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Review Article

Recent Advances in Diagnostic Techniques and New Hope Towards Leprosy Elimination in the Post Elimination Era

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Abstract

Leprosy has been known since 1500BC, but the early diagnosis of leprosy still remains a challenge. Many newer techniques have been developed to diagnose leprosy at the field level so that surveillance of contacts can be done in a better way. Early diagnosis of leprosy helps to block the transmission and also in preventing the deformity. As there is increasing incidence of resistance to rifampicin, clofazimine and dapsone, we are in a need of newer combination therapy for leprosy. This article reviews about the recent techniques for early diagnosis of leprosy and the newer combination therapy for drug resistance cases.

Keywords: Diagnostic techniques; Leprosy; Leprosy treatment; PCR

Introduction

Leprosy becomes an ever challenging disease to entire medical fraternity, by the existence of fresh untreated cases, treated butrelapsed cases, multi drug resistant cases in every continent in spite of global approach with multibacillary drugs. Leprosy has left behind a terrifying image in the history as well as in human memories as a result of mutilation, rejection and exclusion from the society [1].

Leprosy control measures and elimination strategies are very effective in bringing down the global leprosy prevalence which has dropped down from 4.2 per ten thousand in 2002 to 0.69 in 2015. The leprosy control program has been a successful one but for few

hurdles. In India the National Leprosy Control Program (NLCP) has been in vogue since 1955. The annual new case detection rate in India was estimated as 9.73 per 100,000 population in 2015 [2]. With continuous implementation of elimination strategies in India almost thirty-three states/ union territories have achieved the level of leprosy elimination i.e., Prevalence Rate (PR) less than one case per ten thousand population. However, some rural areas at district level, still form the pockets of endemic regions with higher prevalence rates [3]. India alone accounted for 58.85% of the global leprosy burden [4]. According to September 2016 statistics, there are 102,178 cases in India. However, there is still an occurrence of considerable number of new cases in India, indicating an ongoing transmission, with an Annual New Case Detection Rate (ANCDR) of 9.98/100,000 population [4].

The diagnosis of leprosy is a challenge as there are no gold standard methods to differentiate between infection and disease. Leprosy is also an endemic disease in developing countries, where detection rates show only a slight trend toward a decrease in the disease prevalence. It is accepted that transmission occurs from human to human through the upper airways, although intermediate hosts like armadillos may play a role in certain countries like United States. Multi bacillary leprosy patients harbour millions of bacilli that can potentially contaminate their close relatives of house hold contacts. Surveillance of these contacts would be an easy control strategy to block transmission, as suggested by the World Health Organization (WHO). As we are not able to bring down the new case detection rate further as expected, recent studies focus on early diagnosis [5]. This article reviews various emerging advanced techniques in the diagnosis of leprosy. Treatment of leprosy has revolutionized the disease since the introduction of Multi Drug Therapy (MDT) in 1981. However, as there is an increasing incidence of resistance among commonly used drugs like rifampicin, dapsone and clofazimine, there is a necessity for newer effective combinations with bactericidal drugs. This article also reviews various newer drugs effective against leprosy and their combinations to improve the patient compliance and treatment outcome.

Advances in diagnosis of leprosy

Diagnosis of leprosy is based on slit skin smear for demonstration of Acid Fast Bacilli (AFB) for a long time. It is understood that there are many disadvantages of this procedure like AFB is not demonstrable in all patients especially paucibacillary group like True Tuberculoid (TT) and Borderline Tuberculoid (BT) types [6]. Demonstration of AFB needs trained technicians and referral laboratories and hence this procedure is followed at present only in the teaching institutions and not in the field level [7,8].

Histopathological examination of biopsy specimens from skin or nerve is another procedure in the diagnosis of leprosy which gives more information on the type of infiltrate and nerve involvement [9].

Immunological tests like 'Lepromin' which indicates the immune status in the patients is also outdated as both lepromin and Dharmendra antigens are not available freely at present. Hence we have to depend on alternate and newer techniques to make early diagnosis of leprosy.

Polymerase Chain Reaction (PCR) based diagnosis of leprosy

Studies on PCR were first carried out by Williams in the year 1990 [10]. The technique was specific and detected *M. leprae* DNA in biopsies from leprosy patients. More recently PCR has been used for extraction, amplification, and identification of *M. leprae* DNA in clinical specimens [5]. PCR can be done on samples from skin biopsy, skin smears, nerves, oral or nasal swabs, blood, ocular lesions and urine samples [11-13]. Different sequences are used for PCR for example, genes encoding 36 kDa antigen, 18 kDa and 65 kDa antigens [14]. Real-time PCR technology has further improved detection [15].

