Dear Readers,

Greetings from the Research and Training Domain of TLMTI!

You will be glad to know that TLMTI is developing a training resource for capacity building in all aspects of Leprosy, and from this year all news and updates on Training activities will be brought to you along with ‘Research’ news through this Newsletter.

TLMTI has been engaged in providing comprehensive training in all aspects of leprosy for many years, through its recognised training centres at Naini, Salur, Miraj, Kothara, Shahdara and Purulia. Currently TLM conducts the majority of training in TLMTI through its dedicated Training Unit at Naini. It is gradually becoming a serious concern throughout the ‘leprosy world’ that leprosy related expertise is on the wane, and it will be challenge to keep up the diagnostic and management skills in leprosy as cases become fewer in number. So TLMTI is developing a Training resource, which will provide capacity building in leprosy and all related areas such as Disability Management, Community Mobilisation, Rehabilitation, advocacy etc. Dr. Suresh Verghese, with many years of experience in various field of leprosy, is the Training Coordinator for TLMTI. We are attaching the Training calendar of TLM Community Hospital Naini, for any who are interested in this information.

In this issue, Dr. Jerry Joshua (Head, Health Programmes) has shared his ideas on a much needed change of focus as we care for insensitive feet. There are also the abstracts of some of the papers published by our staff.

On the Research Front, things are moving forward, with papers published in different peer reviewed journals last year and a number awaiting publication this year. We have completed 3 large multicentric field projects and four are ongoing. These projects are on such diverse topics as ENLs, Drug Resistance, methods for early detection. Funds are awaited for several approved projects.

Recently, on 11th and 12th April, the Mid Term Conference of the Indian Association of Leprologists was held at Hyderabad. It was a good meeting where urgent issues such as early detection, prophylaxis, and drug resistance were discussed. The high point of the Conference for TLM was when Dr. Utpal Sengupta, Consultant for SBL was awarded a Lifetime Achievement Award in recognition of his contribution to leprosy work. We are fortunate to have him to guide our young scientists at SBL and give the benefit of his experience. And two TLMTI staff, Dr. Itu Singh from SBL and Mr. Pankaj Gupta from TLMTI Shahdara did us proud by tying for the Best Paper Award.

Please do write in to us with any news, cases, articles or Feedback We would be very happy to hear from you. This issue has been a little delayed due to unavoidable circumstances, but we will endeavour to make it up with the interesting contents.

Happy reading!

Annamma S. John
Editor & Head (Research & Training)
Whenever we discuss the most common residual problem because of leprosy, the answer is ‘ulcer’. Unfortunately, ulcers are the common problem that people come to us with as a problem requiring our attention. We too begin to address the problem of healing the ulcer and rarely look beyond at the larger problem of the slowly disintegrating limb.

Non-healing and recurrence of plantar ulcers.

Classical teaching tells us that healing goes through three stages. The first stage of healing is inflammation (Inflammatory phase). This lasts 2 to 3 days. The second stage of wound healing is granulation and epithelialisation. (The Proliferative phase). This starts on the 2nd day or 3rd day and carries on till the end of the 3rd month. The third stage of wound healing is the stage of fibrosis. (Remodelling or maturing phase). This starts in the later part of the first week and peaks around the 2nd and 3rd month and continues up to the end of a year.

In many cases the ulcers, if they have healthy granulating floor and edges and if the edges are epithelialising, heal with ‘rest’. If the edges of the ulcer and or the floor of the ulcer are fibrosed and the base of the ulcer is also fibrosed, such an ulcer does not heal. This is because the third stage of healing (which is fibrosis) has taken over before the second stage (the stage of granulation and epithelialisation) has consolidated into the area of the ulcer. Vascularity of such an ulcer is poor as vessel growth through fibrous tissue is difficult. These ulcers have thickened, fibrous and hyperkeratotic edges which retard the healing process. This happens because of repeated irritation of the site of the ulcer during the second stage of wound healing. (Granulation and epithelialisation). This repeated irritation can be because of repeated weight bearing and shearing stresses on the wound. This wound would have healed with ‘rest’ when it was in the second stage of healing. ‘Rest’ alone will not work if the wound healing has already entered the third stage without bridging the gap between the edges.

