China-born (Alex) Haoci Liu is currently doing his PhD at UWA’s School of Medicine and Pharmacology, at Royal Perth Hospital.

Before coming to UWA in 2010, Alex completed a Bachelor of Biotechnology in his home country, and then enrolled in a 4-year ‘Doctor of Medicine’ course at Shanghai Jiao Tong University. He suspended his medical training (with two years left to go), when he was offered a scholarship to do a PhD at UWA, sponsored by the China Scholarship Council and UWA.

Alex’s research focuses mainly on the health benefits of nitrate-rich leafy green vegetables and beetroot, and he will attend the 8th Asia Pacific Conference on Clinical Nutrition in Tokyo to present some of his work later this year.
Acute benefits of a rich green-leafy vegetable diet on arterial stiffness and blood pressure in healthy volunteers

Alex Haoci Liu

Supervisors: Prof. Jonathan Hodgson
Prof. Kevin Croft
Acknowledgement

Supervisors:
   Prof. Jonathan Hodgson    Prof. Kevin Croft

Technical and laboratory support: Lisa Rich

Clinician:
   Dr. Trevor Redgrave

Funding:  NHMRC

Spinach supply:
   Logan Farms
Health benefits of spinach

What we are facing:
CVD Australia’s biggest killer

46,100 deaths in 2009: 1/3 of all

What we have found:
An easy and affordable approach
## Nitrate and green leafy vegetables

<table>
<thead>
<tr>
<th>Nitrate content (mg/100 g fresh)</th>
<th>Green leafy vegetable varieties</th>
</tr>
</thead>
<tbody>
<tr>
<td>Very high, &gt;250</td>
<td>Celery, cress, chervil, lettuce, red beetroot, <strong>spinach</strong>, rocket, radish</td>
</tr>
<tr>
<td>High, 100 to &lt;250</td>
<td>Celeriac, Chinese cabbage, endive, fennel, kohlrabi, leek, parsley</td>
</tr>
<tr>
<td>Middle, 50 to &lt;100</td>
<td>Cabbage, dill, savoy cabbage</td>
</tr>
<tr>
<td>Low, 20 to &lt;50</td>
<td>Broccoli, cauliflower, chicory</td>
</tr>
<tr>
<td>Very low, &lt;20</td>
<td>Artichoke, asparagus, green beans</td>
</tr>
<tr>
<td>Article</td>
<td>Subject</td>
</tr>
<tr>
<td>--------------------</td>
<td>------------</td>
</tr>
<tr>
<td>Bahra et al, 2012</td>
<td>healthy</td>
</tr>
<tr>
<td>Kapil et al, 2010</td>
<td>healthy</td>
</tr>
<tr>
<td>Webb et al, 2008</td>
<td>healthy</td>
</tr>
</tbody>
</table>
Metabolism of nitrogen oxides

1. Nitrite ingested from food
2. Bacteria in the mouth reduce nitrate to nitrite
3. In the acidic environment of the stomach, nitrite is reduced to nitric oxide
4. Nitrate and remaining nitrite is absorbed in the intestine
5. Nitrate is actively taken up from the blood into salivary glands
6. Nitrate and nitrite blood originate from the food and from systemic NO production
7. Nitrate is excreted by the kidneys
Nitric oxide-mediated vasodilation
Aim

To assess the acute effects of a nitrate-rich meal containing spinach on arterial stiffness and blood pressure in healthy men and women.
Participants

General Population

Advertisement

Healthy volunteers

Telephone and physical screening

Study participants

6 males and 20 females
### Characteristics of participants

<table>
<thead>
<tr>
<th></th>
<th>Range</th>
<th>Mean</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>38 - 69</td>
<td>58.8</td>
<td>7.6</td>
</tr>
<tr>
<td>Height (m)</td>
<td>1.48 - 1.80</td>
<td>1.65</td>
<td>7.7</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>44.8 - 91.5</td>
<td>69.5</td>
<td>11.7</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>19.1 - 30.4</td>
<td>25.4</td>
<td>3.3</td>
</tr>
<tr>
<td>SBP (mm Hg)</td>
<td><strong>110 - 130</strong></td>
<td>119.4</td>
<td>7.0</td>
</tr>
<tr>
<td>DBP (mm Hg)</td>
<td><strong>58 - 80</strong></td>
<td>70.8</td>
<td>6.5</td>
</tr>
<tr>
<td>Total cholesterol (mmol/L)</td>
<td>4.3 - 7.4</td>
<td>5.6</td>
<td>0.8</td>
</tr>
<tr>
<td>HDL cholesterol (mmol/L)</td>
<td>0.9 - 2.8</td>
<td>1.6</td>
<td>0.5</td>
</tr>
<tr>
<td>Triglycerides (mmol/L)</td>
<td>0.4 - 1.8</td>
<td>0.9</td>
<td>(0.8, 1.0)</td>
</tr>
<tr>
<td>Glucose (mmol/L)</td>
<td>4.5 - 5.7</td>
<td>5.1</td>
<td>0.4</td>
</tr>
</tbody>
</table>
Study design

Randomized controlled cross-over trial

Low nitrate dinner

Spinach meal (250 g spinach / 220 mg nitrate)

Control meal (rice milk 100ml as energy control)
Study day schedule

Baseline BP

Saliva collection

Spinach / control

30 min BP

60 min BP

90 min BP

120 min BP

150 min BP

180 min BP

210 min BP

Arterial stiffness assessments

PWV / Alx

Saliva collection

Artery elasticity

PWV / Alx

Artery elasticity
Pulse wave velocity (PWV)
Augmentation index (Alx)

- Reflected wave
- Forward wave
- Augmentation pressure (AP)
- Pulse pressure (PP)
- Mean arterial pressure
- Diastolic pressure
- Time to wave reflection (Tr)
Artery elasticity
Small artery elasticity index (ml/mm Hg X 100)

Large artery elasticity index (ml/mm Hg X 10)

Control

Spinach

P<0.001

P=0.98
Summary

dietary nitrate → nitrite and NO ↑

relaxation of SMC and vasodilation

arterial stiffness and blood pressure ↓
Future work

To determine if longer-term consumption has the same effects.

To determine if the same in population with hypertension and other CVD.