Clinical Validation Report on IVD Reagents

Product name: Malaria pf/pan Rapid Test Kit

Model and specification: 25 tests/box, each test strip packaged separately

Type of clinical trial: Clinical validation

Completion date: April 17, 2022

Testing agency: DSP Center for Clinical Laboratory

(Romanian Public Health Directorate – DSP trough The Laboratory of the National Institute of Public Health – INSP and/or other Authorised Laboratories under OMS validation).

Abstract

To evaluate the Malaria pf/pan Rapid Test Kit (the "Test Kit" for short) produced by Wuhan Jucheng Medical Technology Co., Ltd. ("the Company" for short) for clinical application in qualitative detection of the content in clinical samples (blood), DSP Center for Clinical Laboratory conducted a clinical study on the test strip therein. A total of 200 blood samples were selected as the study objects, including 100 positive samples and 100 negative samples confirmed diagnosis and treatment protocol. The kits used for diagnosis was PhoenixDx produced by Procomcure Biotech GmbH, was used as the reference kit. Based on the test result of the reference kit, the study objects were divided into PhoenixDx antigen positive group and PhoenixDx antigen negative group. At the same time, these samples were tested with Test Kit, and the test results of the Test Kit and the reference kit were compared and statistically analyzed. The results showed that the negative coincidence rate, positive coincidence rate and total coincidence rate between the Test Kit and the reference kit all were greater than 95%, indicating that the Test Kit is in good consistency with the reference kit, and suitable for clinical auxiliary diagnosis.

I. Introduction

The research and development work of the Test Kit produced by the Company has been completed. Clinical validation work has been started to validate the suitability and accuracy of the test strip in clinical application. Entrusted by the Company, DSP Center for Clinical Laboratory undertook the clinical trial on 200 test samples with the Test Kit produced hereby in the clinical study.

II. Purpose

The clinical performance of the Test Kit produced by the Company will be systematically studied to validate its suitability and accuracy in clinical application.

The purpose of this clinical trial is to conduct the comparative experimental study for the same clinical sample with the Test Kit " Malaria pf/pan Rapid Test Kit " produced by the Company and the reference kit ”PhoenixDx” produced by Procomcure Biotech GmbH. Statistical analysis was carried out on the test results to calculate the negative coincidence rate, positive coincidence rate and total coincidence rate. According to the results of statistical analysis, it was validated that the Test Kit is equivalent to the reference kit, to validate the suitability and accuracy of the Test Kit for clinical auxiliary diagnosis.

The results of this clinical trial are an important basis for evaluating the efficacy and safety of the Test Kit.

III. Test Management

1. Introduction to management structure

The clinical trial was conducted by the clinical trialing agency DSP Center for Clinical Laboratory. As the applicant, the Company was responsible for communication and contact during the clinical trial.

2. Quality control in the laboratory

1) All researchers participating in this clinical trial passed the qualification examination and had professional background and capacity related to clinical trial. Before the clinical trial, all researchers had a full understanding of the specific contents about the clinical trial protocol and all indexes through training.

2) The quality control of the laboratory met the requirements of quality control of clinical laboratory to ensure the standardization of test procedure.

3) Quality control before the analysis: Sample collection and treatment was checked for compliance with the requirements and, sample number and other information were checked for correctness.

4) The execution and completion of clinical trial was inspected regularly. The completeness and precision of clinical sample information was checked, and the test results were verified.

3. Statistics and data management

1) All selected cases were filled in the clinical outcome summary sheet, including the subject's sample number, age, gender, and so on. The testers filled the test results of the reference kit and the Test Kit in the clinical outcome summary sheet.

2) After finishing data entry, the main researchers, testers, and applicant jointly reviewed the data and locked the data when they had no doubt.

3) The clinical outcome summary sheet was then sent to analysts for statistics and analysis. The obtained statistics and analysis results were incorporated into corresponding parts of the clinical report.

4. Data preservation

The testing agency and the applicant kept one copy of clinical trial data respectively, including the following contents:

Clinical Trial Agreement, Clinical Test Protocol, Ethics Committee Instructions, Clinical Test Report (testing agency's report), General Report on Clinical Trial, and Clinical Outcome Summary Sheet.

5. Problems found in the study and treatment measures

In clinical trials, when a small number of samples are tested, the results of control samples and test samples are inconsistent. In this case, the clinical quantitative data of these samples or other common clinical strips produced with the same principle are used for re-test.

