Working together to eliminate cyanide poisoning, konzo, tropical ataxic neuropathy (TAN) and neurolathyrism.

Cassava Cyanide Diseases & Neurolathyrism Network

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Fatal cassava food poisoning in Eastern Province, Kenya

One of the cassava characteristics is the presence of linamarin, a cyanoglycoside that is hydrolysable under certain conditions to release hydrogen cyanide in a process called cyanogenesis. The hydrolysis is facilitated by the enzyme linamarase and hydroxynitrile lyase (found in cassava leaves). Toxic hydrogen cyanide (HCN) generation takes place when cassava tissues are broken down¹. Cassava is the fourth most important food source worldwide after wheat, rice and maize². Cassava roots have total cyanide content of 1-1550 mg HCN equivalents/kg fresh weight (ppm)³. The maximum safe cyanide level by WHO for cassava flour is 10 ppm.⁴ Two families, a family of 6 from Makueni District and a family of 7 from Kathonzweni District, all from Eastern Province, Kenya were affected after consuming raw and cooked cassava brought from a farmer and relative respectively in August and September 2011. A 4 year old child died in the first family, see Figure, while the other members vomited, general body weakness and some fever. The Health Officer collected the cooked and uncooked cassava and fresh samples from the same plants where cassava was harvested. The cassava tasted bitter as claimed by the family members. The area had experienced drought for the last 3 years. The cyanide content was determined by a modified picrate method.⁵ Chromatographic paper⁶ was used instead of Whatman filter paper⁷. Cyanide determinations were done in triplicate. Fresh samples M1 and M2 were collected from the farm where cassava had been harvested, while sample M3 was raw cassava obtained from the home of the affected family. M4 was boiled cassava which had been eaten by the family. Mean cyanide results (ppm) were M1 53.4, M2 47.8, M3 52.3, M4 46.0. The boiled cassava, sample M4 had lower cyanide content than the uncooked cassava, M3. This is attributed to the fact that boiling of cassava reduced the cyanide content. Samples C1 and C2 were collected from the farm where second family obtained the cassava that caused death to one family member. C1 had cyanide content of 73.2 ppm while C2 was 56.4 ppm. It has been shown that cyanoglycoside levels in cassava root vary widely between cassava cultivars, plants of the same cultivars, different tissues of the same plant, roots of the same plant, and even within the root parenchyma.⁸ The samples also had greatly exceeded the accepted level as recommended by WHO.⁴ This could explain why the levels were fatal to the two children in the two families and caused poisoning of the other family members.
In the remote rural areas of Africa where konzo is common, health service coverage is often low and therefore polio vaccine coverage and disease surveillance may be poor. Where konzo is common and neglected, communities may see no reason to report cases of acute paralysis. The konzo areas may thus pose several challenges for polio eradication: low polio vaccine coverage, poor disease surveillance, and possible confusion in the diagnosis. The following table shows the differences and similarities between the two diseases.

<table>
<thead>
<tr>
<th>konzo</th>
<th>poliomyelitis</th>
</tr>
</thead>
<tbody>
<tr>
<td>diet of bitter cassava</td>
<td>yes</td>
</tr>
<tr>
<td>more than one case in the family</td>
<td>often</td>
</tr>
<tr>
<td>seasonal</td>
<td>yes</td>
</tr>
<tr>
<td>predominant age groups</td>
<td>3-15 years, adult women</td>
</tr>
<tr>
<td>sudden onset</td>
<td>yes</td>
</tr>
<tr>
<td>type of paralysis</td>
<td>spastic</td>
</tr>
<tr>
<td>symmetry of paralysis</td>
<td>symmetrical</td>
</tr>
<tr>
<td>visual disturbances</td>
<td>often</td>
</tr>
<tr>
<td></td>
<td>no</td>
</tr>
</tbody>
</table>

### Recommendations in konzo areas

1. Teach health workers and community leaders the difference between acute flaccid paralysis (suspected polio) and acute spastic paralysis (konzo).
2. Make konzo a reportable disease.
3. Provide physical rehabilitation to all cases of paralysis.
4. Ensure high polio vaccine coverage.

### Recommendations on konzo and neurolathyrism

Konzo and neurolathyrism show very similar clinical symptoms while the apparent dietary determinants of the two diseases are very different: cassava cyanogens for konzo and a neuro-active amino acid in grass pea for neurolathyrism. This similarity in clinical symptoms has been discussed before in this Newsletter and at the Ghent workshop. Could there be any similarity in the molecular etiology with a possible junction of the two pathways? In this short contribution we summarize some facts and discuss some ideas.