‘Rest’ in the early stages of wound healing, promotes complete healing. If this rest wasn’t timely, then in the later stages of wound healing, rest alone will not suffice as the fibrosis serves as an impediment to vascularisation and epithelialisation. The fibrous tissue needs to be excised and replaced with living tissue. If this is not done, it remains as a chronic or non-healing ulcer.

Further, the thin epithelium that covers over the fibrous tissue, results in an unstable scar, especially over weight bearing areas and over tendons and moving tissue. This is liable to ulceration because, the scar is tethered and interrupts free movement of the surrounding tissue. As the tethered scar interrupts or resists the free soft tissue movement, it tears, giving way, to permit the movement. This is another reason for the need for soft tissue replacement of the scar. Scars subject to chronic irritation subsequently undergo metaplasia (becoming abnormal cells) and later anaplasia (malignant change).

Mechanisms and causes of ulcers

(i) Continuous pressure: If any part of the sole is deprived of its blood supply for long, it will die. Standing without shifting weight from foot to foot over time can cause the compressed tissue of the sole to die, undergo necrosis and ulcerate. This is brought on specially if one foot is anaesthetic and the other isn’t. The person continues to bear weight on the anaesthetic foot without shifting weight from foot to foot. The weight bearing points become necrotic over time and ulcerate. So low pressure over time causes ischemic necrosis and ulceration.

(ii) High pressure – This could be brought about by injury on a nail or thorn or even standing on a corn or callus in the foot. This results in mechanical destruction causing the skin to split and forming an ulcer

(iii) Repetitive pressure: such as that produced by walking long distances can cause ulcer by inflammation. The part being repetitively injured by low pressure becomes swollen, red and warm. Shearing stresses (friction) also play a part in stimulating this
inflammation.

(iv) Moderate pressure and infection: Standing and walking with an open ulcer (broken skin) on the foot results in spreading the sepsis picked up by an open ulcer from the external environment to surrounding tissue.

‘The Hot Foot’

Leaving aside ulcers, we need to address the causes of disintegration of the extremity. That is the crumbling of the tarsal and metatarsal bones of the foot and carpal and metacarpal bones of the hand. This is caused by forces acting (i) during heel strike, (ii) during the foot flat or midstance position and (iii) on the propulsive or the push off phase of the gait cycle.

(i) Damage during heel strike: The calcaneum (the heel bone) may be cracked on landing heavily down on the heel while walking, running or jumping. Those with normal sensation feel the pain when the bone cracks and from then on avoid bearing weight on the heel. Those with insensitive feet continue to use the heel for weight bearing and damage the calcaneum further.

(ii) Damage during the midstance: During this phase of the walking cycle, the weight of the body is shared by the heel, the lateral side of the foot and the metatarsal heads. (The front, the lateral side and the forefoot.) There is no danger of ulceration or disintegration except if the foot were supinated (inverted). In this case the lateral border of the foot takes excessive weight and may cave in at the calcaneo cuboid junction or at the base of the 5th metatarsal head. This results in ulceration and possible disintegration due to sepsis. This needs immobilisation until wound healing, a wedge excision of the bones at the base of the healed scar and later a flap cover after excision of the scar.

(iii) Damage during the propulsive phase: During this phase there are forces of the gastrocnemius and soleus muscles acting to pull the heel up and the ground forces acting to push the forefoot up against the body weight acting on the foot, pushing the foot down. Against these forces, the least protected part of the foot between the heel and the forefoot is the top of the arch, the navicula bone. (The head of the talus or the cuneiform bones or the base of the 1st metatarsal may also be the weakest point).

This part of the arch could often crumble and disintegrate resulting in the collapse and reversal of the arch. This often happens when the intrinsic muscles supporting the arch are weakened by paralysis or if the cables of the tibialis anterior and or tibialis posterior are weakened or paralysed. This can be brought about by walking fast, walking for long, running, jumping or stumbling. This is aggravated by continuing to subject the foot to the stresses of standing, squatting, walking or running.

This crumbling need not necessarily be a fracture, but a giving way of ligaments of the joints of the foot. This too can result in foot disintegration.

Again, in the early phases of foot disintegration, rest is the first requirement with immobilisation of the foot in the anatomical position. In later stages of disintegration, surgical correction of this deformity is necessary. In still later stages amputation may be needed.