Early diagnosis of leprosy poses problems in certain patients who are infected, harbour the organism but lack classical signs and symptoms. In such cases, good surveillance of patients' close contacts has increased the detection rate at a very early stage as they have less severe clinical presentations and lower Bacteriological Indices (BI) [16-18]. Immuno diagnostics or immunological diagnosis of leprosy plays an essential role both in the diagnosis as well as the confirmation of leprosy at the early stage of infection. Most of the immunological tools, to detect *M. leprae* are based on their ability to detect major unique components like Phenolic Glycolipid-I (PGL-I) specific proteins by means of mono clonal and poly clonal antibodies or T cell immune response as measured by IFN-γ production. Apart from the lack of availability of good diagnostic techniques in all centres, there are also variations in the immunity among various spectra of leprosy patients. For example, most of the Pauci Bacillary (PB) patients, may not have detectable antibodies at an early stage of the disease and most of the Multi Bacillary (MB) patients do not produce IFN-γ in spite of their high bacillary load which can be picked up by either PCR or anti PGL-I detection [19].

Lack of a gold standard diagnostic test for leprosy and the inability to distinguish infected individuals from those exhibiting active disease makes leprosy diagnosis essentially based on clinical features. This is the reason for late diagnosis and hence spread of infection in the population and difficulties encountered to interrupt the disease in the population. In this context, detection of *M. leprae* DNA by PCR in difficult to diagnose cases favours early identification and correct diagnosis. By this method, pure neural leprosy is more commonly diagnosed, indeterminate leprosy is detected more frequently, and early detection in household contacts is made possible. PCR confirms the disease even in cases of clinical and histopathological dispute [16-18].

Real-time PCR measure total DNA content estimated by molecular levels and could be correlated to bacterial load, corroborating the clinical data, which can be useful to determine a Molecular Bacteriological Index (MBI) [15,20].

Since PCR assays are based on DNA detection and did not reflect live bacilli, to overcome this problem, studies were conducted on Reverse Transcriptase PCR (RT-PCR) based assays for *M. leprae* viability estimation. RNA based tests only reflect nucleic acids from living organisms [21].

PCR is used in contacts of leprosy patients and if found positive the risk of progression to active disease increases. Genome studies also proved that wild armadillos transmitted leprosy to patients in the southern United States. DNA based PCR assays can be 100% specific while the sensitivity ranges from 34-80% in PB patients and 90% in MB patients [5].

Quantitative PCR (qPCR) is a useful tool for the diagnosis of infectious diseases, and has been used to detect several pathogens including *M. leprae* [22]. Skin biopsy specimens or slit skin smears can be subjected for qPCR assessment in leprosy. Studies on qPCR from skin biopsy and slit skin smear of leprosy patients from Brazil showed 85% sensitivity with 85% positive predictive value and 61% negative predictive values respectively [22]. Currently qPCR is considered as a rapid, sensitive and specific method for diagnosing *M. leprae* from tissue specimens. The use of Repetitive Element Regions (RLEP) has the advantage of being more sensitive. In one study from Brazil, qPCR method detected *M. leprae* in the peripheral blood samples in 200 untreated leprosy cases and 826 household contacts [23]. This study revealed that if PCR was positive in blood samples from index cases at the time of detection they were at a greater risk of developing leprosy at a later date [23]. Recently *M. leprae* DNA was amplified from oral mucosal samples. This technique detected bacilli even when they are undetectable by routine examination [24].

Comparison of various types of PCR is presented in table 1.

	Conventional PCR	Real Time Quantitative PCR	RT-PCR
Advantages	Collects data at the end of reaction More sensitive than serological methods	Collects data at the exponential phase High resolution Less time consuming Automated	Able to detect viable bacilli Helpful in assessing response to chemotherapy
Disadvantages	Poor resolutison Time consuming	Expensive	Expensive Only available at research centres

Table 1: Comparison of various PCR methods.

Immunological Tests for Leprosy

Antibody based tests

PGL-1 based serology, particularly in the kit format, continues to play a greater role in leprosy control in the following manner:

a. As a confirmatory marker

b. As a predictor of disease outcome

c. An indicator of nerve damage and exacerbation

d. As a tool for preclinical intervention

Protein based serology in the micro array format adds to diagnostic resolution and may aid in early clinical detection once reduced to a 'field-friendly format'.

