Assessment of Nutritive and Anti–Nutritive Values and Mineral Content of *Vigna subterranea* Thouars (Bambara Nut) Seeds

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Authors’ contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

ABSTRACT

*Vigna subterranea* L. thouars (Bambara nut) seeds were analyzed for their nutritive and anti–nutritive values. The result showed that *Vigna subterranea* L. thouars seeds have moisture content 7.54%, ash content 3.46%, crude fat content 4.79%, crude protein content 22.40%, crude fibre content 1.65%, carbohydrate content 60.16% and available energy 1580.75 KJ. The mineral content of analyzed samples showed that *Vigna subterranea* L. thouars seeds have 57.39% and 55.67% of potassium and calcium, respectively. The high Ca/P ratio indicates that *Vigna subterranea* L. thouars is a very good food source. This study showed that oxalate, nitrate and cyanide values were below WHO standard. The low content of anti –nutrients of *Vigna subterranea* L. thouars showed that Bambara nut will not constitute health hazard when taken as food.

Keywords: *Vigna subterranea* L. thouars; nutritive value; anti – nutritive value; mineral compositions.

1. INTRODUCTION

The increase in number of undernourished people all over the world especially Nigeria has prompted investigation of indigenous plants of nutritional value [1-3]. A lot of indigenous plant species of high nutritive value have been reported [4-6]. However, *Vigna subterranea* L.
thouars is one of those plants with high nutritive and medicinal value.

*Vigna subterranea L. thouars* is a leguminous plant grown in Nigeria. The pods of Bambara nut are 1.5cm long and contains one to two seeds. Varies in colour from black, dark-brown, red, white, creamy or combination of these colours [7]. Bambara is widely distributed in the sub-Saharan Africa and it is the third most important legume food after cowpea and groundnut [8]. It is resistant to high temperature and also suitable for marginal soils where other leguminous crops cannot be grown. It makes little demand on soil and has a high nutritional value [9]. Bambara is an annual creeping leguminous plant, grown primarily for its edible seeds.

In Eastern part of Nigeria Bambara is called “Okpa” among the Igbos, whereas the Yoruba’s of Western part of Nigeria call it “Epa-kuta”. In Northern part of Nigeria, the Hausas call it “Gurjiya or Kwaruru”. Bambara are used for different purposes all over Africa. In Eastern part of Nigeria, Enugu State to be precise they roast and chew Bambara nut with coconut or palm Kernel as snacks. It is also fried with salt and chewed with groundnut. The freshly harvested ones are boiled with salt and eaten like groundnut. They are milled into flour and used to bake cakes.

In Ghana the Bambara nut are boiled with pepper and salt in preparation of their local food called Abobi [10]. The beans of Bambara nut are roasted and pulverized in East Africa to make soup with or without condiments. Bread made from Bambara nut has been reported in Zambia. In Senegal, the leaves are squeezed and applied on wounds. Leaf sap of Bambara is also applied to the eyes by local people to treat epilepsy [7]. The aim of present study is to investigate the proximate, anti-nutritive and mineral composition of seeds of *Vigna subterranea L. thouars* and its usefulness in human nutrition and health.

2. MATERIALS AND METHODS

2.1 Sample Collection and Preparation

Fresh seed samples of *Vigna subterranea L. thouars* were obtained from Eke –Ozzi market in Igbo Eze North Local Government Area of Enugu State, Nigeria. The nuts were sorted, washed, dried under the sun. The dried samples were ground into powder and sieved through 250μm mesh and stored in a plastic container until use.

2.2 Proximate Analysis

The moisture, ash and crude fibre contents of samples of *Vigna subterranea L. thouars* were determined using Standard Chemical Methods as described by Association of Officials Analytical Chemists [11] by drying 2g of the sample at 105°C for 24h for moisture content determination. The ash content was determined by incineration of 2g of the sample in a muffle furnace at 575±25°C for 2h. Soxhlet extraction technique using petroleum ether (40 – 50°C) was used to evaluate the fat content of the sample [12]. Kjedahl method was used to determine the crude protein content of the sample as described by [11]. The carbohydrate content of the sample was estimated by difference (% Carbohydrate = 100% - sum of percentage of moisture, ash, fat, crude fibre and crude protein contents).