The patient usually presents with a swollen foot. On palpation, the examiner notices warmth. There is no pain or tenderness because the limb is anaesthetic. We therefore refer to such a foot as a ‘hot foot’. It is hot because of inflammation due to a fracture or a sprain. The hot foot needs immobilisation for 6 months in a cast. We usually immobilise it in a cast for one month. Then we see that the cast has become loose because the swelling has subsided. So we remove the cast and reapply it. We wait for 4 months, then remove the cast and take a mould for a Fixed ankle brace. We reapply the plaster for a moth and then remove it and fit the Fixed ankle brace. If we do not fit the fixed ankle brace and allow the patient to walk normally, the bones which are by now osteoporotic and brittle will fracture again. The fixed ankle brace should necessarily be worn for a year till the bones regain their calcium.

So we can see that just paying attention to ulcers
alone will not save limbs. We need to pay attention to the mechanism by which these ulcers occur. We need to pay attention to the mechanisms by which limbs disintegrate without ulcerations. We need to educate patients on care of their anaesthetic limbs.

**EVENTS**

**TLMTI Research Committee**

Held on 25th February, 2015 at the conference hall of ICMR. It was the final meeting at ICMR as Dr. Katoch, the Chairman retired soon after that, though he continues to head the Committee.

**TLMTI Ethics Committee**

Held on 17th March, 2015.

**MLCU WORKSHOP ON TEACHING-LEARNING**

*February 24 - 26, 2015*

TLMTI is in the process of affiliating on courses conducted by the Martin Luther Christian University, Shillong. As part of this process a "Teaching-Learning Workshop" was conducted by MLCU faculty at the Media Centre, Noida. The facilitators were Dr. S Maxwell Lyngdoh (Director, Affiliate Education), Dr. Melari S Nongrum (Deputy Director, University Research Cell) and Dr Maribon V Sangma (Head, Department of Counselling Psychology).

The objectives of the workshop were:

- To evaluate multiple intelligences, learning styles and personality and relate these to teaching-learning of students
- To design a structured approach to interactive learning and assessment
- To become familiar with the academic concepts, policies and rules of MLCU

These topics were dealt with in a participatory and innovative way, which the participants enjoyed. The Workshop introduced a number of new concepts and promoted a lot of independent thinking and exploration of new areas related to training. All in all, it was a great experience for the 23 participants, who are looking forward to using these ideas in their training courses. We look forward to a mutually fruitful and congenial partnership with MLCU.

**Multiple Intelligences, Philosophies of MLCU**

- Complementary curriculum
- Experiential learning
- Student-Centered Learning and Assessment
- CFE, GPA, marks
- CFE plan, Application of Blooms Taxonomy
Stanley Browne Laboratory (SBL) is at present engaged in conducting a field based epidemiological study to investigate the route of Mycobacterium leprae transmission in leprosy affected families and the surrounding community, especially in children in endemic regions of Purulia and Champa Districts of West Bengal and Chhattisgarh respectively.

Dr. Utpal Sengupta and Dr. Ravindra P. Turankar of SBL, along with their research team visited the houses of multi-case families of leprosy patients in the villages of Champa and collected samples (slit-skin-scrapings, nasal swabs, saliva and stool) from patients and their contacts including children. The patients and contacts will be followed up for five years and if they develop disease during the follow up period then another sample will be taken from these contacts along with the patients who were already treated during the study.

In addition to this, samples were also collected from their surrounding environment (soil and water) to find out the source of M. leprae in the natural environment where the patients are residing. As the objective of the study is to determine whether M. leprae isolated from nature has the capability of transmitting infection, attempts will be made to infect experimental mice with such M. leprae isolates. Earlier, SBL already showed that M. leprae from such isolates are viable, by demonstration of mRNA of M. leprae from soil and water samples collected from areas inhabited by leprosy patients.

Further, data are being obtained to find out the nature of contact of the children with their source cases. It has been demonstrated by various workers that helminthic infestations lowers the level of protective immunity (Cell Mediated Immunity) towards various diseases like Tuberculosis, AIDS and Leprosy. Therefore, stool samples are being examined from patients and contacts to find out the background of helminthic infection in the community.

Early signals of infection are being studied through analysis of antibody in saliva and presence of M. Leprae in nasal swabs. Permission was obtained from the Ministry of Health, Government of India to carry out the above activities in the field for research purposes to understand the mechanism transmission. Full co-operation was provided by TLM Purulia and TLM Champa.

