Prevalence Of Hyperhomocysteinemia In Patients With Predialysis Chronic Kidney Disease After Folic Acid Food Fortification Of The Canadian Food Supply

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An estimated 1.5 million Canadians have predialysis CKD.

The prevalence of predialysis CKD is expected to increase as a result of the global pandemic of type 2 diabetes, which is a major cause of predialysis CKD.
Stages Of CKD

5 to 10 times more likely to die than reach stage 5 CKD

Stage 1: GFR $\geq$ 90

Stage 2: GFR = 60-89

Stage 3: GFR = 30-59

Stage 4: GFR = 15-29

END STAGE RENAL DISEASE

Stage 5: GFR < 15

GFR = ml/min/1.73m²

Collins AJ, 2003
Prevalence Of Cardiovascular Disease - The General Population

Collins AJ, 2003
Homocysteine And Risk For Cardiovascular Disease (CVD)

- Plasma total homocysteine may be an independent risk factor for CVD in both the general and predialysis chronic kidney disease populations.

The Homocysteine Studies Collaboration, 2002; Jungers P, 1997
Prevalence Of Hyperhomocysteinemia (hHcy) (>15umol/L)

* Significant Difference (p<0.01)

Muntner P, 2004
# Severity Of Hyperhomocysteinemia In Various Populations

<table>
<thead>
<tr>
<th>Population</th>
<th>Range Of Plasma Total Homocysteine (umol/L)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>General</td>
<td>6 - 12</td>
<td>Friedman AN, 2002</td>
</tr>
<tr>
<td>Cardiovascular Disease</td>
<td>8 - 20</td>
<td>Gupta A, 1997</td>
</tr>
<tr>
<td>Predialysis CKD</td>
<td>6 – 50</td>
<td>Parsons DS, 2002</td>
</tr>
<tr>
<td>Classic Homocystinuria (Cystathionine β-synthase deficiency)</td>
<td>&gt; 500</td>
<td>Gupta A, 1997</td>
</tr>
</tbody>
</table>
Glomerular Filtration Rate (GFR) Is A Determinant Of Plasma Total Homocysteine (P tHcy)

Plasma tHcy was found to be inversely associated with GFR ($r = -0.39; p < 0.0001$).
Prevalence Of Hyperhomocysteinemia (hHcy) By Glomerular Filtration Rate (GFR) in Predialysis Chronic Kidney Disease

Parsons DS, 2002
Pathway of Homocysteine Metabolism

**Remethylation Pathway**
- Glycolysis
  - Serine
  - THF
  - Vitamin B6
  - Methyl Acceptor
  - Methylated Acceptor
- Methionine synthesis
  - Methionine synthase
  - Vitamin B12
  - SAM
- MTHF reductase
- 5,10 MTHF
- NADP
- 5-MTHF

**Transsulfuration Pathway**
- Dietary Protein
- Endogenous Protein
- Vitamin B6
- Vitamin B12
- Vitamin B6
- Cystathionine β-synthase
- Cystathionine
- Taurine
- Glutathione & inorganic sulfates.

**Transmethylation Pathway**
- Homocysteine
- Betaine
- Dimethylglycine
- Methyl Acceptor
- Methylated Acceptor
- SAH
- SAM
- Cystathionine
- Cysteine
- Serine
- Glutathione & inorganic sulfates.
Prevalence Of Inadequate Dietary Folate Intake In The General Population

![Bar chart showing prevalence of inadequate dietary folate intake before and after folic acid food fortification.](chart.png)

* Significant Difference (p<0.001)
Red Blood Cell (RBC) Folate In The General Population

![Bar chart showing the mean RBC folate levels before and after folic acid food fortification.](chart.png)

* Significant Difference (p<0.001)

Ray JG, 2002
Serum Folate In The General Population

![Bar chart showing mean serum folate levels before and after folic acid food fortification.](chart.png)

- **BEFORE Folic Acid Food Fortification**: Mean Serum Folate (nmol/L) is approximately 20.
- **AFTER Folic Acid Food Fortification**: Mean Serum Folate (nmol/L) is significantly higher, approximately 30.