2.3 Anti–Nutrient Analysis

The oxalate, cyanide and nitrate contents of samples of *Vigna subterranea L. thouars* were determined using Standard Chemical Methods as described by Association of Official Analytical Chemists [11]. This was carried out by digesting 2g of the sample with hydrochloric acid (HCl). Ammonium chloride (NH₄Cl) was added to the digested sample to make it alkaline. Then titration was carried out for oxalate content determination. The cyanide content of the sample was investigated using spectrophotometer at 540nm wavelength as described by AOAC [11]. The suspension of the sample was incubated for 1hr at 45°C as described by AOAC [11]. The nitrate content was determined by weighing 2g sample into 15ml centrifuge tube and 100ml of distilled water was added. The suspension was incubated for 1h at 45°C as described by AOAC [11].

2.4 Mineral Analysis

Mineral composition of the samples was determined according to method recommended by Association of Official Analytical Chemists [11]. 1g each of the sample of Vigna subterranea L. thouars was digested using 12cm³ mixture of Nitric acid (HNO₃), Sulphuric acid (H₂SO₄) and HClO₄ (9:2:1 V/V) [13]. After digestion of the sample. Copper, iron, zinc, sodium, potassium, calcium and magnesium content were determined using Atomic
Absorption Spectrophotometer (Pye-unicam 969 Cambridge, UK). Phosphorus content of the sample was determined using Flame Photometer.

3. RESULTS AND DISCUSSION

The results of the proximate composition of flour samples of *Vigna subterranean* L. thouars were depicted in Table 1. The result indicates that *Vigna subterranean* L. thouars has moisture content 7.54%. The value was higher than those reported for *Sesamum indicum* linn. (black – seed specie) by [14] and [6] of moisture content 6.00% and 3.46%, respectively. Olaofe et al., [5] reported that *Bombcapsis glabre* seed has moisture content 3.46%. Olayemi and Salihu [15] working on *cucurbita mixima* peel (Pumpkin) reported moisture content 5.20%. It has been reported [4] that *Pentaclethra macrophylla* benth (Africa Oil bean) seed has moisture content 11.87%. The moisture content of African oil bean seed was high when compared to value obtained in this study. The moisture contents of any materials is an index of its water activity and it is used to measure its stability and susceptibility of microbial contamination [16]. The result of this study showed that *Vigna subterranean* L. thouars has low moisture content 7.54%. The low moisture content observed in this study showed that *Vigna subterranean* L. thouars could be stored for a long period of time without deterioration.

The result of this study showed that *Vigna subterranean* L. thouars has ash content 3.46%. It has been reported [17] that *Monodora myristica* seed has ash content 6.50%. Lohdip and Jikmyan [14] reported ash content of 9.62% in *Sesamum indicum* L. pedaliaceae seed. The ash content of *sesamum indicum* L. pedaliaceae was high when compared to the value obtained from *Vigna subterranean* L. thouars. The ash content 3.62% of pumpkin peel as reported by Olayemi and Salihu [15] was similar to ash content of *Monodora myristica* seed. Ash content is an indication of the level of inorganic elements in the sample from which the mineral content could be obtained [18,19]. This suggest that *Vigna subterranean* L. thouars seeds could provide essential, valuable and useful minerals needed for body development.

This study showed that *Vigna subterranean* L. thouars has low fat content 4.79% when compared to fat content 46.95% reported for *Pentaclethra macrophylla* benth seed [4]. Olaofe et al. [5] working on *Bombcapsis glabre* seed reported fat content 34.8%. Paul and Southgate [20] reported fat content 23.5% in soybean seed. It has been reported [15] that pumpkin peel has low Fat content 3.11%. Fats are essential because they provide the body with maximum energy [21], but excess fat have been discovered in cardiovascular disorders such as atherosclerosis, cancer and aging [22]. In view of this, Bambara nut could be consumed to reduce the risk of these diseases. The crude protein content of Bambara nut was 22.40% lower than the value 43.1% obtained for seed of *Luffa Cylindrica* [23]. The crude protein content of this study is comparable to crude protein content 23.7% for gourd seed reported by [24]. It has been reported [25] that any plant food that provides about 12% of its calorific value from protein is considered a good source of protein. Therefore *Vigna subterranean* L. thouars nut could be used as an alternative source of protein in diet/protein supplement especially in underdeveloped countries such as Nigeria where majority of the populace live on starchy food and cereals.