Assays based on cellular immune responses, and the facile detection of gamma interferon, may assist in pre-clinical detection allowing for early diagnosis and appropriate treatment [25].

Diagnosis of leprosy among contacts with sub clinical infection warrants immunological intervention. Various immunological tests which detect leprosy antibodies were compared for their efficacy. The native and synthetic PGL-1 ELISA assays detected antibodies in 22% and 31% in PB patients and the ML flow test failed to detect antibodies in this group. The study concluded that all three tests showed sensitivity ranging from 60-68%. The use of PGL-1 ELISA and ML flow tests are thus recommended as additional tools in the diagnosis and classification of clinical forms, aiding in prescribing the correct

treatment regimen to prevent subsequent nerve damage and disability [26]. PGL-1 ELISA may also be used to detect sub clinical infection in leprosy [26]. PGL-1 recognizes the trisaccharide portion of the molecule whereas the ML flow test is a lateral immune chromatographic flow test which detects the anti PGL-1 antibodies. Both these tests are useful in the assessment of the host immune response to *M. leprae* [26].

Slit skin smear is the most widely used complementary examination in leprosy patients which has been in vogue for many years. It has disadvantages like lack of infra structure like laboratory, specially trained personal and validity of test reading. ML flow test is an immune-chromatographic assay which detects *M. leprae* specific anti PGL-1 IgM antibodies. It is easy to perform and can be used at primary health care centres because the test does not require a specific infrastructure. ML flow test results are recorded as 0 (sero negative), 1+, 2+, 3+ or 4+ according to colour intensity of antigen band [27]. ML flow test is positive in 97.8% of MB patients with BI >2+. It is also positive in 93.5% of all patients with positive skin smears. Interestingly it shows positive in 37.8% of patients with negative skin smears. These patients would have been started on MDT-PB regimen in the absence of these test reports which could lead to relapse after many years [27].

Two fusion proteins ML0405 and ML2331 (LID-1 designated leprosy IDRI diagnostic) have been tested for their antibody reactivity and was found to be more sensitive than PGL-1 [28].

Antigen based tests

1. Skin test antigens: Two new antigen preparations namely: Armadillo derived *M. leprae* (MLSA-LAM) and *M.leprae* Cell wall Antigen (MLCwA) are tested in various studies.

2. Another new antigen MMPII (bacterio ferritin) which is also reactive in PB sera has been tested.

3. Glycolipids like PDIM (phthiocerol dimycocerosate), Glycopeptidolipids, and mycolic acids like Trehalose mono mycolate and Trehalose dimycolate form the cell wall components of *M. leprae* and are assessed by using thin layer chromatography. Antigen detection like antigen 18 is another new area of research identified instead of antibody detection.

4. ESAT-6 (Early Secretory Antigen Target) ML0049 was found to be 100% specific and sensitivity was 82.4% in MB and 19.4% in PB cases.

The major disadvantages of all these tests are the cost effectiveness and hence out of reach of people [29,30].

M. leprae genome

M. leprae genome has been studied extensively. The abundance of pseudogenes compared to functional counterparts within the genome of *M. tuberculosis* helps to explain the intra cellular nature of *M. leprae* and its inability to grow in axenic media. Genomic studies revealed more similarities between *M. leprae* genome from India and Brazil, they are identical in sequence. The phylo-geographical hypothesis shows the evidence that the organism originates from eastern Africa [31].

One current approach is to use bioinformatics and comparative genomics to identify potentially antigenic proteins for diagnostic purposes and future vaccine developments. There are three classes of proteins in *M. leprae,* those restricted to *M. leprae*, those present in *M. leprae* with orthologs in other organisms and those exported or surface exposed proteins. After the completion of genomic sequencing of *M. leprae*, genes that are unique to *M. leprae* and no homologues to *M. tuberculosis* are looked into [32].

Cytokine profiles in leprosy

Leprosy develops in a patient mainly due to altered host response to *M. leprae* which depends on microbiological and immunological characters of the individuals. Studies on cytokines revealed involvement of Th1 cytokines like interleukin-2 and IFN-γ in TT leprosy and Th2 type cytokine like IL-4, IL-5 and IL-10 in LL patients [33].