From early studies on konzo, it was considered that the presence of cyanogens in cassava was involved in the etiology of this disease, perhaps the low protein intake from cassava roots as staple food in a monotonous diet was an aggravating factor. More specifically, the low content of essential amino acids methionine and cysteine in the diet could reduce the formation of glutathione, the central metabolite in the redox homeostasis, and induce the sensitivity to oxidative stress. In early studies on neurolathyrism, the presence of common vetch seeds (Vicia sativa) containing the toxic β-cyanoalanine found in most samples of grass pea seed was blamed for the toxicity. In 1964 a new non-protein amino acid β-N-oxalyl-γ-diaminopropionic acid (β-ODAP) was discovered in grass pea seeds that was shown to have a specific action on nerve cells, and this discovery started a long story to prove that this was the cause of neurolathyrism. A major problem in the research on both konzo and neurolathyrism was the difficulty to produce an animal model mimicking the human condition of konzo or neurolathyrism. Especially the sudden onset of both diseases is as yet unexplained. Very young animals seemed to be more susceptible to the neuro-excitant β-ODAP.

While many papers on neurolathyrism claimed that grass pea contains high quality protein rich in lysine, it was overlooked that the content of the sulfur amino acids methionine and cysteine was the lowest of all commercial legume crops. Soybeans have twice as much of these essential sulfur containing amino acids, needed for the maintenance of the glutathione level and the redox homeostasis in the cells. Even in soybean the sulfur amino acids are the limiting essential amino acids and farmers feeding soybean to their cattle add synthetic methionine as supplement for better performance. An important similarity between the foodstuffs causing konzo and neurolathyrism is thus the deficiency in sulfur containing amino acids. Only after consumption over an extended period, the sudden onset of konzo or neurolathyrism can occur, which makes it difficult for the victims to understand that a food that helped...
them to survive droughts or even famine, can suddenly become toxic. This gave rise to some popular beliefs that konzo is caused by evil spirits or that neurathyrism is caused by walking through a field of flowering grass pea.

The residual cyanogens present in insufficiently treated cassava roots can be detoxified in the liver by the enzyme rhodanese. Hereby the free cyanide is transformed into the less toxic thiocyanate (SCN−) that can be removed via urine. This detoxification needs one molecule of sulfur amino acid per molecule of cyanide. In case of deficiency of methionine and cysteine, the detoxification is not complete and cyanide or cyanate is released from intracellular stores.

Konzo and neurathyrism share many additional effects in the nervous system. The best known effect of β-ODAP is the excitation of the AMPA-receptors, a subset of glutamate receptors, which may result in excessive activation of post-synaptic neurons. β-ODAP is also known to stimulate the release of glutamate into the synaptic cleft, the space between the pre-synaptic cell and the post-synaptic cell. Normally this glutamate is taken up by glial cells that are the energy suppliers of the neurons, and where glutamate is metabolized into glutamine that is returned to the neuron and recycled. The transporter of glutamate into the neurons or glial cells is however inhibited by β-ODAP which increases the level of glutamate in the synaptic cleft. This in turn increases the excitation of post-synaptic neurons to potentially toxic levels.4 Such excessive or prolonged activation of glutamate receptors gives rise to an increase of intracellular Ca2+ concentration, resulting from entry via Ca2+-permeable glutamate receptors or released from intracellular stores such as mitochondria and the endoplasmatic reticulum. The motor neurons have little buffering capacity for Ca2+, and thus are more vulnerable to such events. β-ODAP also inhibits the cysteine-glutamate antiporter on the membranes of glial cells or neurons which lowers the availability of glutathione precursors and increases oxidative stress.5 Besides being an inhibitor of the cysteine-glutamate antiporter, β-ODAP is also a substrate for this transporter and can enter the cells, where it can affect the mitochondrial respiration. In particular, mitochondrial complex-I seemed to be inhibited.6 The mitochondria are also the target for cyanide toxicity, affecting the electron chain and the formation of ATP. The effect of cyanide or β-ODAP on the mitochondria not only causes depletion of ATP, it also increases the production of reactive oxygen species (ROS). The cell normally can protect itself against these aggressive ROS by a battery of enzymes such as superoxide dismutase, catalase and glutathione peroxidase that act in concert to maintain the redox homeostasis. This can however be jeopardized by the reduced availability of glutathione (GSH). The activity of cyanide or β-ODAP as described above, in addition to the deficiency of sulfur amino acids contribute to this low availability of GSH. This also results in the formation of mixed protein-glutathione disulfides (PrSSG) that normally are reduced by GSH. Dysfunction of mitochondria and oxidative stress have been linked also to neurodegenerations with gradual onset,6 while the central role of glutathione in neuro-degeneration has been discussed by Nunn et al.7 Glutathione seems to be at the point of confluence of the molecular etiology of both konzo and neurathyrism, and this seems to be also the case for neurodegenerations with gradual onset where old age is the main risk factor.8 Konzo and neurathyrism share several aspects of neuro-excitotoxicity: i) mitochondrial dysfunction with reduced energy supply to the neurons, ii) oxidative stress that is enhanced by the deficiency of sulfur amino acids in the staple diet, and iii) the modulation of enzymes protecting against ROS and oxidative stress.4,9 Recently, it is a factor increasing the incidence of neurathyrism symptoms in rats was identified as stress.9 The same authors also found transient hemorrhage in the lower part of the spinal cord, potentially indicating disturbance in the blood-brain barrier (BBB). Occurrence of konzo and neurathyrism is mostly restricted to poor and often illiterate people of low socio-economic status. Social stress in combination with malnutrition and physiological oxidative stress during extended periods may increase the fragility of the BBB and the susceptibility for konzo and neurathyrism. The very high incidence of neurathyrism in a forced labor camp during the second world war10 may corroborate this hypothesis.

