Target Product Profile for development of a point-of-care diagnostic test for dermal leishmaniases

Dermal leishmaniases

Localized cutaneous leishmaniasis and its evolving forms diffuse cutaneous leishmaniasis, mucosal leishmaniasis and cutaneous leishmaniasis recidivans, together with the visceral leishmaniasis sequela post-kala azar dermal leishmaniasis, account for about one million dermal leishmaniases cases per year worldwide. Although not lethal, the dermal leishmaniases cause chronic and disfiguring skin lesions, which are an important cause of morbidity and stigma.

Microscopy remains the reference test for diagnosis of dermal leishmaniases; however, it has low and variable sensitivity and requires well-trained personnel. The technical complexity and cost of the more sensitive molecular techniques (e.g. PCR) limits their application in routine diagnosis in endemic areas. [1]. As a result, a high number of patients are put on treatment without laboratory confirmation, exposing a variable number of them to unnecessary toxic treatment [2,3].

Thus, there is a great need for point-of-care (POC) tests for early diagnosis of dermal leishmaniases, in order to benefit both patients and communities by early identification of those that need treatment, reducing the risk of both sequelae and ongoing *Leishmania* transmission.

Public Health Response

In 2007, World Health Assembly 60, in its Resolution on the Control of Leishmaniasis, urged Member States, among other actions:

> i) "to strengthen prevention, active detection and treatment of cases of both cutaneous and visceral leishmaniasis in order to decrease the disease burden; "
>
> ii) "to strengthen the capacity of peripheral health centres to deliver primary and secondary care, so that they provide appropriate affordable diagnosis and treatment and act as sentinel surveillance sites;"

and requested that WHO's Director-General "promote research pertaining to leishmaniasis control, including in the areas of safe, effective and affordable vaccines, diagnostic tools and medicines with less toxicity, and dissemination of the findings of that research".

Available Diagnostic Tools

Microscopy of Giemsa stained samples from lesions, including skin scrapings, fine needle aspirates, or slit-skin smears, remains the reference test for diagnosis of the different forms of dermal leishmaniasis. However, as noted above, microscopy has significant shortcomings, and more sensitive molecular tests have not yet been widely adopted. Other simpler tests for detection of leishmanial DNA, such as loop-mediated isothermal amplification (LAMP), are yet to be implemented. Other potential approaches include serology, which may be useful for screening of PKDL and mucosal leishmaniasis, but cannot be used for confirmation, as presence of antibodies may be due to previous episodes or exposure to the parasite by living in endemic areas. The leishmanin skin test or Montenegro skin test can also aid in

the diagnosis of CL, but again the test is not a marker of active infection, and therefore has limited value [1].

In 2016, the foundation for Innovative New Diagnostics (FIND), involved a panel of 47 international experts on leishmaniasis in a survey to rank diagnostic priorities. A rapid test for CL was identified among the top priorities [4].

Currently there is an FDA cleared and CE marked rapid test targeting *Leishmania* antigen that is designed for CL diagnosis, *CL Detect™ Rapid Test for Cutaneous Leishmaniasis* (InBios International Inc., Seattle, WA) https://inbios.com/cl-detecttm-rapid-test-for-cutaneous-leishmaniasis-intl/. Studies have shown high specificity, but unfortunately the sensitivity is quite variable across *Leishmania* species and endemic regions [5-8].

The WHO Diagnostic Technical Advisory Group for Neglected Tropical Diseases

The WHO Department of Control of Neglected Tropical Diseases (NTD) set up the Diagnostic Technical Advisory Group (DTAG) to be the principal advisory group to WHO on NTD diagnostics. This group works to ensure a unified method will be used to solve NTD diagnostic needs and to direct WHO strategies to develop efficient diagnostic tools. The first meeting of the group occurred in Geneva, Switzerland in 2019 [9]. The DTAG noted the following diagnostic needs for dermal leishmaniases:

- Rapid test for PKDL – to distinguish post-kala azar dermal leishmaniasis (PKDL) from other skin conditions.
- Rapid test for confirmation of suspected cases of cutaneous leishmaniasis (CL) at peripheral health facilities.