High-Resolution Ultrasonography (HRUS) in leprosy

High-Resolution Ultrasonography (HRUS) is a non-invasive, imaging technique, which provides real time examination of deeper tissues including peripheral nerves in static and dynamic states such as blood flow. Since the hall marks of leprosy are nerve enlargement and inflammation, HRUS and colour Doppler imaging can be used to demonstrate nerve damage. Five studies evaluated 111 patients with sonographic studies. These studies revealed that peripheral nerve ultrasound provides information on the exact location of nerve enlargement and morphological alterations in the nerve including echo texture, fascicular pattern and vascularity. This investigation is more useful in pure neuritic leprosy [34].

Most modern sonographic machines use pulsed colour Doppler to assess whether structures, usually blood, are moving towards or away from the probe and its relative velocity. Colour Doppler imaging of each nerve can be performed to look for presence or absence of blood flow signals in the epineural plexus and infra fascicular vessels of nerve trunk. Normally there is hypo-vascularity of nerve trunks. Increased blood flow signals seen in colour Doppler in thick and tender peripheral nerves of leprosy denotes oedematous and hyperaemic changes secondary to inflammation leading to alteration of an effective blood nerve barrier during reactions [35].

Non-invasive tests like HRUS, MRI (Magnetic Resonance Imaging) and nuclear magnetic resonance are available in a few centres only. They have shown arterial and venous flow is altered in damaged nerves and in active ENL and are patient friendly.

Electro-neuromyography

The general principle of electro-neuromyography is that the recording or active electrode is placed over muscle or nerve segment to be studied. Motor and sensory amplitude reduction was the earliest and the most frequent abnormality. Low conduction velocity of the ulnar nerve across the elbow was present in over 55% of the patients [36].

Treatment and Drug Resistance

Current treatment regimen [37]

WHO simplified both classification and treatment of leprosy into two different groups. They are Pauci Bacillary (PB) and Multibacillary (MB). All patients with less than five patches are included into PB group and all patients with five or more than five patches are included into MB group. Similarly patients with more than one nerve trunk

involvement are included into MB. Recent classification includes all patients with smear positivity into MB group. WHO recommendation for treatment of PB and MB leprosy are shown in the table 2 and 3.

Multidrug Therapy for Paucibacillary Disease (MDT-PB)		
Age group	Rifampicin	Dapsone
Adult 50-70 kg	600mg/month Supervised	100mg Daily unsupervised
Child 10-14 year	450mg/month Supervised	50mg Daily unsupervised
Child <10 year	300mg/month Supervised	25mg Daily unsupervised

Table 2: PB regimen.

Duration: Six months.

Multidrug Therapy for Multibacillary Disease (MDT-PB)			
Age group	Rifampicin	Dapsone	Clofazimine
Adult 50-70 kg	600mg/month supervised	100mg daily unsupervised	300mg/month supervised & 50mg daily unsupervised
Child 10-14 year	450mg/month supervised	50mg daily unsupervised	150mg/month supervised & 50mg on alternate day unsupervised
Child <10 year	300mg/month supervised	25mg daily unsupervised	100mg/month supervised & 50mg twice weekly unsupervised

Table 3: MB regimen.

Duration: Twelve months.

Problems of current MDT regimen [38]

1. Current MDT regimen is still complicated as two types of drug administrations, i.e., monthly and daily administration and daily administration is self-administered. Should a patient fail to comply with daily self-administered treatment, he is virtually treated with rifampicin monotherapy. Therefore, the current MB regimen is not resistance proof for rifampicin.

2. Duration of MDT is relatively too long.

3. Emergence of drug resistance is virtually unavoidable in any large scale treatment of an infectious disease with microbial agents. Some patients with rifampicin resistance and multidrug-resistant leprosy have already been reported. A safe and effective alternative regimen should be developed for patients with rifampicin resistance or who cannot tolerate rifampicin [38].

New MDT regimens

New regimens for treatment of leprosy should be developed to simplify the treatment and facilitate supervision of drug administration and to treat rifampicin resistant leprosy or in patients who cannot tolerate rifampicin. Newer anti-microbial agents with powerful bactericidal activity against *M.leprae* should be incorporated into the new regimens. Newer drugs which display bactericidal activity against *M.leprae* are given in table 4 [39,40].

