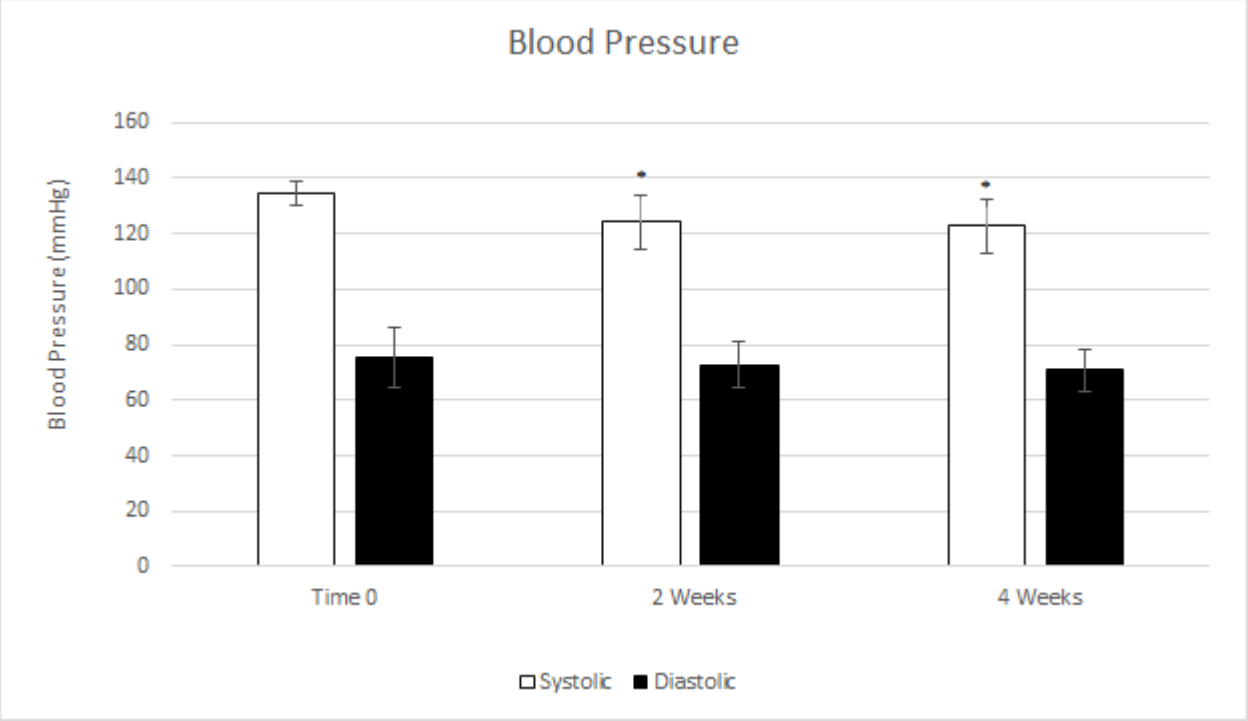
Data Analysis: Statistical analysis was completed at University of Maryland School of Medicine using SAS software (ANOVA and paired t-test analysis).

Note: Salivary nitric oxide test strips measure NO_2^- levels as a proxy for nitric oxide.

Results:

Blood Pressure

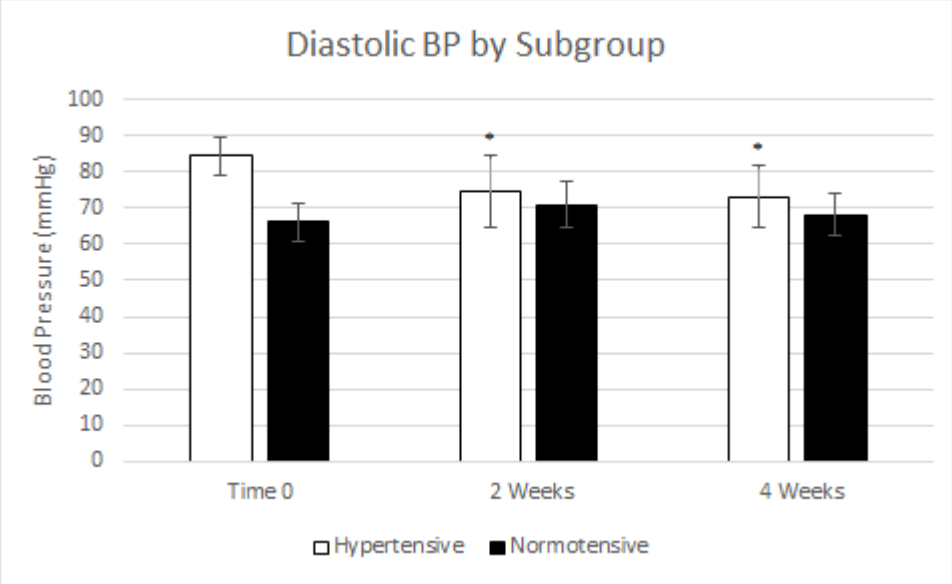
In-clinic systolic blood pressure decreased by 10.3 points at 2 weeks and a further 1.3 points at 4 weeks when compared to baseline at Time 0, statistically significant by ANOVA (p=0.003).



