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The role of VDRL in phirang rog

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Abstract
A perfect scientific medical science by the name of Ayurveda is a gift given by our ancestors which deals with our daily activities to annual activities which help us to maintain our physical and mental health. Many diseases and their treatment have been described by them, some of which are not present today and some still exist. As new diseases arrived in the Indian subcontinent during the period, they came to India through invaders or traders, they were described by our ancestors with their pathogenesis, signs and symptoms, complication, prognosis and treatment. One of them is Phirang rog, which came from Europe, was not found in our scriptures before 16th century. Presently its compatibility with the Syphilis disease can be seen. Timely diagnosis and treatment is necessary to prevent the spreading of disease and the development of irreversible tissue damage. In Syphilis RPR (Rapid Plasma Reagin), VDRL (Venereal Disease Reasearch Laboratory Test), FTA-ABS (Fluorescent Treponemal Antibody Absorption), TP-PA (*Treponema Pallidum* Particle Agglutination assay), Darkfield microscopy are advised for diagnosis. This study has been done to prove the usefulness of VDRL in the diagnosis of Phirang rog.

Keywords: Phirang, syphilis, VDRL, FTA-ABS

Introduction
Since very long time Venereal Disease Research Laboratory (VDRL) test is performed solely by physicians to screen patients for syphilis, yet VDRL is still the most commonly used test all over the world for screening and it still remains unchallenged. Most interesting fact is that it is a test most commonly performed by patients themselves, even in the absence of a medical advice and the result obtained thereof is remembered and reminded to consulting physicians every time they visit them for rest of their life. This holds true when a patient suffers from a disease pertaining to genital system. However, most horrifying fact is that a positive test penetrates so deep into patients' psyche that patient is self-stigmatized and hurt affecting him mentally and physically, affecting his daily activities and performance. Clinically in immuno-competent persons, a negative result is strong evidence against presence of the disease. In human immunodeficiency virus (HIV)-infected persons, the serological response is unusual as in many other diseases, so while interpreting a serological test result, great care should be given. The aim of this article is to prove the usefulness of VDRL in the diagnosis of Phirang rog. Phirang is firstly described by Bhavamishra (16th century) in Bhavaprakash (BPM-9). It is also called 'Gandha Rog' [1]. It occured mainly in Europe and near 16th century has come from Europe (Phirangi desh) so it is named 'Phirang' (comes from Phirangi desh) [2]. Before 16th century it was not prevailing or not notifying in Bharatiya continent. So it is absent in our Ayurvedic litreture before 16th century. Phirang is classified as Agantuja vyadhi which converts into Nija vyadhi after few days [3]. According to Bhavamishra Phirang is transmitted through Female who is suffering from Phirang [1]. Kaviraj Gananath Sen described Phirang in Siddant Nidan [4]. According to him Phirang is more similar to Syphilis. RPR (Rapid Plasma Reagin), VDRL (Veneral Disease Reasearch Laboratory Test), FTA-ABS (Fluorescent Treponemal Antibody Absorption), TP-PA (*Treponema Pallidum* Particle Agglutination assay), Darkfield microscopy are advised for Syphilis diagnosis. Among these VDRL is relatively quick and easy to perform.

Aims: To evaluate the role of VDRL in Phirang rog.

Objectives
1. To study of Phirang.
2. To study of VDRL
3. To evaluate the role of VDRL in Phirang

Materials and Methods

Under this study following steps are adopted-

1. Indication of test
2. Explain the test

- Preparation of the patient.
- Principle
- Procedure
- Risk factors
- Precautions

3. Interpretation

4. How to check clinically on patient by Ayurvedic method.

Materials are collected from different Modern medical and Ayurvedic literature. All collected materials are critically analyzed and a healthy discussion has been done. Finally a fruitful conclusion is drawn.

Phirang

Bhavamishra is the first person who represents the Phirang. He explained the disease which comes from outside the Bhartiya continent on 16th century. Phirang was not preveling or may be not notified before 16th century hence Phirang was not described in Ayurvedic literature before 16th century. He depicts the transmission of Phirang through female who is suffering from Phirang. Kaviraj Gananath Sen illustrates 'Triponima' as a causative organism for Phirang in Siddhant Nidanam. It looks like a whirl of horn and enters the body through abrasions on the sexual part during sexual contact[5]. He represents Phirang into three stages-Prathama, Dvitiya and Treetiya while Bhavamishra describes three types of Phirang - Bahya, Abhyantar and Bahya-abhyantar.

Lakshana

According to Acharya Bhavamishra [6]

- Bahya Phirang – Visphota with mild pain, lesions rupture and turns into vrana, sukhasadhya.
- Abhayantar Phirang - Manifest in sandhi and produces pain, similar to Amavata and associated to shotha.
- Bahya abhayantar Phirang - Like both types.