* Significant Difference (p<0.001)

Ray JG, 2002
Prevalence Of Hyperhomocysteinemia (hHcy) (≥13umol/L) In The General Population

* Significant Difference (p<0.001)

Jacques P, 1999
Prevalence Of Hyperhomocysteinemia (hHcy) (≥12umol/L) in people with predialysis CKD after Folic Acid Fortification of the Food Supply
Energy Intake By Glomerular Filtration Rate (GFR) In Predialysis Chronic Kidney Disease

* P<0.05 as compared with a GFR of ≥ 43

Duenhas MR, 2003
Protein Intake And Glomerular Filtration Rate (GFR) In Predialysis Chronic Kidney Disease

Protein Intake By GFR Quartile

* P<0.05 as compared with a GFR ≥ 43

Duenhas MR, 2003
In the predialysis CKD population:

- The prevalence of hyperhomocysteinemia has not been described after folic acid food fortification
- The prevalence of inadequate intakes of folate, vitamins B12 and B6, energy and protein have never been described and
- There is uncertainty as to the need for the supplementation of the vitamins folate, B12 and B6
Primary Objectives

In the pCKD population:

- To estimate the prevalence and severity of hyperhomocysteinemia
- To measure intake of folate, vitamins B12 and B6, protein and energy from food and supplements and estimate the prevalence of inadequate intake of these nutrients from food alone
Secondary Objectives

In the pCKD population:
1. To describe the associations between:
   Fasting plasma total homocysteine and
   - dietary and supplemental intakes of folate, vitamins B$_{12}$ and B$_{6}$, protein and energy.
   - vitamin status as measured by RBC folate and serum folate, B$_{12}$ and B$_{6}$.
   - glomerular filtration rate.
   - energy-protein status.
2. To describe the associations between:

Dietary intakes of folate, vitamins B\textsubscript{12} and B\textsubscript{6}, protein, energy and

- Gastrointestinal symptoms of uremia
- Glomerular filtration rate
Hypothesis

In the pCKD population:

- The prevalence of hyperhomocysteinemia (tHcy > 12 umol/L), after folic acid food fortification, will be 75%.
- The majority of these patients will have inadequate intakes of folate, vitamins B\textsubscript{12} and B\textsubscript{6}, protein and energy from food.
Methodology

Study Design
Prospective cross-sectional descriptive

Subjects
All new patient referrals to the predialysis clinics at St. Michael's Hospital and the University Health Network.
# Inclusion and Exclusion Criteria

<table>
<thead>
<tr>
<th>Inclusion criteria</th>
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<tbody>
<tr>
<td>≥ 18 years old</td>
<td>Liver or kidney transplant</td>
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<tr>
<td>GFR &lt; 60 mL/min/1.73m²</td>
<td>Cancer or liver disease</td>
</tr>
<tr>
<td>Able to read and understand English</td>
<td>Vitamin B₁₂ deficiency</td>
</tr>
<tr>
<td>Able to provide informed consent</td>
<td>Taking an antibiotic 7 days prior to blood sample</td>
</tr>
<tr>
<td></td>
<td>In another study that interferes with Hcy metabolism</td>
</tr>
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</table>
1ST visit

Fasting blood samples

Interview For:
- 24 hour dietary recall
- Micronutrient supplement use
- Gastrointestinal symptoms of uremia

Perform & Obtain Information For:
1. Subjective Global Assessment (SGA)
2. Anthropometrics including: height, weight, triceps skinfold (TSF), mid upper arm circumference (MAC)
Data Collection

1st visit

- Fasting blood samples
- Interview for:
  - 24 hour dietary recall
  - Micronutrient supplement use
  - Gastrointestinal symptoms of uremia
- Perform and obtain:
  - SGA
  - Anthropometrics including: BMI, TSF, MAC

1 week

2nd visit

- 24 hour dietary recall
<table>
<thead>
<tr>
<th>Sample</th>
<th>Analytical Method</th>
</tr>
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<tbody>
<tr>
<td>Plasma total homocysteine</td>
<td>Competitive immunoassay</td>
</tr>
<tr>
<td>Serum B&lt;sub&gt;12&lt;/sub&gt;</td>
<td>Competitive immunoassay</td>
</tr>
<tr>
<td>RBC folate</td>
<td>Microbiological assay</td>
</tr>
<tr>
<td>Serum Folate</td>
<td>Microbiological assay</td>
</tr>
<tr>
<td>Serum pyridoxal 5 phosphate</td>
<td>Tyrosine decarboxylase assay</td>
</tr>
</tbody>
</table>
Sample Size

- Sample size calculation \( n = 72 \) subjects

- Sample size estimate based on:
  1. A predicted prevalence of hyperhomocysteinemia of 75%
  2. A 95% confidence level
  3. A 20% confidence interval
Clinical Implications Of Our Research

- This will be the first study to describe the prevalence of hHcy in a pCKD population in the era of Folic Acid Fortification, and will elucidate if elevated plasma tHcy is still a health concern in the Canadian pCKD population.
This will be the first study to describe the prevalence of inadequate dietary intake of the vitamins involved in homocysteine metabolism in the CKD population in the era of Folic Acid Fortification. This information, considered in conjunction with vitamin status and p tHcy, may provide evidence to guide dietetic practice regarding whether or not dietitians should be prescribing vitamin supplements to people with pCKD.