Onchocerciasis associated epilepsy and nodding syndrome research.

Richard Idro
Makerere University College of Health Sciences, Kampala, Uganda
Objectives

- Describe the clinical, EEG, imaging features and short term outcomes of both NS and OAE in Uganda

- Determine if NS is a neuro-inflammatory disorder with antibodies to O.volvulus cross reacting with host neuron proteins
  - Determine if a similar relationship exists between OAE. May NS and OAE both be the same disease with different manifestations.
  - Can we develop a diagnostic test?

- Can Doxycycline be used for the treatment of nodding syndrome?
Head nodding in an adolescent
Where in the northern Uganda are the patients nodding syndrome found?

GPS locations of patients with epilepsy in Pader, Kitgum and Lamwo districts

GPS locations of patients with NS in Pader, Kitgum and Lamwo districts

Map of Lamwo, Kitgum and Pader showing black fly breeding sites
Natural history of untreated nodding syndrome

- **Stage 1**: Prodromal stage
  - Weeks
  - Increasing
  - Inattention
  - Dizziness
  - Lethargy
  - Staring, sleepy

- **Stage 2**: Head nodding
  - 1 – 3 years
  - Tonic clonic
  - Myoclonic
  - Atypical
  - Absence & focal seizures

- **Stage 3**: Development of other seizures
  - 3 – 8 years
  - Motor
  - Severe cognitive
  - Behaviour & psychiatric
  - Malnutrition, growth failure
  - Muscular skeletal deformities
  - Lip changes

- **Stage 4**: Development of other complications
- **Stage 5**: Severe debilitation

*Idro et al 2013*
Nodding syndrome is a neurological disorder characterized by epilepsy.

Mean number of seizures

No. of seizures 2-3 wks after starting sodium valproate
Principles of Management of Nodding Syndrome in Uganda

- Treatment guidelines developed by a multidisciplinary team of clinicians, nurses, and therapists

- Trained health workers provide care in 17 treatment centres based on the national guidelines

- The goal of treatment is to relieve symptoms, prevent disability and offer rehabilitation to improve function.
  - In the absence of a known cause, care is symptomatic.

- Initial management focuses on the most urgent needs
We assessed the outcomes of treatment one year after starting Nodding syndrome, N=484 and Other convulsive epilepsies, N=476.

<table>
<thead>
<tr>
<th></th>
<th>Nodding syndrome, N=484</th>
<th>Other convulsive epilepsies, N=476</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>Seizure free &gt; 30 days</td>
<td>8 (1.7%)</td>
<td>121 (25.0%)</td>
</tr>
<tr>
<td>Daily clusters of head nods, median (IQR)</td>
<td>4 (IQR 3,6)</td>
<td>1 (IQR 0,2)</td>
</tr>
<tr>
<td>Behaviour and emotional difficulties</td>
<td>327 (67.6%)</td>
<td>133 (27.5%)</td>
</tr>
<tr>
<td>Independence in basic self care</td>
<td>174 (36.0%)</td>
<td>402 (83.1%)</td>
</tr>
</tbody>
</table>
### Nodding Syndrome in Uganda - 2017

<table>
<thead>
<tr>
<th>District</th>
<th>Number of Cases</th>
<th>Deaths</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pader</td>
<td>806</td>
<td>81</td>
</tr>
<tr>
<td>Kitgum</td>
<td>544</td>
<td>33</td>
</tr>
<tr>
<td>Lamwo</td>
<td>339</td>
<td>10</td>
</tr>
<tr>
<td>Gulu and Omoro</td>
<td>323</td>
<td>1</td>
</tr>
<tr>
<td>Amuru</td>
<td>58</td>
<td>4</td>
</tr>
<tr>
<td>Lira</td>
<td>13</td>
<td>-</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>2083</strong></td>
<td><strong>129</strong></td>
</tr>
</tbody>
</table>
What causes nodding syndrome?

- We asked ourselves several questions?
  - Is it caused by a toxin or an environmental chemical in water or food?
  - Is it genetic?
  - Is it caused by an infection?
- We conducted a series of research to answer these questions.
Postmortem examinations

First set of brain specimens – obtained late

Next \( \frac{3}{4} \) specimens – high carbon density material

Similar findings in next set of brain specimens
Fig. 2 Histologic findings in nodding syndrome. a Tau-immunoreactive in frontal cortex, mostly in gryal crowns (AT8, scale bar: 1000 µm). b Cortical neurofibrillary tangles (Bielschowsky stain, scale bar: 100 µm). c Neurofibrillary tangles, dystrophic neurites and dot-like immunoreactivity containing phosphorylated tau in cerebral cortex (AT8, scale bar: 200 µm). d Neurofibrillary tangles and dystrophic neurites in neurons in the base of pons (AT8, scale bar: 75 µm).
The relationship between antibodies to voltage gated potassium channel (VGKC) complex proteins and nodding syndrome

- VGKC are key in generation and propagation of electrical impulses in the CNS.