The proposed regimens for simplifying MDT are [39,40]

1. Rifapentine 900 mg (or Rifampicin 600mg) + Moxifloxacin 400 mg + Clarithromycin 1000mg (or Minocycline 200 mg) administered once monthly under supervision for 12 months.

2. For rifampicin resistant leprosy:

a. Intensive phase: Moxifloxacin 400 mg + Clarithromycin 500mg + Minocycline 100mg + Clofazamine 50 mg daily for six months.

b. Continuation phase: Moxifloxacin 400 mg + Clarithromycin 1000 mg + Minocycline 200 mg once monthly for addition of 18 months.

3. ROM (Rifampicin + Ofloxacin + Minocycline) therapy: A review on meta-analysis comparing 6 studies on comparing ROM therapy with MDT and 8 studies that evaluated only ROM therapy was done. It showed that in PB patients ROM therapy as a single dose was less effective. However, ROM therapy as multiple doses showed equal efficacy that of MDT MB in MB patients. Multiple doses can be considered as another alternative for PB patients [41].

4. Gatifloxacin is a FDA approved fluoroquinolone antibiotic that inhibits the bacterial enzymes DNA gyrase and topoisomerase IV. The dose ranges from 100-400 mg/day.

5. Linezolid is a synthetic oxazolidine drug approved by FDA to treat infections by gram positive organisms. It acts as an inhibitor of protein synthesis of bacteria. The dose ranges from 600 mg per day. It can cause bone marrow suppression or optic atrophy. More than 36% of that group discontinued linezolid due to adverse effects [42].

Drugs	Class	Bactericidal Activity Mice	Bactericidal in Activity Humans	Unit Cost in
Pefloxacin	Fluroquinolone	++	++	Moderate
Ofloxacin	Fluroquinolone	++	++	Moderate
Moxifloxacin	Fluroquinolone	+++	+++	High
Clarithromycin	Macrolide	++	++	Moderate
Minocycline	Tetracycline	++	+=	Moderate
Rifapentine	Rifamycin	+++	Not done	High
R207910	Diarylquinoline	+++	Not done	Not yet available
Linezolid	Oxazolidinone	+	Not done	High

Table 4: Newer drugs in leprosy.

Limitations of newer drugs

1. Paucibacillary patients treated with ROM therapy were twice as likely to have a relapse compared with those that were treated with WHO MDT.

2. There are insufficient data to conclude on the efficacy of multidose ROM therapy in MB leprosy [32].

3. Though the bactericidal activity of moxifloxacin and rifapentine are comparable to rifampicin, they are too costly to afford.

4. Since WHO recommended regimens have displayed excellent results in the field, the effectiveness and possible side effects of any newly proposed drug regimen must be carefully tested in controlled clinical trials and in field trials before being applied to patients [43].

Drug resistance

Emergence of rifampicin resistance would create a lot of difficulties for an individual patient, and its widespread dissemination would pose a problem to the community and a threat to leprosy control.

Although rifampicin resistance was not reported in more than 10 million patients who completed MDT, this could be due to following reasons:

1. Post-MDT surveillance for relapse has been discontinued.

2. Rifampicin susceptibility testing facilities are available only in referral centres.

Polymerase Chain Reaction (PCR) based DNA sequence analysis of the rpo-B gene of *M. leprae* was in full concordance with those of susceptibility testing in mouse footpad system. This approach may lead to the diagnosis of 80% rifampicin resistant strains of *M. leprae*. Secondary rifampicin resistance could also emerge in some patients who initially improved with rifampicin. Similarly multi drug resistance to *M. leprae* also has been encountered [44,45].

Drug resistance-mechanisms and molecular diagnostics

MDT for leprosy has been introduced in 1981 to prevent the spread of drug resistant *M. leprae*. It is unclear whether or not the frequency of dapsone resistance has declined since the implementation of MDT, since prior to that time the majority of patients with isolates with dapsone resistance mutations harboured low degree mutations strains in many years. Dapsone resistance in *M. leprae* is the result of specific mutations in condons 53 and 55 within the fol-P1 gene coding Dihydropteorate Synthase (DHPS). Rifampicin resistance is conferred by mutations in the β sub unit of RNA polymerase coded by rpo-B gene [32].