NTD Road Map 2021-2030

Cutaneous leishmaniasis is one of the diseases targeted for control in the 2021–2030 NTD road map. The main 2030 target for CL is that at least 87% of cases are detected, reported and treated. At least 64 countries are expected to be validated for elimination of VL as a public health problem by 2030; which means PKDL (which plays an important role in transmission) needs to be specifically addressed in the Indian sub-continent and some countries in eastern Africa [10].

To achieve these goals, more effective and user-friendly diagnostics for CL and PKDL are needed. Enabling decentralized testing is key for both individual cases and mass screening in the visceral leishmaniasis near-elimination context; i.e. testing at the public health centre and/or at the community level.

A rapid test targeting *Leishmania* antigens common across *Leishmania* species will address major diagnostic needs for dermal leishmaniasis, and by addressing PKDL may also contribute to the control and elimination of visceral leishmaniasis.

Background and scope for the TPP

Thus there is a great need for point-of-care (POC) tests for early diagnosis of dermal leishmaniases, in order to benefit both patients and communities by early identification of those that need treatment, reducing the risk of both sequelae and ongoing *Leishmania* transmission. It is then important that new POC tests to be developed meet the needs of the target population and the requirements for implementation in resource-limited settings, where most cases of dermal leishmaniasis occur. To this end we present here a Target Product Profile (TPP) for a POC test for dermal leishmaniases. The TPP was defined through several rounds of discussions and by consensus with stakeholders and experts in dermal leishmaniases from different type of organizations and endemic regions.

Audiences engaged and external consultations to develop the TPP

A draft TPP was developed by leishmaniasis experts at the Foundation for Innovative New Diagnostics (FIND) and Drug for Neglected Diseases initiative (DNDi). This first draft was presented at the 2nd redeLEISH Meeting (Medellín, Colombia, July 2015) for discussion with a panel of 70 experts on leishmaniasis [11]. The draft was refined based on the inputs received from experts in that meeting, and a new document prepared to be shared with a second panel of experts. The second panel was composed of 31 experts from different organizations and endemic regions. Consensus was reached to define a TPP with 29 priority features [1]. This TPP has been presented at different stakeholder meetings, including: the 6th World Congress on Leishmaniasis [12] and the Biennial meeting of the Global Buruli Ulcer Initiative, 2017 [13].

In March 2016, and as part of a consultation to develop FIND's leishmaniasis strategy, 40 experts on leishmaniasis helped identify and rank leishmaniasis diagnostic priorities. The experts had experience on the different forms of leishmaniasis from different endemic regions and represented the academy, international organizations and product development partnerships, among others. The experts identified Rapid POC test for dermal leishmaniasis as one of the highest priorities. This was further discussed at the 6th World Congress on Leishmaniasis [12].

After further refinement, this TPP was published in 2019 in *Parasite epidemiology and Control* with authors from FIND, DNDi and WHO [1]. A version of this TPP, has been reviewed by the WHO DTAG and is provided as Annex I to this document.

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Target Product Profile for a point-of-care diagnostic test for dermal leishmaniases

1. Scope of the test	**Minimum**	**Ideal**	**Annotations**
1.1 Goal of test. Intended use	Detection of active localized cutaneous leishmaniasis (LCL)	Detection of active cutaneous leishmaniasis (CL, any form) or post-kala azar dermal leishmaniasis (PKDL) with the purpose of initiating treatment during the same clinical encounter (or same day)	LCL is the most prevalent of dermal leishmaniases (>80% of the cases). This clinical form is present in all CL endemic regions. All *Leishmania* species cause LCL. Other forms of CL usually evolve from LCL
1.2 Target population	Individuals with clinical signs suggestive of LCL	Individuals with clinical signs suggestive of any form of CL, or PKDL	
1.3 Target operator of test	Trained laboratory staff	Health worker at PHC level without laboratory training	Most patients go to health facilities with limited human resources
1.4 Lowest setting for implementation	Decentralized health care facilities with minimum laboratory infrastructure	Decentralized health care facilities with no laboratory infrastructure, or mobile team	This test could replace microscopy, as has happened with other diseases (e.g. malaria)
1.5 Target analyte to be detected	*Leishmania antigen*		
2. Performance characteristics	**Minimum**	**Ideal**	**Annotations**
2.1 Clinical sensitivity	95% in parasitologically confirmed cases	100% in parasitologically confirmed cases	Measured in frozen or fresh samples from parasitologically confirmed patients (microscopy and/or culture and/or PCR from skin scrapings, swabs, biopsies, aspirates, etc.). A combined reference standard according to each region should be considered
2.2 Clinical specificity	>90%	>95%	Tested against reference standard (according to each endemic setting), including subjects with other diseases affecting the skin
2.3 *Leishmania* species-specificity	*Leishmania* genus-specific	*Leishmania* species-specific	Different treatment options might be needed for different species
2.4 Type of analysis. Quantitation	Qualitative		There is no need for quantification as parasite burden will not guide therapy