	Baseline	2 weeks	4 weeks
Systolic (mmHg)	134.4 ± 4.5	124.1 ± 9.9	122.8 ± 9.7
Diastolic (mmHg)	75.3 ± 10.8	72.8 ± 8.2	70.8 ± 7.5

Of the 12 participants, 6 had normotensive diastolic readings, while 6 were mildly hypertensive for diastolic (Diastolic hypertensive defined as >=80 Diastolic normotensive defined as <80 normotensive subjects ranged from 60-75). Overall ANOVA analysis of the 12 participants revealed that diastolic blood pressure did not significantly change (p=0.464).

An analysis was undertaken to assess the impacts of the proprietary nitric oxide support supplement on the participants (n=6) with hypertensive diastolic in-clinic blood pressure measurements. Paired t-tests show that the change in diastolic blood pressure within this subgroup approaches significance at 2 weeks (p=0.057) and is significant at 4 weeks (p=0.021). The 6 normotensive participants showed no reduction in diastolic blood pressure.

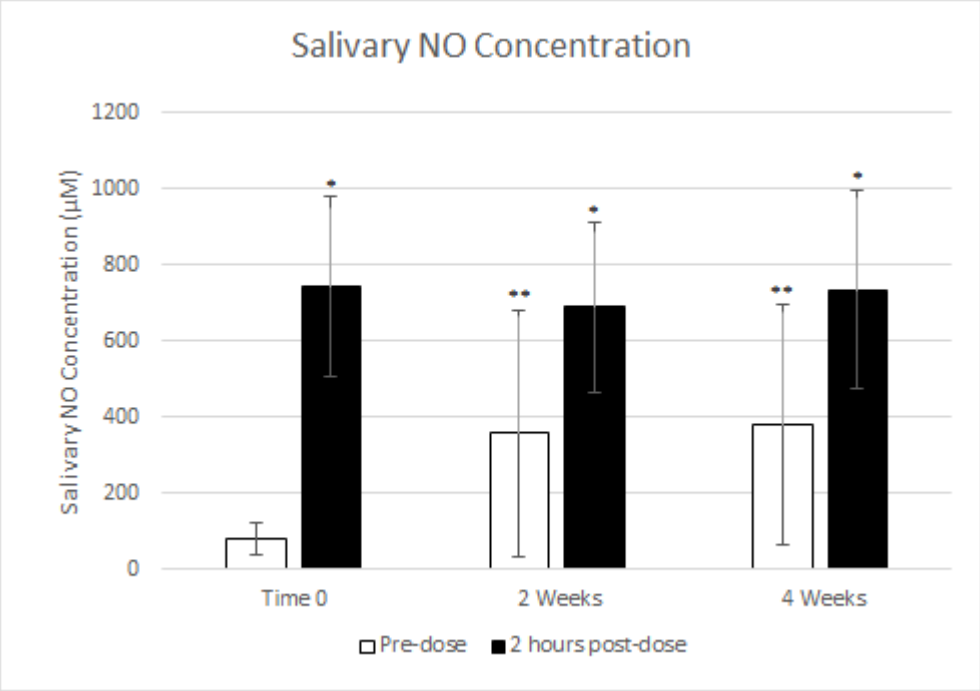


	Baseline	2 weeks	4 weeks
Hypertensive (≥80)	84.4 ± 5.4	74.5 ± 10.0 *(p<0.057)*	73.2 ± 8.4 *(p<0.021)*
Normotensive (<80)	66.2 ± 5.1	71.1 ± 6.4	68.3 ± 6.1

