since 42% of the patients required growth factor treatment. FOLFIRINOX compared with gemcitabine was associated with a statistically significant higher incidence of grade 3–4 neutropenia (45.7% *v*. 21%), febrile neutropenia (5.4% *v*. 1.2%), thrombocytopenia (9.1% *v*. 3.6%), diarrhoea (12.7% *v*. 1.8%), and sensory neuropathy (9% *v*. 0%), as well as grade 2 alopecia (11.4% *v*. 1.2%).

Another issue with FOLFIRINOX is that it will be a difficult combination to build on with targeted therapeutics as severe skin rash from erlotinib combined with neutropenia can become life-threatening.

Despite these concerns, FOLFIRINOX represents a new standard of care against which other therapies will need to be compared.

This study was financed by the pharmaceutical company Amgen, the French government and the French National League against Cancer.

SHAILESH V. SHRIKHANDE
Department of Gastrointestinal and Hepato-pancreato-biliary Surgery

SUDEEP GUPTA
VIKAS OSTWAL
Department of Medical Oncology
Tata Memorial Hospital
Mumbai
Maharashtra

Evaluation of newer rapid diagnostic tests for typhoid fever

Keddy KH, Sooka A, Letsoalo ME, Hoyland G, Chaignat CL, Morrissey AB, Crump JA. (Enteric Diseases Reference Unit, National Institute for Communicable Diseases of the National Health Laboratory Service, Sandringham, South Africa; Directorate of Research and Innovation, Tshwane University of Technology, Pretoria, South Africa; National Health Laboratory Service, Nelspruit, South Africa; World Health Organization, Geneva, Switzerland; Department of Medicine, Duke University Medical Center, Durham, United States of America.) Sensitivity and specificity of typhoid fever rapid antibody tests for laboratory diagnosis at two sub-Saharan African sites. *Bull World Health Organ* 2011;**89:**640–7.

SUMMARY

A validity study of newer rapid diagnostic tests for typhoid fever was done in sub-Saharan Africa which has a lower burden of typhoid fever as compared to Asia. Three types of antibody tests were evaluated against blood culture (Bac-T Alert, bioMérieux) as the gold standard. The Widal test (with a cut-off of ≥1:80 for positivity) was done using Linear Cromotest® (Linear Chemicals) using two techniques—semiquantitative slide agglutination test and single tube agglutination test, testing separately for the H and O antigens. The other two tests were TUBEX® and Typhidot®. Two different types of the Typhidot® test were used—one testing for IgG and the other for IgM. Thus, 3 tests using 7 techniques were compared with the gold standard. The study was conducted at 2 hospitals, one in Mpumalanga province, South Africa and the other in Moshi, Tanzania. Consecutive febrile patients with a history of fever or documented fever >38 °C were enrolled. A disease-positive patient was defined as one whose blood culture grew *Salmonella typhi* and a disease-negative patient as one with no growth or growth of other organisms in the culture. Trained laboratory personnel did the tests under blinded conditions. Blood culture was done as soon as the patient was admitted whereas the sera for antibody testing were stored and tests were done at an interval of 6 months to 2 years after collection. The total sample size was 92 (53 in South Africa and 39 in Tanzania). *Salmonella typhi* was isolated in 28 samples. The sensitivity, specificity and predictive values were calculated for each of the 7 techniques. Further, predictive values were calculated for 2 hypothetical scenarios of low (5%) and high (50%) pre-test probabilities because the sample prevalence of 30% was not considered reflective of field conditions. The 5% figure was taken to represent the lower burden of typhoid fever in this part of Africa and the 50% figure was taken to simulate an outbreak situation.

The most sensitive test was Cromotest® semiquantitative slide agglutination O test with a sensitivity of 95.2% (95% CI 86.5–99.0) and the most specific was Typhidot® IgG with a specificity of 70.4% (95% CI 49.8–86.2).