3. Test procedure	Minimum	Ideal	Annotations
3.1 Training needs. Time dedicated to training session for end users	One day for any level health care worker. Job aid provided	Less than half a day for any level health care worker. Job aid provided	
3.2 Sample type	Lesion fine needle aspirate, skin scrapping, biopsy, etc.	Lesion swab	Minimally invasive sampling procedures will be preferred
3.3 Sample preparation. Total steps	3–5 simple steps procedure	Direct testing from lesion swab	
3.4 Number of steps to be performed by operator	< 10; 1 timed steps	< 3; 1 timed steps	
3.5 Need for operator to transfer a precise volume of sample	Acceptable with a disposable transfer device provided	No	Sample may need to be eluted in specific buffer (included in the kit)
3.6 Time to result	< 1 h	< 20 min	
3.7 Internal control	Included		Positive control to confirm validity of the test
3.8 Reading system. Interpretation of results	Visual (naked eye) or simple reading device	Visual (naked eye)	See 3.9
3.9 Auxiliary equipment	Test reader (for lateral flow assay, dual path platform, or similar)	None, instrument free (required materials are included in the kit)	There are RDTs that generate a fluorescent signal that increases sensitivity, a reader is needed to detect this signal. In these cases a connectivity option could be desirable, enabling sending results to a reference lab, coordinator, reporting system, etc. thresholds
3.10 Power Requirements	Battery operated	None required	If a reading device is needed it should be small, portable or hand-held instrument (<1 kg) that can operate on rechargeable battery or solar power lasting at least 4 h (8 h preferred)
3.11 Need for maintenance/spare parts	None		

4. Operational characteristics	Minimum	Ideal	Annotations
4.1 Operating conditions	–40 °C, up to 80%relative humidity (RH), 0–2000 m above sea level	5–50 °C, up to 90% RH, 0–4000 m above sea level	High environmental temperatures and high humidity are often a problem in countries where CL is endemic. Some laboratories for CL diagnosis are located at high altitude (e.g. La Paz, Bolivia)
4.2 Reagent kit transport	No cold chain required; tolerance of transport stress for a minimum of 48 h at −15 °C to 50 °C	No cold chain required; tolerance of transport stress for a minimum of 72 h at −15 °C to 50 °C	Refrigerated transport is costly and often cannot be guaranteed during the entire transportation process. Frequent delays in transport are common
4.3 Reagent kit storage/stability	No cold chain required. Up to 12 months at 40 °C, up to 70% relative humidity	No cold chain required. Up to 24 months at 50 °C, up to 90% relative humidity	Should be able to tolerate transport stress (48 h at 50ºC). To include test quality detector (for surpassed temperature or humidity)
4.4 Reagents reconstitution. Need to prepare the reagents prior to utilization	Few simple steps	All reagents ready-to-use	Simple steps like resuspension of lyophilized reagent
4.5 In use stability	>1 h for single use test after opening the pouch		High environmental temperatures and high humidity are often a problem in countries where CL is endemic
4.6 Biosafety requirement. Level of protection to be made available for the staff and the samples	No need for biosafety cabinet. Standard biosafety precautions when handling potentially infectious materials. No contraindications to routine use		
5. Pricing	**Minimum**	**Ideal**	**Annotations**
5.1 Maximum price for individual test.	< 5 USD per test	< 1 USD per tesT	Assumption that the test is produced at a large scale, transport costs from manufacturing company not included
5.2 Maximum price for instrumentation. If needed	< 2000 USD	< 2000 USD	In case a test reading device is needed
5.3 Expected scale of manufacture	1.0 million tests/year	2.5 million tests/year	Based on 0.7–1.2 million estimated CL cases; and provided the test has better performance than microscopy