References

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Tropical spastic paraparesis (konzo): a major neurologic problem in Caungula - Angola: first report in Angola

A particular kind of spastic paraparesis was first reported in Republic of Zaire about 80 years ago. Since then other reports were made in other tropical countries such as Mozambique, Tanzania and Cameroon, related to cyanide intake of cassava. The typical clinical picture is spastic paraparesis with abnormality of gait while walking or running with onset less than a week and irreversible course. No reports have been made before in Angola. In order to clarify an unusual frequency of unknown disease at the site, we went to Caungula, which is a small rural village in the northeast of Angola near the border with the Democratic Republic of Congo. We observed 20 patients affected with spastic paraparesis with different degrees of gait limitation. In 10 of them we collected blood samples for cyanide, HIV and HTLV virus analyses. The patients were from 5 to 34 years old, mostly female. From those whose blood was collected, all the blood samples were non-reactive to HIV and HTLV and 1 and 2, but all of them had high levels of cyanide.

We conclude that tropical spastic paraparesis (konzo) in Angola is related to consumption of cassava with a high concentration of cyanide.


[Ed: This is the abstract of a poster given at the 20th World Congress of Neurology, Marrakesh, Morocco, (2011). http://wcn.kenesapp.com/WCN_268/poster_16595/poster.aspx . An initial report from the same source was given in CCDN News No 16, 3-4, (2010).] Dr Julie Clifff reports that Dr Miguel Bettencourt stated to the Angolan Press Agency that the region of Caungula continued to report innumerable cases of konzo, and that the disease was a continuing serious public health problem.]

Control of konzo in DRC

Konzo is an irreversible paralysis of the legs that occurs mainly in children and young women after childbirth, due to intake of large amounts of cyanide containing compounds from bitter cassava. It was first discovered by Dr Trolli in 1938 in Popokabaka Health Zone, Bandundu Province, Belgian Congo, now the Democratic Republic of Congo (DRC) and has been brought under control for the first time in the same location. In Kay Kalenge village in Popokabaka Health Zone there were 34 cases of konzo in a population of 1250. The women of the village were taught that konzo is due to a poison present in cassava flour that causes konzo and how to treat the cassava flour to remove the poison by using the wetting method. As follows: The cassava flour was placed in a bowl and a mark made on the inside at the level of the dry flour. Water was mixed into the flour until the wet flour came up to the same mark. The wet flour was spread out on a basket in a thin layer for about 2 hours in the sun and for about 5 hours in the shade. The enzyme (linamarase) present in the flour decomposed the cyanide compound (linamarin) with production of hydrogen cyanide gas. The wetting method is truly a gift from God to these people. The damp flour was then cooked in the traditional way, by mixing with boiling water to produce a thick porridge called fufu, consumed with beans or some other flavouring. The women accepted the wetting method willingly and found that it produced much tastier fufu than that produced from untreated flour, which was bitter due to the presence of bitter linamarin, and the sweet fufu could be stored for longer (2 days) than fufu from untreated flour. The total cyanide content of the cassava flour after the wetting treatment was reduced to less than 10 ppm, the FAO/WHO limit for cassava flour. The mean urinary thiocyanate content of 100 school children (which measures the total cyanide intake over the previous week or so) was reduced from 352 μ mole/L to a safe level of 130 μ mole/L. The number of urinary thiocyanate samples that exceeded a dangerous level of 350 μ mole/L decreased from 26 at the beginning of the intervention (March 2010) to zero by May 2011. No new cases of konzo occurred in Kay Kalenge during the intervention (March 2010 to September 2011) which included two dry seasons when incidence of konzo normally peaks, due to large intakes of total cyanide over the cassava harvest.

The success of this method in preventing konzo is due to (1) education of all women to understand that konzo comes from a poison (cyanide) present in cassava flour and is not due to witchcraft, (2) training in use of the wetting method to remove cyanide compounds, helped by use of laminated posters in the language (http://online.anu.edu.au/BoZo/CCDN N) and (3) full commitment to use of the wetting method on a regular basis. This result was achieved over the period of the intervention by visits every 4 months by the full team, interspersed with visits every month by the Caritas team from Popokabaka. The same methodology is now being used in a one year intervention in three villages where the prevalence of konzo in Boko Health Zone, Bandundu Province, supported by funding from the Australian Agency for International Development (AusAID) and further interventions to prevent konzo in other villages are being planned.

References
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