sequence previously obtained from the infected mother of the index case of the Prince of Wales Hospital outbreak (isolate Su-10, GenBank accession number AY282752; table). The table shows that only three nucleotide differences (at positions 3852, 11 493, and 28 696) were noted between patient 1 and the Su-10 sequence, which provides molecular evidence that patient 1 probably acquired the infection from the hospital. Two of these nucleotide changes did not result in aminoacid substitutions. The remaining one (at position 28 696) resulted in a change from cysteine to glycine, but this substitution is unlikely to be related to the distinctive clinical features of the Amoy Gardens outbreak because a glycine at this position is seen for many other SARS-CoV isolates, including the Tor2 and Urbani isolates (table).[4,5]

The viral genomic sequence for patient 2 was identical to that of patient 1, whereas that for patient 3 differed at two nucleotides (positions 17 166 and 28 102; table). To further elucidate the importance and prevalence of these latter changes, we sequenced the five nucleotide positions referred to in the table for viral isolates obtained from naso-pharyngeal aspirates from two additional Amoy Gardens patients (patients 4 and 5; table). Patient 4 had mild diarrhoea and did not require admission to intensive care. Patient 5 had no diarrhoea. Each of these additional viral isolates showed a sequence identical to those from patients 1 and 2, which suggests that the nucleotide alterations at positions 17 166 and 28 102 in the viral isolate from patient 3 were a special case.

The viral isolates obtained from the Amoy Gardens outbreak were typified by that of patient 1. Our findings further support the likelihood of patient 1 being the index of the outbreak. The unique nucleotide differences that distinguish this viral isolate represent the molecular fingerprint of the Amoy Gardens viral isolate. The recurrent demonstration of the presence of this isolate among studied Amoy Gardens patients lends further support to the proposed environmental route of transmission implicated in the outbreak. Sequence variation in the SARS-CoV genome is not responsible for the distinct clinical features of the Amoy Gardens outbreak. Thus, further work should be focused on investigating the possible role of other unique features on this outbreak in explaining the clinical characteristics of this cohort. Our findings highlight the usefulness of molecular investigations in supplementing investigations in clinical and environmental epidemiology.

Contributors

S S C Chim and S K W Tsui contributed equally to this article. S S C Chim, S K W Tsui, K C A Chan, T C C Au, Y K Tong, and E K O Ng did genomic sequencing of viral isolates. E C W Hung and R W K Chiu assessed the correlation between clinical and molecular data. P K S Chan and J S Tam did the virus isolation work. J J Y Sung, C M Chu, and K Y Yuen collected the clinical data. K P Fung, M M Y Waye, and C Y Lee reviewed the sequencing results. Y M D Lo planned the study and interpreted the data.

Members of the CUHK Molecular SARS Research Group

S Chung (Department of Surgery); C Au-Yeang (Department of Microbiology); K K W Au, A H Chan, C W Chan, C Y C Kou, W Y Lam, S K Lau, Y M Lau, S L Law, P T W Law, M L Y Li, A W K Wan, C H Wong, W H Yiu (Department of Biochemistry); N Lee, A Wu, C H Tse (Department of Medicine and Therapeutics); L Y S Chan (Department of Obstetrics and Gynaecology, Chinese University of Hong Kong, Hong Kong Special Administrative Region, China).

Conflict of interest statement

None declared.

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1 Lee N, Hui D, Wu A, et al. A major outbreak of severe acute respiratory syndrome in Hong Kong. *N Engl J Med* 2003; **348:** 1986–94.
2 Outbreak of severe acute respiratory syndrome (SARS) at Amoy Gardens, Kowloon Bay, Hong Kong: main findings of the investigation. http://www.info.gov.uk/info/ap/pdf/amoy_e.pdf (accessed Sept 8, 2003).
3 Peiris JSM, Chu CM, Cheng VCC, et al. Clinical progression and viral load in a community outbreak of coronavirus-associated SARS pneumonia: a prospective study. *Lancet* 2003; **361:** 1767–72.
4 Rota PA, Oberste MS, Monroe SS, et al. Characterization of a novel coronavirus associated with severe acute respiratory syndrome. *Science* 2003; **300:** 1394–99.
5 Marra MA, Jones SJM, Astell CR, et al. The genomic sequence of the SARS-associated coronavirus. *Science* 2003; **300:** 1399–404.