IV. Test Design

1. Description of overall test design and protocol

With reference to the *Guideline of Clinical Study on In Vitro Diagnostic Reagents,* the appropriate study objects are selected and the PhoenixDx produced by Procomcure Biotech GmbH that was approved for marketing was used as the reference kit to conduct blinding simultaneous comparison, for analyzing the negative coincidence rate, positive coincidence rate and total coincidence rate of the Test Kit and the reference kit.

The trial protocol was to select 200 blood samples as the study objects. Samples were divided into positive group and negative group according to the test results of reference kit. At the same time, the samples were tested with qualitative test strip and reference agent, the test results of the Test Kit and the reference agent were compared and statistically analyzed to calculate the negative coincidence rate, positive coincidence rate and total coincidence rate, so as to judge the clinical suitability and accuracy of the Test Kit, and whether the test result of the Test Kit was consistent with that of the reference kit.

2. Research method

1) Sample collection, storage, transportation

After the samples were collected, they were tested and validated by the protocol.

2) Determination of method for comparison

The samples with inconsistent test results in the test group and the control group can be compared and checked by clinical quantitative results and clinical diagnosis results.

3) Names, specifications, sources, lot number, expiry dates and preservation conditions of the products for clinical study

Product name for clinical study is Malaria pf/pan Rapid Test Kit, and the specification is 25 tests/kit. The product is provided by the Company – Wuhan Jucheng Medical Technology Co., Ltd. The lot number is 20220110, and its shelf- life is 24 months. The storage condition is 4℃- 30℃.

The reference kit is PhoenixDx produced by Procomcure Biotech GmbH, the specification is 96 tests/kit, its shelf-life is 6 months, lot number is PCCSGU15395 and the storage condition is dark place with -20 ℃±5 ℃.

4) Quality control method

The execution and completion of clinical trial is inspected on a regular basis. The completeness and precision of clinical sample information is checked and the test results are verified.

5) Clinical trial method

All test samples were simultaneously tested with the control test strip and the Test Kit, and the test results of the two were compared. When all clinical samples were tested, the recorded test results of the Test Kit and the reference kit were statistically analyzed, to calculate the negative coincidence rate, positive coincidence rate and total coincidence rate and then evaluate whether they were equivalent according to these statistical indexes.

6) Statistical analysis methods for clinical study data

Calculate the negative coincidence rate, positive coincidence rate and total coincidence rate of the test results of the Test Kit and the reference kit. Determine whether each index meets the

clinical evaluation criteria to validate the accuracy and suitability of the product in clinic. Test the Test Kit with different types of samples and statistically analyze the test results. At the same time, test different types of samples of subjects simultaneously with the Test Kit, and compare the test results. When all clinical samples are tested, the recorded test results are statistically analyzed to calculate the negative coincidence rate, positive coincidence rate and total coincidence rate. And then evaluate whether they are equivalent according to these statistical indexes.

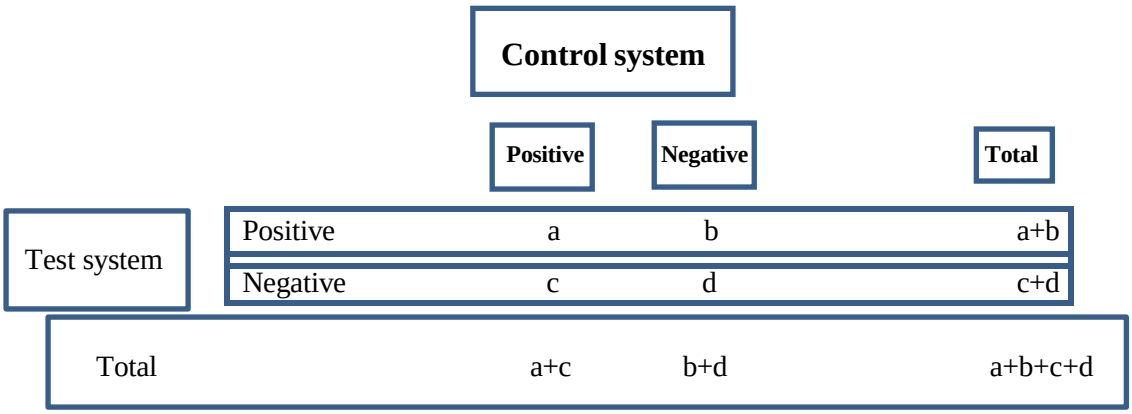
7) Clinical evaluation criteria

Compare the Test Kit with the marketed reference kit to calculate coincidence rate. Product performance shall meet the following requirements.