Dapsone resistance was recorded in 1964, rifampicin in 1976 and for ofloxacin in 1996 [45]. Resistance to anti leprosy drugs dapsone, rifampicin and ofloxacin evolves by amino acid substitution at the site of action of these drugs [32]. Recent studies identified point mutations in the fol-P1 gene that encodes dihydropteorate synthase in dapsone resistant *M. leprae* [32]. Rifampicin resistance is associated with mutations in the rpoB gene that encodes the β sub unit of RNA polymerase. Resistance to ofloxacin is associated with mutation in gyrA gene encoding the A sub unit of DNA gyrase of *M. leprae*. The susceptibility testing for these drugs are possible by a rapid DNA based PCR direct sequencing method. PCR-SSCP (Single strand Conformation Polymorphism) and PCR solid phase hybridization assays have been developed to detect rifampicin resistant *M. leprae* in human specimens [46,47].

Frequency of drug resistant mutations

In a study from Myanmar, Indonesia and Philippines it was found that out of 252 isolates obtained from new cases, 3% were dapsone resistant and 2% were rifampicin resistant. In samples from 53 relapsed patients this ranged from 15% for dapsone and 8% for rifampicin. Two patients had mutations for both dapsone and rifampicin. There were no mutations for quinolone resistance in this study. The authors recommended monitoring drug resistance globally by testing *M. leprae* DNA from relapse cases and from a representative sample of new cases [48,49].

Multi-drug therapy and drug resistance

To simplify the treatment of leprosy, Uniform MDT (U-MDT) has been recommended. This includes treating all leprosy patients with all three drugs for 6 months. The assessment of U-MDT is underway but as yet there are no clinical studies carried out on efficacy of MDT in pauci Vs multi bacillary leprosy cases in large scale. There are also no reports available on the exact status of relapse cases with U-MDT regimen. Patients with MB disease treated with a 6 months MDT regime did not show much improvement in clinical and histological parameters as those treated for 12 months. The use of molecular tools for monitoring drug resistance is feasible and simplification of techniques may allow such tools to be used in field laboratory settings.

The major disadvantages of testing newer drugs in leprosy are [50]

1. Drugs with varying degree of bactericidal activity are needed.

2. Research should be done into cost and cost-benefit scenarios.

3. If preliminary trials are successful, well tolerated and provide early evidence of effectiveness for treating MB leprosy, then controlled clinical studies should be undertaken.

4. In the controlled clinical trials, multiple regimens, including current MB regimen as positive control should be compared. Each arm of the trial requires at least several hundreds of MB patients, and periodical follow up of 7 years after completion of treatment.

5. Better methodologies are required to accelerate the development of new MDT regimens [50].

Reaction in Leprosy

Type-1 reaction

Pathogenesis of Type-1 lepra Reaction (T1R) involves Th1 type cells and lesions in reaction express pro inflammatory IFN-γ, IL-12 and the oxygen free radical producer nitric oxide synthase. The plasma levels of chemokine IP-10 (CXCL10) were increased in type-1 reaction. Treatment of the reaction causes clinical improvement but changes in the inflammatory cytokines lag behind by some considerable time and in some remain unchanged. This variation in inflammatory activity within different compartments is important to study T1R and may explain why treatment is always not effective. It also has been found that there is a local conversion of endogenous corticosteroids in the skin lesions of leprosy with T1R. The gene expression of the enzyme 11β-hydroxysteroid dehydrogenase type-II which converts active cortisol back into inactive cortisone is decreased in patients with T1R compared with controls with no reaction [51,52].

Type 2 reaction

ENL has a predominant Th2 cytokine profile with increased expression of interleukins (4, 5, 8, 10 & 17). Serological markers like IL-7, TNF-α, CCL-11, Alpha 1 acid glycoprotein and MMP-9 are found to be elevated in type 2 reactions.

Similar to type 1 reaction, autoimmunity may also play a role in ENL [52].

Corticosteroids for treating nerve damage in leprosy

Corticosteroids are commonly used for treating nerve damage in leprosy. Three RCTs involving 513 people were studied. Evidence from RCTs does not show significant long term effect for either long standing nerve function impairment or mild sensory impairment. A 5 month corticosteroid regimen was significantly more beneficial than a 3 month corticosteroid regimen. A standard 12 week course of prednisolone is recommended by WHO which can be safely used in the field. The critical dose to control reaction after the initial period was considered to be 15 -20 mg daily [53].

Chemo Prophylaxis

Since NCDR has not diminished following implementation of MDT, there has been renewed interest in chemoprophylaxis. Dapsone and acedapsone had been used earlier for this purpose. Two basic principles of chemoprophylaxis are [54].