The crude fibre content of flour of Bambara nut was found to be 1.65%. The value is lower than crude fibre contents 2.80% and 2.40% for gourd seeds and cowpea seeds as reported by Ogungbenle [6] and Aremu et al., [26] respectively. Fibre has useful role in providing roughage that aids digestion, increase dietary bulk, and makes stool larger and softer, due to their ability to absorb water, so preventing constipation [27,28]. Report has shown that low fibre diet has been associated with heart diseases, cancer of the colon and rectum, varicose veins, phlebitis, obesity, appendicitis, diabetics and even constipation [29]. This study showed that crude fibre content of Bambara nut was low when compared to crude fibre content of gourd seed and cowpea seed. Therefore, Bambara nut could not be used as an alternative source of dietary fibre. The carbohydrate content 60.16% analyzed for Bambara nut was high when compared to values 37.67%, 21.72% and 9.89% for pumpking peel, black seeds and gourd seeds as reported by Olayemi and Salihu [15], Lohdip and Jikmyan [14] and Ogungbenle [6], respectively. The high carbohydrate content of *Vigna subterranean* L. thouars suggests that Bambara nut is a better source of carbohydrate which is necessary for energy in man. Carbohydrate supplies energy to cells such as brain, muscles and blood. It contribute to fat metabolism and spare proteins as an energy source and act as mild natural laxative for
human beings and generally add to the bulk of the diet [30, 31]. The calculated metabolizable energy 1580.75 KJ/100g for this study was comparable to value 1595.34 KJ/100g for legumes as reported by Aremau et al., [26]. Alinnor and Oze [4] working on African oil bean seed reported calculated metabolizable energy to be 2344.56KJ/100g. The value was higher than the value obtained in this study. The high energy value of bambara nut may be attributed to high carbohydrate content.

The cyanide content 0.006mg/100g was analyzed for Bambara nut in this study. The value obtained was lower than the World Health Organization [32] recommended value of 0.54mg for children and 40mg for adult. Hydrogen cyanide is poisonous chemical with a fast acting effect. However, low levels of cyanide are found in nature and in product we commonly eat. Large doses of cyanide prevent cells from using oxygen and eventually these cells die. Organs such as heart, respiratory system and central nervous system are most susceptible to cyanide poisoning. Hydrogen cyanide inhibits enzyme containing metal such as cytochromoxidase containing iron. Cytochromoxidase is responsible for the energy-providing processes in the cell where oxygen is utilized [33,34].

The nitrate content of 2.60mg/100g was observed in Bambara nut in this study. The result was below the WHO [32] recommended value of 3.7mg for children and 222mg for adult. Nitrate is an inorganic chemical that is soluble in water. Presence of nitrate in food causes health hazard because of its conversion to nitrite in the body. Once ingested, conversion of nitrate to nitrite takes place in the saliva of people of all age group, and in the gastrointestinal tract of infants [35]. Nitrite in the body changes the normal form of hemoglobin which carries oxygen in the blood to the rest of the body, into a form called methemoglobin that cannot carry oxygen. High concentration of nitrate in drinking water can result in a temporary blood disorder in infants called methemoglobinemia, commonly called “blue baby syndrome”. If severe cases are untreated it may result to brain damage, leukemia and can cause death as a result of suffocation due to lack of oxygen [36]. After nitrate is converted to nitrite in the body, it can react with any amine-containing substances found in food to form nitrosamine. Nitrosamine is a potent cancer causing chemical. Also nitrosamine formation may be inhibited by antioxidant that may be present in food such as vitamin C and vitamin E [37].

The oxalate content of Vigna subterranean L. thouars was 0.0047mg/100g. This study showed that oxalate value was below WHO recommended value of 4mg for children and 40mg for adult. Oxalates occur naturally in plants, animals and humans. Oxalates are very powerful oxidants that initiate free radicals. If oxalic acid is very high in the blood being filtered by the kidney, it may combine with calcium to form insoluble calcium oxalate crystals. The crystals formed may block urine flow and cause severe pain as it passes through. These crystals also form in the bones, joints, blood vessels, lungs, nerves and brain. In the bone, oxalate crystals may crowd out bone marrow cells, and can lead to anemia and suppressed immunity. Oxalate can also lead to autism especially in children [38,39].