The semiquantitative slide agglutination and single tube Widal tests had positive predictive values (PPVs) of 25% (95% CI 0.6–80.6) and 20% (95% CI 2.5–55.6), respectively. The newer tests had comparable PPVs with that of TUBEX® being 54.1% (95% CI 36.9–70.5); of Typhidot® IgM 56.7% (95% CI 37.4–74.5); and of Typhidot® IgG 54.3% (95% CI 36.6–71.2).

For a pre-test probability of 5%, PPVs were 11% (95% CI 6.6–17.9) for TUBEX®, 9.1% (95% CI 5.8–14.0) for Typhidot® IgM and 11% (95% CI 6.3–18.4) for Typhidot® IgG. For a pre-test probability of 50%, PPVs were 70.2% (95% CI 57.3–80.5) for TUBEX®, 65.6% (95% CI 54.0–75.6) for Typhidot® IgM and 70% (95% CI 56.0–81.1) for Typhidot® IgG.

The authors concluded that the newer rapid antibody tests performed poorly and cannot be used in routine settings but they can be used judiciously in outbreak situations when pre-test probability would be high.

COMMENT

The diagnosis of typhoid has been based on 2 major tests. The first is blood culture. This is not widely used because of the need for trained laboratory personnel and expensive equipment. The second is the Widal test, one of the most widely used tests for diagnosis of typhoid in India. It has been shown in several studies that Widal test lacks sensitivity resulting in high false-negative rates.[1–5] The need for better and rapidly performed diagnostic tests led to the introduction of tests such as TUBEX® and Typhidot® based on antibody detection. The results of the present study need to be interpreted in the light of certain limitations in the methods used.

The inclusion criteria did not include the duration of illness, which is an important component. Based on the WHO case definition of a person with suspected typhoid fever, many studies

have included patients with fever for at least 3 days.[6–8] No exclusion criterion has been mentioned by the authors. Individuals previously vaccinated against typhoid and those who have previously had typhoid should have been excluded as current antibody response is modified due to an anamnestic reaction.

The setting in which the patients were recruited, whether outpatient or inpatient, is not clearly stated. The severity of illness has also not been mentioned. This may have introduced 'spectrum bias', if either only severely ill or mildly ill patients were studied.

The average duration of illness of the participants has not been specified. If most patients were seen during the first week of illness, antibody response would have just begun and the index tests would under-perform. Later in the course of the illness, antibody response would be greater and the index tests would perform better. It is prudent to do a week-wise analysis of performance of the index test as shown by Olsen *et al*.[4]

On calculating the sensitivity and specificity of each test, it seems that the authors have, possibly inadvertently, switched the columns for these two parameters in the second table of the article. The discussion too seems to be based on these mislabelled numbers.

After correctly assigning the values for sensitivity and specificity, it can be seen that sensitivity ranged from 3.6% for Cromotest® semiquantitative slide agglutination O test to 70.4% for Typhidot® IgG test, and specificity ranged from 69.2% for Typhidot® IgG test to 95.2% for both Cromotest® semiquantitative slide agglutination O test and Cromotest® tube agglutination H test.

Conversely, if the values in the table and the accompanying discussion are correct, then the values given under the heading 'estimates' in the results could be wrong.

The diagnoses in patients who did not have typhoid have not been reported. This is important to know the differential diagnoses that the index test was able to discriminate typhoid from.[9] We need a test that can differentiate typhoid from commonly confused febrile conditions such as malaria, pneumonia, etc.

The sample size was inadequate to make any statistical inference about the results (reflected in the wide confidence intervals). Sample size calculation performed as suggested by the Tropical Diseases Research (TDR) Diagnostics Evaluation Expert Panel report,[10] for a sensitivity of 80% and absolute precision of 10% gave the number of 'truly disease-positive' persons to be recruited as 170. If we assume that 30% (the sample prevalence achieved in this study) of all patients meeting the inclusion criteria for suspected typhoid will be true positives, then the total number of suspected cases to be recruited will be 566. The authors recruited only 92 suspected cases of which 28 had a positive blood culture for *Salmonella typhi*. Simple normograms based on required levels of sensitivity and specificity for such calculations are described by Carley *et al*.[11]

The strength of the study was that 3 different kinds of tests were subjected to a head-to-head comparison in a low prevalence field situation. Such a situation provides a good opportunity to study cost which would be crucial for policy recommendations. However, an economic analysis was not done in this study.