*p values are for paired t-tests comparing with baseline

Salivary Nitric Oxide Levels

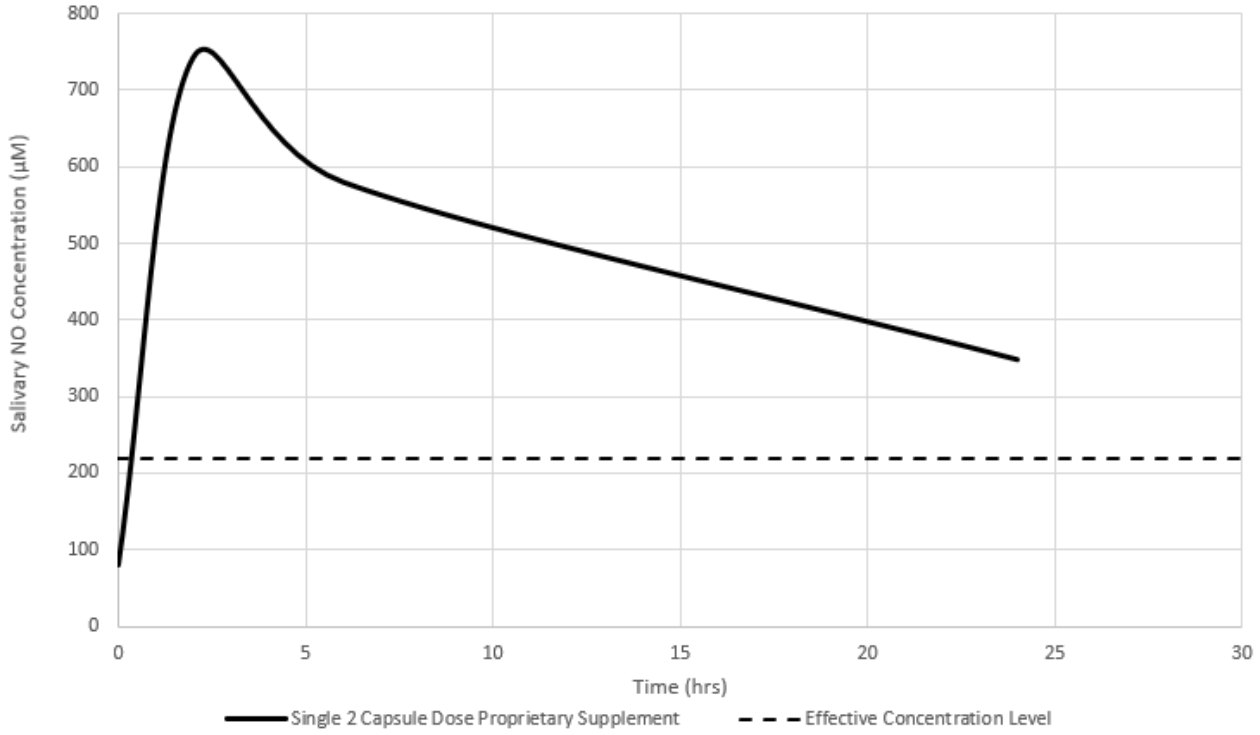
Baseline in-clinic salivary test strip mean was 80.0 (SD +/-44.3) micromolar (μM). At 0, 2, and 4 weeks, salivary nitric oxide levels (measured by salivary test strips) showed significant increases from pre-dose baseline to 2 hours post-dose (see Figure). Importantly, a significant change in pre-dose salivary NO levels was observed by ANOVA (p=0.014). (2 weeks: 356.3 SD +/-325.8 micromolar; 4 weeks: 381.7 SD +/-315.5 micromolar).



	Time 0: Pre-Dose	Time 0: Post-Dose	Week 2: Pre-Dose	Week 2: Post-Dose	Week 4: Pre-Dose	Week 4: Post-Dose
Mean (μM)	80.0 ± 44.3	743.3 ± 235.2	356.3 ± 325.8	688.8 ± 224.8	381.7 ± 315.5	734.2 ± 258.4
p value		<0.001		<0.008		<0.003

*p values are for paired t-tests comparing pre- and post-dose means at each time point

Salivary nitric oxide levels (measured by salivary test strips) of participants showed prolonged statistically significant increases after the first single dose of the proprietary nitric oxide support supplement, rising above 300 µM within 2 hours, and staying above 300 µM for more than 24 hours.



Time	0	2 Hours	6 Hours	24 Hours
NO (µM)	80.0 ± 44.3	743.3 ± 235.2	580.4 ± 321.8	348.3 ± 342.5

Conclusion:

The proprietary nitric oxide support supplement, which contains a synergistic blend of nitric oxide (NO) and hydrogen sulfide (H2S) precursors, resulted in improved metrics for the study population. Salivary nitric oxide strip measurements increased to above 300 µM within two hours after a single dose and remained above 300 µM for 24 hours. Blood pressure was within the normal range at 2 and 4 weeks, a statistically significant improvement. Importantly, no significant change was observed in diastolic blood pressure within the six individuals with diastolic normotensive readings at baseline, an important safety consideration. There were no reports of adverse events during the study.

This pilot study offers promising results that warrant further study with an increased number and diversity of participants.

Disclosures:

Study funded by Calroy Health Sciences, LLC.
Calroy was not involved in patient selection, data collection, or statistical analysis of the data.
* Dr. Houston is a member of Calroy's Scientific Medical and Advisory Board, and he is compensated for his participation in board meetings.
** Proprietary nitric oxide support supplement (Vascanox HP) supplied by Calroy Health Sciences.

Citations for further learning:

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