Table 3 presents the mineral composition of Vigna subterranean L. thouars. Minerals are important component of diet because of their physiological and metabolic function in the body. The result shown in table 3 indicates that Vigna subterranean L. thouars has sodium content 4.50mg/100g. Sodium is an important mineral that assist in the regulation of body fluid and in the maintenance of electric potential in the body tissue. The sodium facilitates the improvement of brain function. It helps to prevent muscle cramp. It helps to maintain glucose absorption and

Table 1. Proximate composition of Vigna subterranean L. thouars (% dry weight)\(^a\)

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Vigna Subterranean L. Thouars (Bambara nut)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture content</td>
<td>7.54±0.01</td>
</tr>
<tr>
<td>Ash Content</td>
<td>3.46±0.02</td>
</tr>
<tr>
<td>Crude fat Content</td>
<td>4.79±0.01</td>
</tr>
<tr>
<td>Crude Protein Content</td>
<td>22.40±0.01</td>
</tr>
<tr>
<td>Crude fibre content</td>
<td>1.65±0.02</td>
</tr>
<tr>
<td>Available carbohydrate</td>
<td>60.16±0.02</td>
</tr>
<tr>
<td>Available energy (KJ)(^b)</td>
<td>1580.75</td>
</tr>
</tbody>
</table>

\(^a\) Values as mean ± standard deviation of triplicate determination; \(^b\) Calculated metabolisable energy (KJ/100g Sample): (protein x 17 + Fat x 37 +carbohydrate x17)
Table 2. Anti–nutrient level of *Vigna subterranean* L. thouars (mg/100g)*a*

<table>
<thead>
<tr>
<th>Parameters</th>
<th><em>Vigna Subterranean</em> L. Thouars (Bambara nut)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cyanide</td>
<td>0.006</td>
</tr>
<tr>
<td>Nitrate</td>
<td>2.60</td>
</tr>
<tr>
<td>Oxalate</td>
<td>0.0047</td>
</tr>
</tbody>
</table>

* Values are mean ± standard deviation of triplicate determination

Table 3. Mineral Composition of *Vigna subterranean* L. thouars samples (mg/100g)*a*

<table>
<thead>
<tr>
<th>Parameters</th>
<th><em>Vigna Subterranean</em> L. Thouars (Bambara nut)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium</td>
<td>4.50±0.01</td>
</tr>
<tr>
<td>Potassium</td>
<td>57.39±0.02</td>
</tr>
<tr>
<td>Calcium</td>
<td>55.67±0.02</td>
</tr>
<tr>
<td>Magnesium</td>
<td>15.78±0.03</td>
</tr>
<tr>
<td>Iron</td>
<td>2.08±0.01</td>
</tr>
<tr>
<td>Copper</td>
<td>0.012±0.02</td>
</tr>
<tr>
<td>Zinc</td>
<td>8.61±0.02</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>0.60±0.02</td>
</tr>
<tr>
<td>Na/K</td>
<td>0.08</td>
</tr>
<tr>
<td>Ca/P</td>
<td>92.78</td>
</tr>
<tr>
<td>Ca/Mg</td>
<td>3.53</td>
</tr>
</tbody>
</table>

* Values are mean ± standard deviation of triplicate determination

contraction of the heart. Deficiency of sodium lead to diarrhea, vomiting, headache and attack to the nervous systems. Also too much sodium in the body can cause high blood pressure, increase chance of heart disease, stroke, kidney damage, swelling of the legs and hand. It can also lead to confusion, seizures and even coma [40]. The World Health Organization’s (WHO) recommended intake of sodium per day is 150mg for adult and children [32]. The result indicates that sodium content of *Vigna subterranean* L. thouars sample was below WHO recommended standard, therefore *Vigna subterranean* L. thouars is not a good source of sodium.