The senior persons of the household were approached for participation and after discussing and explaining the details of the project, excellent co-operation from the community was available. The consent forms and epidemiological questionnaires were filled with full co-operation of the participants. Villages in Champa District are scattered along the highway and hence travel of 100km/day by motorcycle was made for 4 days with the help of the Field Technician, Mr. Pratap Singh of Champa.

The Medical Superintendent and staff, especially Mr Sentil Kumar, Physiotherapist of The Leprosy Mission, Champa were a great source of help and inspiration for execution of field activities smoothly. The outcome of the field study might be of great relevance to understand the nature of transmission and to plan and adopt strategies for the control and transmission of leprosy under the National programme.

Dr. Ravindra P Turankar
Research Officer, Stanley Browne Lab

(Photos: Research Staff collecting samples in Champa District of Chhattisgarh)
ABSTRACTS

Report of rpoB mutation in clinically suspected cases of drug resistant leprosy: A study from Eastern India

Mallika Lavania, Utpal Sengupta et al
2015;81(2):155-161

Background: The current strategy for leprosy control depends mainly on early case detection and providing the recommended multidrug therapy (MDT) dosage. Understanding the molecular mechanisms of drug resistance to each of these drugs is essential in providing effective treatment and preventing the spread of resistant strains in the community. The progress of molecular biology research provides a very efficient opportunity for the diagnosis of drug resistance by in vitro method.

Aim: We aimed to investigate the point mutations within the rpoB gene region of the Mycobacterium leprae genome, which are responsible for resistance to rifampicin, in order to determine the emergence of drug resistance in leprosy in the Kolkata region of West Bengal.

Methods: A total of 50 patients with a relapse of leprosy were enrolled in the study. Skin smears were obtained for estimation of bacillary index and biopsies were obtained in 70% alcohol for extraction of DNA. The extracted DNA was amplified by M. leprae-polymerase chain reaction (PCR) targeting rpoB gene region. Every single nucleotide base in the sequence is aligned to reference sequence and identity gaps were determined by NCBI - BLAST. Later in-silico analysis was done to identify the changes in the translated protein sequences.

Results: A mutation at the base pair position 2275405 where G is replaced by C in the M. leprae genome, which corresponds to the coding region of rpoB gene (279 bp - 2275228 to2275506), was observed in two patients. This missense mutation in CAC codon brings about a glutamic acid to histidine change in the amino acid sequence of RNA polymerase beta subunit at the position 442 (Glu442His), a region specific for rifampicin interaction, which might be responsible for unresponsiveness to rifampicin by manifesting a stable bacteriological index in these 2 patients even after completion of 24 months of multibacillary multidrug therapy (MB-MDT).

Limitations: The major limitations of multiple-primer PCR amplification refractory mutation system (MARS) assay is that it capable of detecting mutation at codon 425 and cannot distinguish any silent amino acid changes.

Conclusion: The study indicates the existence of Rifampicin drug resistance in Eastern India.

Molecular mimicry between Mycobacterium leprae proteins (50S ribosomal protein L2 and Lysyl-tRNA synthetase) and myelin basic protein: a possible mechanism of nerve damage in leprosy

Singh Itu, Sengupta U et al.
Microbes Infect. 2015 Jan 8. pii: S1286-4579(14)00337-2. doi:

Autoantibodies against various components of the host are known to occur in leprosy. Nerve damage is the primary cause of disability associated with leprosy. The aim of this study was to detect the level of autoantibodies and lympho-proliferative response against myelin basic protein (MBP) in leprosy patients (LPs) and their correlation with clinical phenotypes of LPs. Further, probable role of molecular mimicry in nerve damage of LPs was investigated. We observed significantly high level of anti-MBP antibodies in LPs across the spectrum and a positive significant correlation between the level of anti-MBP antibodies and the number of nerves involved in LPs. We report here that 4 B cell epitopes of myelin A1 and Mycobacterium leprae proteins, 50S ribosomal L2 and lysyl tRNA synthetase are cross-reactive. Further, M. leprae sonicated antigen hyperimmunization was responsible for induction of autoantibody response in mice which could be adoptively transferred to naive mice. For the first time our findings suggest the role of molecular mimicry in nerve damage in leprosy.

Calendar of Training - IELP 2015
(Please click above or visit http://bit.ly/1P4hyH7)