1) Negative coincidence rate: the proportion of samples whose test results obtained with the Test Kit and the reference kit are negative in the samples whose test results obtained with the reference kit are negative. The negative coincidence rate shall be greater than 90%.

2) Positive coincidence rate: the proportion of samples whose test results obtained with the Test Kit and the reference kit are positive in the samples whose test results obtained with the reference kit are positive. The positive coincidence rate shall be greater than 90%.

3) Total coincidence rate: the proportion of samples whose test results obtained with the Test Kit and the reference kit are the same in the total number of samples. Total coincidence rate shall be larger than 90%.



Generally, formulas of positive coincidence rate and negative coincidence rate are as follows:

Positive coincidence rate = a/ (a+c) * 100%

Negative coincidence rate = d/ (b+d) * 100%

Total coincidence rate = (a+d)/(a+c+b+d) * 100%

If the positive coincidence rate and negative coincidence rate meet the clinical requirements, the two methods or products are equivalent; if the difference between the positive coincidence rate and negative coincidence rate is too large, the clinical protocol shall be redesigned.

8) Modification of the protocol during the study

No modification.

V. Results and Analysis of Clinical Trial

A total of 200 samples were selected. All selected samples were tested.

Make consistency statistics on the test results of Test Kit (test product) produced by the Company and the PCR Kit, analyze their diagnostic sensitivity and specificity, and list them in the form of four-fold table.

Test result of reference kit			
Test Kit	Positive	Negative	Total
Positive	True positive (A)	False positive (B)	A+B
Negative	False negative (C)	True positive (D)	C+D
Total	A+ C	B+D	A+B +C+D

Generally, formulas of diagnostic sensitivity and specificity are as follows:

Diagnostic sensitivity = A/ (A+C) x 100%

Diagnostic specificity = D/ (B+D) x 100%

Total coincidence rate= (A+D)/ (A+B+C+D) x 100%

Table 1: Statistics of Test Results of Test Kit and Reference Kit

	Positive result of reference kit	**Negative result of reference kit**	**Total**
Positive result of Test Kit	97	1	98
Negative result of Test Kit	3	99	102
Total	100	100	200

Item	**Formula**	**Result**	**95% CI**
Diagnostic sensitivity (%)	A/(A+C)*100%	97.00%	83.63%-96.28%
Diagnostic specificity (%)	D/(B+D)*100%	99.00%	95.92%-99.87%
Total Coincidence Rate (%)	(a+d)/(a+b+c+d)*100%	98.00%	

It can be seen from Table 1 that among the 100 samples in the positive group tested with the Test Kit, 97 cases are positive, and 3 cases are negative. Among the 100 samples in the negative group tested with the Test Kit, 99 cases are negative and 1 case is positive. The results show that the negative coincidence rate, positive coincidence rate and total coincidence rate all are greater than 95%, indicating that they are in good consistency with those of the reference kit.

VI. Discussion and Conclusion

(I) Discussion

The Malaria pf/pan Rapid Test Kit strip produced by the Company contains the monoclonal antibody fixed in the test area and the corresponding antigen in the quality control area (C). The rapid test of the antibodies in blood samples is used clinically for auxiliary screening of the patients. The purpose of the clinical trial is to evaluate the clinical performance of the product. The test conditions are presented as follows:

Comparative analysis results of the Test Kit and PhoenixDx produced by Procomcure Biotech GmbH:

Test results of the Test Kit and the reference kit: The diagnostic sensitivity and specificity are greater than 95%, indicating that they are in good consistency with those of the reference kit.

(II) Test conclusion

After validation, the negative coincidence rate, positive coincidence rate and total coincidence rate between the test results of the Test Kit and PhoenixDx produced by Procomcure Biotech GmbH are relatively high, and the results of the statistical analysis also show that there is no significant difference between the test results of the Test Kit and the reference kit, the two methods have good diagnosis consistency and are equivalent. At the same time, the diagnostic sensitivity and specificity of the Test Kit and the nucleic acid test results are both greater than 95%, indicating that they are in good consistency with those of the reference kit.

VI. Description of Special Circumstances on Clinical Studies

There is no special circumstance to be explained in this clinical study.

Dr. MIHAI MAGAZIN
Medic Specialist
Medicină de laborator
Cod: D00213

SOCIETATEA COMERCIALA GENETIC LAB S.R.L. BUCUREȘTI - ROMÂNIA