1. Rifampicin should be one of the components.

2. Treatment should be administered in a single dose.

There are only few trials available on this. One of these studies showed a protective effect of 35-40% [54]. The limitations of chemoprophylaxis are:

1. Chemoprophylaxis is confined to household contacts only.

2. The effect of chemoprophylaxis is only temporary and has to be repeated.

Leprosy Vaccine

One should explore new vaccines which are inexpensive and

potent for leprosy control. *Mycobacterium habana, Mycobacterium vaccae*, Ag85 A/B, Indian Cancer Research Center bacilli (ICRC Vaccine), BCG, *Mycobacterium indicuspranii* (Mycobacterium w) are some of these. They can be grouped under:

1. Protein antigens known to be homozygous with TB proteins.

2. Proteins shown to induce protective immunity in mouse foot pad assay.

From financial point of view vaccines are not cost effective measures when applied universally to stop transmission of low incidence diseases. A therapeutic vaccine could be offered to individuals in close contact with an index case and therefore thought to be of at risk of infection [55,56].

Adverse effects of MDT in leprosy

Dapsone is associated with severe adverse effects such as Dapsone Hypersensitivity Syndrome (DHS), dermatitis, hepatitis, a granulocytosis and severe hemolysis. In Brazil, Deps reported in 54% the drug had to be withdrawn due to its adverse effects [57]. Dapsone allergy rates to 3.1% in 636 newly diagnosed MB patients in Bangladesh and Nepal [58]. In the event of severe dapsone toxicity WHO recommends no modification of MDT other than immediately stopping dapsone is required in the case of those receiving MDT-MB. In case of PB patients, dapsone should be replaced by clofazimine in the same dose as that of MDT-MB [58].

In a study conducted at a tribal region, Chhattisgarh state, India 176 patients records were analyzed for adverse effects. The authors found 79 patients had adverse effects due to one or more components of MDT. Seventy three patients out of seventy nine had adverse effects due to dapsone, eight due to rifampicin and 16 due to clofazimine [59]. The introduction of MDT in 1981, was aimed to control primary and secondary dapsone resistance in leprosy emerging at that time. MDT introduction had additional benefits such as rifampicin as bactericidal drug, intense coverage, reaching remote areas, removing gender bias and integrating leprosy with other services thus achieving a substantial reduction in leprosy patients.

Among the three drugs in the regimen, rifampicin is included in both regimens. Rifampicin is considered as a safe drug, but still this has produced side effects like maculopapular eruptions, thrombocytopenic purpura, hepatitis, flu-like syndrome, haemolytic anemia, shock, respiratory insufficiency and acute renal failure. Clofazimine is well tolerated and non-toxic in the usual dosage.

Adverse effects of MDT ranged from 38-45% of patients in various studies. Dapsone syndrome was noticed in 2% of patients and had a mortality rate of 0.5%. One or more drugs had to be stopped in 5% of patients in one study due to adverse effects and alternative regimen was instituted. Difference in the rate of adverse effects in various studies could be explained due to genetic susceptibility and racial and ethnic factors in the drug metabolism [59].

Recently described entity like Acute Generalized Exanthematous Macular Eruption (AGEP) is accompanied by non-follicular sub corneal sterile pustules all over the skin and is predominantly a drug induced dermatosis like dapsone. This condition was earlier considered as a variant of pustular psoriasis. The onset is acute, accompanied by episode of fever, which regresses in a few days. Resolution of pustules occurs within 4-10 days after discontinuing the suspected drug. The main triggering factors are β lactum antibiotics. Dapsone hypersensitivity syndrome is not associated with sub corneal pustules. AGEP is benign, unlike Drug Hypersensitivity Syndrome (DHS) [60].

Conclusion

Leprosy has been prevalent for centuries. Early diagnosis is still a challenge. PCR techniques fill this gap to a large extent but, they are costly and not available for use at the field level. *Mycobacterium leprae* was first to be identified but yet to be cultured and hence susceptibility testing to new drugs are carried out only in animals. Gene probe studies have overcome this difficulty to a certain extent but it is a distant dream as they are not available for routine use. MDT has cured over 1.2 million patients since its implementation, but the incidence of leprosy in children has not decreased up to our expectations. Six months or one year treatment is considered as one of the reasons for noncompliance in many studies. Hence we have to look for trials with newer combinations with shortened regimens. We still hope to evolve newer diagnostic techniques and drugs in future.

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