Departments of Chemical Pathology (S S C Chim PhD, K C A Chan MRCP, Y K Tong MPhil, R W K Chiu MB, E K O Ng PhD, Prof Y M D Lo DM)**, Biochemistry** (S K W Tsui PhD, T C C Au MPhil, K P Fung PhD, M M Y Waye PhD, Prof C Y Lee PhD)**, Paediatrics** (E C W Hung MB)**, Microbiology** (P K S Chan MRCPath, J S Tam PhD)**, and Medicine and Therapeutics** (Prof J J Y Sung MD)**, Chinese University of Hong Kong, Prince of Wales Hospital, Hong Kong, China; Department of Medicine, United Christian Hospital, Hong Kong** (C M Chu FRCP)**; and Department of Microbiology, Queen Mary Hospital, University of Hong Kong, Hong Kong** (Prof K Y Yuen MD)

Correspondence to: Prof Y M D Lo, Department of Chemical Pathology, Chinese University of Hong Kong, Room 38023, 1/F Clinical Sciences Building, Prince of Wales Hospital, 30–32 Ngan Shing Street, Shatin, New Territories, Hong Kong Special Administrative Region, China (e-mail: loym@cuhk.edu.hk)

Food-aid cereals to reduce neurolathyrism related to grass-pea preparations during famine

Haileyesus Getahun, Fernand Lambein, Michel Vanhoorne, Patrick Van der Stuyft

Neurolathyrism is a spastic paraparesis that can be caused by excessive consumption of the drought-resistant grass pea *(Lathyrus sativus)*. Devastating neurolathyrism epidemics have occurred during major famine crises in various parts of the world. We investigated in a case-control study the effects of food aid on risk of paralysis. Risk increased with consumption of boiled grass pea (adjusted odds ratio 2·78, 95% CI 1·09–7·13 with cereals; 5·22, 2·01–13·55 without cereal) and raw unripe green grass pea (1·96, 1·16–3·31; p=0·011), but not with the fermented pancake, unleavened bread, and gravy preparations. In a correlational study there was an inverse relation between the number of new cases and the amount of food-aid cereals distributed per person. During famine, cereals and nutritional information should reach people before they have grass pea as the only food.

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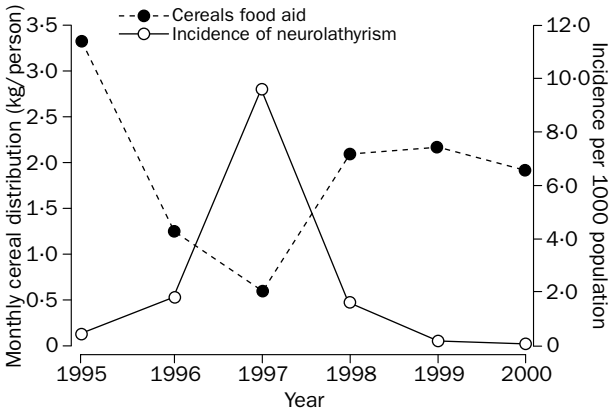
See Commentary page 1775

Neurolathyrism is a neurodegenerative and irreversible spastic paraparesis that can be crippling and lead to complete dependency. This disorder can be caused by excessive consumption of the drought-resistant pulse grass pea (*Lathyrus sativus*). Grass pea contains the glutamate analogue neurotoxin β-N-oxalyl-L-α,β-diaminopropionic acid (β-ODAP), which is thought to cause neuronal damage through excitation of the AMPA-activated receptors.[1] All major famines and chronic food shortages in Ethiopia from the mid-1970s onwards have been accompanied by reports of



Monthly cereal food aid per person and incidence of neurolathyrism in Delanta Dawint, 1995–2000

neurolathyrism epidemics.[1] Of 14 million people presently affected by drought and food shortages,[2] a third are living in neurolathyrism-prone areas.[3] We explored whether addition of food-aid cereals to grass pea foods reduces the risk of neurolathyrism during severe famine.