Although certain other studies have reached similar conclusions, this study lacks sufficient methodological rigour to support its results.[8,9,12]

Relevance to India

Typhoid, an endemic disease in India, is a disease of improper sanitation and hygiene. Its geographical distribution is heterogeneous with urban slums having a high prevalence.[13] Many studies in India have evaluated the validity of Typhidot® and TUBEX® tests. Three hospital-based studies done in Mysore, Vellore and Delhi, with sample sizes ranging from 80 to 563, have shown that Typhidot® test has a very high sensitivity, can be used in early diagnosis and is easy to do.[7,14,15] However, a population-based study conducted in urban slums of Kolkata reported that Typhidot® and TUBEX® tests performed poorly and were comparable to the Widal test.[8]

An Indian Academy of Pediatrics (IAP) Task Force Report in 2006 recommended that in children blood culture is the diagnostic method of choice and the Widal test can be used with caution in the second week of illness. There were no recommendations on the use of newer rapid tests.[16] The Integrated Disease Surveillance Project (IDSP) Manual for District Public Health Laboratories has a provision for reporting cases of typhoid based on Typhidot®, in addition to blood culture and Widal test.[17]

There is a need to develop guidelines on the use of newer diagnostic tests. The critical aspects that should be addressed in this process are: (i) endemicity of the disease, (ii) setting in which the diagnosis is required (hospital or primary healthcare as the pre-test probability and resources will vary), (iii) duration of illness (first week or later, when a certain type of test may be more useful than another), (iv) prior use of antibiotics, and (v) cost, feasibility and field robustness of different tests. Operational research to ascertain the feasibility of using different types of tests for different stages of illness and different settings is also required because every type of test has its own advantage in certain stages of the illness.[18]

In addition to treating typhoid early, it is important to prevent the emergence of antibiotic resistance which is becoming increasingly common due to incorrect diagnostic test results, unnecessary delay in diagnosis and consequent inappropriate use of antibiotics.[19,20]

REFERENCES

1 Shukla S, Patel B, Chitnis DS. 100 years of Widal test and its reappraisal in an endemic area. *Indian J Med Res* 1997;**105:**53–7.
2 Olopoenia LA, King AL. Widal agglutination test—100 years later: Still plagued by controversy. *Postgrad Med J* 2000;**76:**80–4.
3 Bakr Wafaa M, El Attar LA, Ashour MS, El Toukhy AM. The dilemma of Widal test—which brand to use? A study of four different widal brands: A cross sectional comparative study. *Ann Clin Microbiol Antimicrobials* 2011;**10:**7.
4 Olsen SJ, Pruckler J, Bibb W, Nguyen TM, Tran MT, Nguyen TM, *et al*. Evaluation of rapid diagnostic tests for typhoid fever. *J Clin Microbiol* 2004;**42:**1885–9.
5 Clegg A, Passey M, Omena M, Karigifa K, Suve N. Re-evaluation of the Widal agglutination test in response to the changing pattern of typhoid fever in the highlands of Papua New Guinea. *Acta Trop* 1994;**57:**255–63.
6 World Health Organization. *WHO background document: The diagnosis, treatment, and prevention of typhoid fever*. WHO, 2003. Available at *http://whqlibdoc.who.int/hq/2003/WHO_V&B_03.07.pdf* (accessed on 14 Oct 2011).
7 Narayanappa D, Sripathi R, Jagdishkumar K, Rajani HS. Comparative study of dot enzyme immunoassay (Typhidot-M) and Widal test in the diagnosis of typhoid fever. *Indian Pediatr* 2010;**47:**331–3.
8 Dutta S, Sur D, Manna B, Sen B, Deb AK, Deen JL, *et al.* Evaluation of new-generation serologic tests for the diagnosis of typhoid fever: Data from a community-based surveillance in Calcutta, India. *Diagn Microbiol Infect Dis* 2006;**56:**359–65.
9 Bhutta ZA, Mansurali N. Rapid serologic diagnosis of pediatric typhoid fever in an endemic area: A prospective comparative evaluation of two dot-enzyme immunoassays and the Widal test. *Am J Trop Med Hyg* 1999;**61:**654–7.
10 Banoo S, Bell D, Bossuyt P, Herring A, Mabey D, Poole F, *et al.* Evaluation of diagnostic tests for infectious diseases: General principles. *Nat Rev Microbiol* 2006;**4** (12 Suppl): S16-S28 doi:10.1038/nrmicro1523.
11 Carley S, Dosman S, Jones SR, Harrison M. Simple nomograms to calculate sample size in diagnostic studies. *Emerg Med J* 2005;**22:**180–1.
12 Membrebe FA, Chua JA. The clinical utility of typhidot in the diagnosis of typhoid fever. *Phil J Microbiol Infect Dis* 1999;**28:**1–4.
13 Ochiai RL, Acosta CJ, Danovaro-Holliday MC, Dong Baiqing D, Bhattacharya SK, Agtini MD, *et al.* A study of typhoid fever in five Asian countries: Disease burden and implications for controls. *Bull World Health Organ* 2008;**86:**260–8.