This study indicates that potassium content of *Vigna subterranean* L. thouars was 57.39mg/100g. This study showed that potassium is the most abundant mineral. The observation was in agreement with the report of Olaofe and Sanni [41], Olaofe et al., [24], Aletor and Aladetimi [42], Badifu and Ogunsina [43] and [4] who have reported that potassium is the most abundant in agricultural products. Potassium is important in the regulation of heart beat, neurotransmission and water balance of the body. Potassium helps to prevent stroke and it maintains muscle function and optical nerve. It helps to maintain the normality of blood pressure in the body. It helps people suffering from stress and anxiety by ensuring efficient mental performance. Deficiency of potassium leads to fatigue and weakness in muscles, inactive reflexes, abnormal heart beat, heart palpitation and stroke. If potassium level get too high, nerve can fire randomly or not at all. This in turn affects involuntary muscles such as the heart. High level of potassium can cause irregular heart beat and heart attack. Skeletal muscle may also behave irregularly [44]. The recommended intake of potassium per day is 2000mg for adult and 1600mg for children. This study revealed that potassium content of *Vigna subterranean* L. thouars sample was below WHO standard, therefore *Vigna subterranean* L. thouars is not a good source of potassium.

The result of the analysis showed that calcium content of Bambara nut was 55.65mg/100. The recommended daily intake of calcium by WHO is 800mg for both adult and children. The result indicates that calcium content of *Vigna subterranean* L. thouars was below WHO standard. Calcium is an important mineral required for bone formation and neurological function of the body. It helps to protect the cardiac muscles by helping them contract and relax properly.

Calcium helps to prevent colon cancer. It helps to maintain health teeth and gum. It helps to prevent kidney stone and muscle cramps. Calcium deficiency can lead to Osteoporosis. It can lead to loose teeth and gum disease. Excess of calcium in the body can lead to muscle
weakness or pain and fatigue, stomach discomfort, excessive thirst and urination. Excessive calcium in the body can cause reduction in the body's absorption of zinc and iron [45]. This study indicates that magnesium content in Vigna subterranea L. thouars was 15.78mg/100g. The WHO recommended Dietary Allowance (RDA) for magnesium in adult is 350mg/100g, while children is 170mg/100g. The result showed that magnesium content of Vigna subterranea L. thouars was below WHO standard. Therefore Vigna subterranea L. thouars is not a good source of magnesium. Magnesium plays vital role in calcium metabolism in bones and also involve in prevention of circulatory diseases. It helps in regulating blood pressure and insulin releases. It helps to prevent asthma. It helps in collagen production and curing psychiatric disorders like panic attack, stress and anxiety. Deficiency of magnesium in the body can lead to diarrhea and cardiac arrest [46].

The result presented in Table 3 revealed that iron content of Vigna subterranea L. thouars was 2.08mg/100. The RDA for iron in adult and children is 10mg/day, while adult female is 15mg/day. This study indicates that iron content of bambara nut was below WHO standard. Iron is required for blood formation and it is important for normal functioning of the central nervous system [47]. It also facilitates the oxidation of carbohydrate, protein and fats. It helps to transport oxygen to the cells from the lungs. It helps in energy production in the cells by converting sugars, fats, proteins into adenosine triphosphate (ATP). It helps to protect the body from free radical damage by forming an enzyme known as catalase. It helps in cognitive function. Deficiency of iron leads to anemia and reduced immune function [48]. Excess of iron in the body causes liver problems (Cirrhosis, cancer), heart failure. In some cases excess iron can affect the reproductive system and lead to decreased libido in men, early menopause and impotence, alzheimer and parkinson disease [49]. This study revealed that copper content of Vigna subterranea L. thouars was 0.012mg/100g. The RDA for copper in adult is 3mg/day, while children is 2mg/day. The result indicates that copper content of the sample was below WHO recommended standard. Therefore bambara nut could not serve as a good source of copper. Copper is required in the body for enzyme production and biological electron transport. It helps to prevent and cure arthritis. Copper helps to protect the skeletal, nervous and cardiovascular systems. It helps to ensure proper functioning of the thyroid gland. It helps to reduce bad cholesterol and increase good cholesterol. Copper gives pigmentation to the hair and eyes by producing melanin which imparts colouration to the skin, hair and eyes. It also functions as an anti-oxidant. Deficiency of copper leads to low skin pigmentation, uneven heartbeat, dilated veins, disease [50]. Wilson disease is a disease that prevents the body from getting rid of excess copper. The copper builds up in the liver and it release the copper directly into the blood stream. This can cause damage to the brain, kidneys and eyes [51]. This study showed that zinc content of Vigna subterranea L. thouars was 8.62mg/100g. It has been reported [17] that zinc content of Monodora myristica whole seeds and dehulled seeds were 21.41mg/100g and 11.35mg/100g, respectively. Alinnor and Oze [4] reported high zinc content 1134.36mg/100g in Pentaclethra macrophylla benth samples. The WHO recommended standard for zinc in adult and children are 15mg/day and 10mg/day, respectively. The result of the analysis showed that zinc content of the sample was below WHO standard.