During the latest epidemic in Ethiopia between 1995 and 1999[4] we established a neurolathyrism surveillance system in Delanta Dawint, one of the most affected districts. We did a correlational study of the amount of food aid that reached the population. We also compared, in a retrospective case-control study, the types of grass pea preparations and cereal mixtures consumed by all people who developed neurolathyrism and by controls in Asim Elana, a severely hit village in this district. Ethics clearance was given by the Ethiopian Science and Technology Commission and by Ghent University. Oral informed consent was obtained from all participants, since more than 50% were illiterate and written consent was impossible.

For the case-control study we identified cases through the surveillance system and randomly selected controls from households in the village. All participants were from the Amhara ethnic group. Information on the proportion and type of cereals added to grass-pea foods was obtained from the female household member who prepared food, through use of a pretested questionnaire marked by trained enumerators. We collected this information for 6 months before the first case was detected and until the end of the epidemic. In addition, we asked these women to show the amount of cereal, if any, generally mixed into each type of grass-pea preparation during the study period. After two demonstrations, the survey enumerator classified the proportion of food-aid cereal to grass pea as at least a third or less than a third.

Spearman's correlation coefficient was calculated to assess the association between the incidence of neurolathyrism and the amount of cereal food aid distributed. We used odds ratios, adjusted for age and sex, in the case-control study to assess the risk of paralysis associated with consumption of particular grass-pea food preparations.

Delanta Dawint district had an estimated population of 165 000 in 2000, and a crude death rate of 17·8 per 1000 population. Between Sept 1, 1995, and Dec 31, 2000, a total of 2035 new cases of neurolathyrism were detected in the district (period prevalence rate 12·3 per 1000). The figure shows the distribution of new cases per 1000 population and the per-person amount of food aid distributed (food-aid values for 2000 are extrapolated). For 1995–99 there was a significant negative correlation (p=0·03) between these two variables. The food aid mainly consisted of wheat (*Triticum aestivum*) and maize (*Zea mays*), with limited supplementary rations of vegetable oil and high-calorie cereal preparations for vulnerable groups. Families received 12·5 kg per person of cereal food aid monthly, up to 62·5 kg per family each month. However, delivery became irregular and delayed and the amount of food aid fell, which coincided with the peak of the epidemic in 1997, when 1454 new cases (9·6 per 1000) were reported.

	Cases (n=170)	Controls (n=818)	Adjusted odds ratio (95% CI)*	p
Variable				
Sex				<0·0001
Male	114 (67·1%)	389 (47·6%)	1·0	
Female	56 (32·9%)	429 (52·4%)	0·39 (0·27–0·58)	
Age-group (years)				<0·0001
0–9	13 (7·6%)	193 (23·6%)	0·69 (0·30–1·60)	
10–19	88 (51·8%)	219 (26·8%)	4·20 (2·19–8·05)	
20–49	56 (32·9%)	269 (32·9%)	2·30 (1·18–4·50)	
⩾50	13 (7·6%)	137 (16·7%)	1·0	
Grass-pea-food consumption				
Green unripe				0·011
No	55 (32·4%)	457 (55·9%)	1·0	
Yes	115 (67·6%)	361 (44·1%)	1·96 (1·16–3·31)	
Roasted				0·46
No	53 (31·2%)	434 (53·1%)	1·0	
Yes	117 (68·8%)	384 (46·9%)	1·44 (0·54–3·85)	
Boiled				0·001
No	52 (30·6%)	457 (55·9%)	1·0	
Yes, with ⩾a third cereal	52 (30·6%)	213 (26·0%)	2·78 (1·09–7·13)	
Yes, with <a third cereal	66 (38·8%)	148 (18·1%)	5·22 (2·01–13·55)	
Bread				<0·0001
No	80 (47·1%)	442 (54·0%)	1·0	
Yes, with ⩾a third cereal	44 (25·9%)	228 (27·9%)	0·31 (0·17–0·56)	
Yes, with <a third cereal	46 (27·1%)	148 (18·1%)	0·79 (0·46–1·34)	
Pancake				<0·0001
No	60 (35·3%)	324 (39·6%)	1·0	
Yes, with ⩾a third cereal	41 (24·1%)	267 (32·6%)	0·28 (0·15–0·54)	
Yes, with <a third cereal	69 (40·6%)	227 (27·8%)	0·59 (0·31–1·11)	
Gravy				0·64
No	2 (1·2%)	8 (1·0%)	1·0	
Yes	168 (98·8%)	810 (99·0%)	1·52 (0·26–8·65)	

*All variables listed entered in one logistic regression model.