14 Jesudason MV, Sivakumar S. Prospective evaluation of a rapid diagnostic test Typhidot for typhoid fever. *Indian J Med Res* 2006;**123:**513–16.
15 Sherwal BL, Dhamija RK, Randhawa VS, Jais M, Kaintura A, Kumar M. A comparative study of Typhidot and Widal test in patients of typhoid fever. *J Indian Acad Clin Med* 2004;**5:**244–6.
16 Kundu R, Ganguly N, Ghosh TK, Yewale VN, Shah RC, Shah NK. IAP Task Force Report: Diagnosis of enteric fever in children. *Indian Pediatr* 2006;**43:**875–83.
17 Manual for laboratory diagnosis of common epidemic prone diseases for district public health laboratories, Integrated Disease Surveillance Project. Ministry of Health and Family Welfare, Government of India, IDSP, 2011. Available at *http://idsp.nic.in/idsp/IDSP/Lab_Manual_260511.pdf* (accessed on 25 Oct 2011).
18 House D, Chinh NT, Diep TS, Parry CM, Wain J, Dougan G, *et al.* Use of paired serum samples for serodiagnosis of typhoid fever. *J Clin Microbiol* 2005;**43:**4889–90.
19 Menezes GA, Harish BN, Khan MA, Goessens WH, Hays JP. Antimicrobial resistance trends in blood culture positive *Salmonella typhi* isolates from Pondicherry, India, 2005–2009. *Clin Microbiol Infect* 2011 Apr 25. doi: 10.1111/j.1469-0691.2011.03546.x.
20 Zaki SA, Karande S. Multidrug-resistant typhoid fever: A review. *J Infect Dev Ctries* 2011;**5:**324–37.

RIZWAN S. A.
BARIDALYNE NONGKYNRIH
Centre for Community Medicine
All India Institute of Medical Sciences
New Delhi
sarizwan1986@gmail.com

Timing initiation of parenteral nutrition: Does it impact the outcome?

Casear MP, Mesotten D, Hermans G, Wouters PJ, Schetz M, Meyfroidt G, Van Cromphaut S, Ingels C, Meersseman P, Muller J, Vlasselaers D, Debaveye Y, Desmet L, Dubois J, Van Assche A, Vanderheyden S, Wilmer A, Van den Berghe G. (Department of Intensive Care Medicine and Medical Intensive Care Unit, Department of Internal Medicine, University Hospitals of the Catholic University of Leuven, Leuven; Department of Anaesthesia and Intensive Care, Jessa Hospitals, Hasselt, Belgium.) Early versus late parenteral nutrition in critically ill adults. *N Engl J Med* 2011;**365:**506–17.