Therefore bambara nut could not serve as a good source of zinc. Zinc is an essential micronutrient associated with number of enzymes, especially those associated with synthesis of ribonucleic acid [52]. Zinc help in male and female fertility by protecting the prostate gland from infection, maintaining sperm count and mobility and normal levels of serum testosterone and in treating menstrual problems. It activates areas of the brain that receive and process information from taste and smell sensors. It help to accelerate the renewal of skin cells. Zinc prevents the development of cataract and also prevents night blindness etc. Zinc deficiency can lead to diarrhea and pneumonia in children, impaired cognitive function, memory impairment, reduced taste and smell. Excess zinc in the body can lead to gastrointestinal reactions including nausea, vomiting, cramps and discomfort. High doses of zinc might impair the status of other nutrients especially copper, calcium and iron [53,54].

The phosphorus content of bambara nut was 0.60mg/100g. Olaofe et al., [23] have shown that Luffa cylindrica seeds have phosphorus content 700mg/100g higher than that obtained from bambara nut. The RDA for phosphorus in adult and children is 800mg/day. The value of phosphorus obtained in this study was far below
WHO recommended standard. Phosphorus helps in bone formation and in achieving healthy gums and tooth enamel. It helps in excretion by keeping the kidney in normal condition and maintains proper brain function. Phosphorus ensure that hormones, especially those required for good reproductive health are always in balance form. It contributes in the repair and maintenance of various body cell which are prone to daily wear and tear etc. Phosphorus deficiency can lead to weak bones, discomfort in various body joints, rickets, tooth decay, tremors, anxiety etc. Excess of phosphorus in the body can lead to diarrhea, but high phosphate blood levels allow the phosphorus to bind with calcium causing calcification or hardening to the organ and soft tissues [55].

The ratio of sodium to potassium in the body is of great concern for prevention of high blood pressure. A food source having Na/k ratio of less than value of 1 has impact in lowering blood pressure. The Na/k value of 0.08 was obtained from Vigna subterranea L. thouars. The value indicates that Vigna subterranea L. thouars will not promote high blood pressure because of low Na/k value. This study showed that Ca/P ratio of Vigna subterranea L. thouars was 92.78. If the Ca/P ratio is low (low calcium high Phosphorus in take), more than the normal amount of the calcium may be lost in the urine. Food is considered “good” if Ca/P ratio is above 1 and “poor” if the ratio is less than 0.5, while Ca/P ratio above 2 help to increase the absorption of calcium in the small intestine.

This study revealed that Vigna subterranea L. thouars is a very good food source because of its high Ca/P value. The Ca/Mg ratio for Vigna subterranea L. thouars was 3.53 above the recommended value of 1.00 [56].

4. CONCLUSION

The result of this study showed that Vigna subterranea L. thouars was rich in protein and it could be used as alternative source of protein in diet/protein supplement especially in developing countries such as Nigeria. This study revealed that bambara nut was rich in carbohydrate as well as high available energy. The high Ca/P value indicates that Vigna subterranea L. thouars is a very good food source. The result of the analysis showed that anti-nutrient content of bambara nut were below WHO recommended standard. In view of this, eating bambara nut will not constitute health hazard.

ACKNOWLEDGEMENT

The authors are grateful to Mr. Chijioke Odoh and Mr. L.C. Anyika for taking some measurements of the sample. The authors are also grateful to Department of Agriculture, Federal University of Technology, Owerri for making use of their research laboratory.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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