Adjusted odds ratio for type of grass-pea foods consumed during neurolathyrism epidemic in Delanta Dawint district

In the case-control study, the consumption of grass pea in roasted, boiled, and raw unripe seed form was associated with an increased risk for neurolathyrism, whereas no raised risk was noted for the fermented pancake, unleavened bread, and gravy preparations (table). Roasted grass pea and unripe green seeds are traditionally consumed on their own, but various condiments are added to the gravy form. Cereals are sometimes mixed with grass pea in the boiled, fermented pancake and unleavened bread forms. Use of cereal and grass-pea flour mixtures for these preparations reduced the risk of paralysis if they contained more than a third cereal.

Susceptibility for neurolathyrism varies among individuals and communities, and an increased risk of paralysis is associated with male sex and young age.[1,3] We controlled for the effects of age and sex in the logistic regression analysis. Furthermore, the participants in our case-control study all belong to the same ethnic and socioeconomic group and share the same culture and tradition. We were, however, unable to perfectly control for socioeconomic variables and interfering acute-illness episodes, and further confounding biases cannot be entirely excluded, particularly in the correlational part of the study.

Although our data might not convey the same impression of accuracy as the information obtained from strictly quantitative 24-h food recall, the observed effects of type of grass-pea preparation are biologically plausible. The condiments added to the gravy form may improve the micronutrient balance, and its complex processing may wash out the water-soluble toxin. Reduced β-ODAP content and the improved aminoacid score after fermentation may explain the lower toxicity associated with the pancake form.[5] The addition of wheat and maize into grass-pea preparations could compensate for the deficiency of methionine and cysteine, as well as diluting the concentration of toxin.

Reports show that only grass pea is resisting the current drought in most neurolathyrism-prone areas and that the population is increasingly relying on this pulse.[3] Food aid should thus not be restricted to the almost starving but should also be urgently sent to people in neurolathyrism-prone areas before they are forced into exclusive grass-pea consumption. Dietary information, education, and communication on safe grass-pea preparations are also needed.

Contributors
H Getahun did the field study. All researchers were involved in the design, analysis, and writing of the study.

Conflict of interest statement
None declared.

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1 Getahun H, Mekonnen A, Teklehaimanot R, Lambein F. Epidemic of neurolathyrism in Ethiopia. *Lancet* 1999; **354:** 306–07.
2 Moszynski P. Ethiopia openly appeals for help with looming famine. *Lancet* 2002; **360:** 1950.
3 US Agency for International Development and Chemonics International Inc. Famine early warning systems network: pre-famine conditions confront parts of Ethiopia. http://www.fews.net/resources/gcontent/gcontent.cfm?submit=y&f=al&g=1000211&d=al&l=en (accessed on July 29, 2003).
4 Getahun H, Lambein F, Vanhoorne M, Van der Stuyft P. Pattern and associated factors of the neurolathyrism epidemic in Ethiopia. *Trop Med Int Health* 2002; **7:** 118–24.
5 Kuo Y-H, Bau H-M, Quemener B, Kahn J, Lambein F. Solid state fermentation of *Lathyrus sativus* seeds using *Aspergillus oryzae* and *Rhizopus oligosporus* sp T-3 to eliminate the neurotoxin β-ODAP without loss of nutritional value. *J Sci Food Agricult* 1995; **69:** 81–89.

South Gonder Health Department, Debre Tabor, Amhara Regional State, Ethiopia (H Getahun MD)**; Public Health Department** (H Getahun, Prof M Vanhoorne MD, Prof P Van der Stuyft MD) **and Laboratory of Physiological Chemistry, Faculty of Medicine and Health Sciences,** (Prof F Lambein PhD)**, and Institute of Plant Biotechnology for Developing Countries (IPBO), Ghent University, Ghent, Belgium** (Prof F Lambein)**; and Epidemiology Unit, Institute of Tropical Medicine, Antwerp, Belgium** (H Getahun, Prof P Van der Stuyft)

Correspondence to: Prof Fernand Lambein, Josef Kluyskensstraat 27, B-9000 Ghent, Belgium
(e-mail: fernand.lambein@UGent.be)