SUMMARY

Guidelines from Europe[1] and the USA[2] recommend that enteral nutrition (EN) should be commenced within 24–48 hours of admission to the intensive care unit (ICU) for patients who are not expected to be on a full oral diet within 3 days. This recommendation is supported by a recent meta-analysis, which showed that early EN (within 24 hours) compared with standard care was associated with a significant reduction in mortality and rate of pneumonia.[3] However, caloric goals are often difficult to attain with EN in critically ill patients, resulting in underfeeding and a number of associated risks.

The ESPEN (European Society for Parenteral and Enteral Nutrition) guidelines[1] advocate that supplemental parenteral nutrition (PN) should be considered whenever EN is insufficient to meet the caloric targets after 2 days.[4] This recommendation is based mainly on a meta-analysis, which showed a significant benefit in favour of early PN among patients in whom EN could not be initiated within 24 hours of admission to an ICU.[5] In contrast, the ASPEN (American Society of Parenteral and Enteral Nutrition) guidelines[2] recommend that, in the absence of malnutrition, PN should be initiated only after the first week of admission to an ICU, if EN is not tolerated despite strategies to maximize delivery of EN. This recommendation is based on 2 meta-analyses[6,7] of studies undertaken before the mid-1990s that produced contradictory results.

The EPaNIC study was a prospective, randomized, controlled, parallel-group, open-label, multicentre clinical trial comparing early (within 24–48 hours) versus late initiation (day 8) of PN when EN on its own failed to reach the calculated caloric goal.

From 1 August 2007 to 8 November 2010, all adults who were admitted to 1 of 7 participating ICUs in Belgium were screened for inclusion in the study, if they had upon admission to ICU, a nutritional risk screening (NRS) score of 3 or more, indicating a risk of developing malnutrition. The exclusion criteria were do-not-resuscitate (DNR) code at the time of admission to ICU, death expected within 12 hours, home ventilation, diabetic coma, BMI <17 kg/m^2, short bowel syndrome, pregnancy or breastfeeding, enrolment in another trial, oral nutrition or established nutritional therapy on admission, and absent clinical indication for central intravenous catheterization.

After informed consent was obtained, the patients were stratified according to diagnostic categories and randomly assigned in a 1:1 ratio to one of the two study groups, labelled 'early-initiation PN' (top-up of EN) or 'late-initiation PN' (top-up of EN) using initially sequentially numbered, sealed and opaque envelopes that were later replaced by a central computerized randomization system. Treatment assignments were made in permuted blocks of 10 per stratum.

All patients who were unable to eat on the second evening of ICU stay (<36 hours) and were without formal contraindications, received EN while being nursed in a semi-recumbent position. The infusion rate for EN and the use of prokinetic agents and duodenal feeding tubes were specified in the standing orders. Trace elements, minerals and vitamins were administered in all patients as clinically indicated irrespective of the group they were assigned to.

Patients who were assigned to the early-initiation PN (top-up of EN) group received intravenous 20% glucose solution; the target for total energy intake was 400 kcal/day on ICU day 1 and 800 kcal/day on day 2. On day 3, PN with OliClinomel or Clinimix (Baxter, Brussels) was initiated at a rate matching the gap between actual energy intake delivered by EN and the calculated caloric goal based on corrected ideal body weight (IBW), age and sex. PN was continued until EN alone constituted at least 80% of the calculated caloric goal. In contrast, patients who were assigned to the late-initiation PN (top-up of EN) group received 5% glucose solution equal in volume to PN the patient would receive in the early-initiation group. If EN was insufficient after 7 days, PN was initiated on day 8 to reach the caloric goal.

The primary outcome measure of this study was the length of stay in ICU. This was defined as the time when the patient was no longer in need for vital-organ support and met at least two-thirds of the caloric requirements as oral feeding.

Over a 39-month period, 4640 patients were randomized and included in an intention-to-treat analysis with minimal protocol violations and loss to follow up. The two groups were well-matched at entry. The patient population was fairly representative for a northern European ICU with a mean (SD) age of 64 (15) years and a dominance (>60%) of the male gender. The study population had a mean (SD) APACHE II score of 23 (10) at admission to the ICU. The