- We measured serum antibodies against VGKC-complex proteins in 31 patients and 11 sibling controls:
  - 15/31 (48.3%) established cases of NS tested positive for these antibodies compared to:
    - 1/11 (9.1%) controls
CDC measured antibodies to another protein – leiomodin and found an association with NS

- Antibodies against leiomodin-1 were found in 11/19 (58%) cases

- LMOD1 shares 83% sequence similarity with a conserved region of *O. volvulus* tropomyosin.

- The antibodies were neurotoxic in mice brain suggesting cross-reactivity.

- Johnson et al
Pathogenesis & treatment of NS project in Uganda

Aims of the study

1. Examine an immune-mechanism for the pathogenesis of NS
2. Conduct an exploratory trial of doxycycline or placebo as treatment for NS

Work which led to the project

1. Strong epidemiologic association between NS and O. volvulus
2. Two pilot studies showing antibodies to NSPs and O. volvulus
3. No specific treatment. Only symptomatic treatments for NS
4. Trials of doxycycline as treatment for O. volvulus

Experimental design

Case control study
- Of 154 WHO defined NS, 154 epilepsy controls and 154 frequency matched sibling or neighbourhood controls to examine the relationship between NS, antibodies to VGKC/leiomodinin and O. volvulus.

Phase IIb clinical trial
- Randomised trial of doxycycline 100mg daily for six weeks or placebo as treatment for NS in 230 patients.

Expected outcome

- Relationship between NS, antibodies to NSPs and O. volvulus
- Early manifestations of NS, EEG and brain MRI features of NS
- Biomarkers in NS
- Preliminary evidence of Doxycycline as treatment for NS
- Proof of concept that NS is treatable with antibiotics: search for similar
SUMMARY OF STUDY PROCEDURES

Throughout the 24 months, all participants will continue to receive standard of care therapy and follow every 1-2 months as is local practice. Adherence to standard therapy will also be monitored.

Home visits at wks 2, 4, 6 and 3 months
- Adherence monitoring
- Seizure log
- Adverse event documentation

Six month follow up
- Hospitalise for 1 week
- Repeat all pre-randomisation procedures except lumbar puncture
- Measure primary and secondary end points

Follow up at 12 and 24 months
- Measure secondary end points
- Clinical and cognitive assessment
- Seizure burden, complications and disease stage
- Microfilaria density

Tests for the case control study and pre-randomisation procedures for the trial
- Hospitalise for 1 week
- Clinical assessment, seizure burden, complications & disease stage
- Baseline tests – cognition, EEG +/- MRI, blood draw for standard tests, CRP, C3a, C3b, NSPs/leiomodin, skin snips for microfilaria density, lumbar puncture for CSF inflammation – neopterin, oligoclonal bands. Optimise sodium valproate dose

Complete case control study

Recruit subjects for Case Control study

ELIGIBLE PATIENTS
## Current progress

<table>
<thead>
<tr>
<th>Participant Group</th>
<th>No. to be enrolled, N</th>
<th>No. enrolled</th>
<th>% enrolled</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nodding syndrome patients – also randomized to doxycycline or placebo</td>
<td>230</td>
<td>240</td>
<td>&gt;100%</td>
</tr>
<tr>
<td>Non nodding syndrome epilepsy controls</td>
<td>154</td>
<td>154</td>
<td>100%</td>
</tr>
<tr>
<td>Normal community controls</td>
<td>154</td>
<td>154</td>
<td>100%</td>
</tr>
</tbody>
</table>

Recruitment is complete - Results expected beginning of 2020
Preliminary studies

1. Describe the clinical, EEG and corresponding 1.5 Tesla MRI features of NS. ------ 40 patients

2. Clinical, EEG and imaging studies and 2 yr outcomes of OAE epilepsy.

3. Examine the relationship between NS and antibodies to specific NSPs.

4. In cases, investigate for inflammatory markers in CSF and plasma and determine the relationship between these, O. volvulus specific IgG4 and antibodies to NSP-Abs/leiomodin and microfilaria density.

5. Describe types of Wolbachia super-groups in cases with NS and controls and examine the relationship between the super-groups and NS - Germany

6. Conduct an exploratory mass spectrometry study of plasma and CSF to identify potential diagnostic and prognostic biomarkers and explore mechanisms of disease.

7. Genetic studies
Other than research, what is the Ministry of Health doing?

- Plan to eliminate Onchocerciasis
  - Aerial spraying
  - Twice yearly ivermectin to everybody
  - River douching/larvaciding
Proportion of nodding syndrome patients testing positive for *Onchocerca volvulus* microfilaria
Acknowledgments

Funding: ARL Award from MRC/DFID, University of Oxford, Waterloo foundation, Government of Uganda (Ministry of Health and Kitgum District Local Government)