According to Kaviraj Gananath Sen[7] (mostly similar to Primary, Secondary and Tertiary stage of Syphilis) [8, 9]

- **Prathama Dasha:** It is the stage of ksatotpatti (formation of ulcer), which lasts for 2-3 months. After 2-3 fort–night after sexual contact it gives rise to an eruption on glans penis, labia, mouth, finger or anus, which it turns into the ulcer in a few days. It is similar to tarunasthi to touch, is round, has slight pain and exudation, has warms inside, dries up leaving off dry scales. Near the lesion lymphglands become swollen resembling betelnut but do not ripe, if properly treated the ulcer heals well.
- **Dvitiya Dasha:** It starts after six months involving the skin, muscles and bone which continues for one or two years if not promptly treated. Visha spreads to all the dhatus and ashaya and produces mandalas on the skin with irregular borders, multiple in number and without itching, prominently in foreleg, abdomen, palms but not on the face. Vishamjwar, severe headache at night, enlarged- hard –moveable lymphglands, hair fall, emaciation, paleness, falling off teeth and sometimes Kotha (Gangrene) of hanu sandhi (lower jaw). Develops Chatrak arsas (*Condylomata lata*) which are cirkari and resemble the head of Shilindhra (mushroom) produced by shelshama on lips, anuus, vagina, penis. In some cases nasal bridge may break.
- **Treetiya Dasha:** It is very dreadful, difficult to diagnose and treat. It continues for twenty or thirty years, this period depending on less or more amount of toxins. During this period, toxin blisters develop externally or internally called Visha gandak. The internal Visha gandak (abcess) may present in lungs, liver, spleen, vertebral column, brain, spinal cord.

Complication: Lean and thin body, loss of immunity, depressed nose, loss of appetite, loss of bony tissue, bowing of legs [10], tremors on one side, convulsion, Insanity, difficulty in speech, depression, shoth in great vessels of heart [11].

Prognosis [12]

- Sadhya – Bahaya, new and less complicated Phirang.
- Kastsadhaya - Abhayantar Phirang.
- Asadhya – Bahaya abhayantar, old, sarvasharirgata and more complicated.

VDRL (Venereal Disease Research laboratory test) [13, 14]

It is Nontreponemal tests for detecting syphilis. The VDRL test checks for the patient's antibodies make in response to antigens *Treponema pallidum*, produced by cells damaged by the bacteria in Syphilis, a sexually transmitted infection (STI). The principle of the VDRL test is the agglutination reaction between the VDRL antigen and the reagent. The reaction may be seen macroscopically on the test slide as the clumping of the carbon particles.

Early symptoms that may prompt to order this test includes:

- One small, painless sore
- Swelling in lymph nodes near the sore
- A skin rash that doesn't itch

No special preparation is needed.

Requirements- Patient's Serum, Water bath, Freshly prepared cardiolipin antigen, VDRL slide, Mechanical rotator, Pipettes, Syringe with unbeveled needle, Microscope, Known reactive and non-reactive serum controls

A blood sample is needed but can also be done using a sample of spinal fluid in Neurosyphilis.

Blood sample collection

- Technician may tie a tourniquet, above the injection site before inserting the needle to easier withdrawal of blood.
- Inserting a hollow needle into a vein in the elbow or on the back of the hand.
- The blood flows into an airtight collection tube attached to the other end of the needle.

CSF sample collection

1. Collect samples of CSF through a lumbar puncture or spinal tap.
2. During the procedure, a person will lie on their side and pull their knees towards their chest.
3. Technician will disinfect and numb the injection site with a local anesthetic.
4. Then he\she will insert a spinal needle into the lower spine, which they use to extract a small quantity of CSF.

The test can be performed both quantitatively and qualitatively

Qualitatively Test

- Patient's serum is inactivated by heating at 56 deg c for 30 mins in a water bath to remove non-specific inhibitors.

- VDRL antigen suspension controls and samples are brought to room temp.
- One drop (50 µl) of the test specimen positive and negative controls is pipette onto separate reaction circles of the disposable slide.
- A drop of diluted antigen suspension is added to the measured volume of specimen, positive and negative controls.
- Using a mixing stick the test specimen and the VDRL reagent is mixed uniformly over the entire reaction circle.
- The slide gently rotated and continuously either manually or on a mechanical rotator at 180 rpm.
- Flocculation is checked microscopically using 10x objective and eye piece at about 8 mins.

Quantitative Test

- Dilute serum sample to an endpoint titer. Quantitative tests for 3 serum specimens through the 1:8 dilution may be performed on one slide.
- Place 50 µl of 0.9% saline in circles numbered 2 through 4. Don't spread saline.Place 50 µl of serum in circle 1 and 50 µl of serum in circle 2.
- Mix the saline and the serum in circle 2 by drawing the mixture up and down in the safety pipette 8 times.
- Transfer 50 µl from circle 2 (1:2) to circle 3 and mix.
- Transfer 50 µl from circle 3 to 4, mix and then discard the last 50 µl. Gently re- suspend the antigen suspension.
- Add exactly 1 free falling drop (17 µl) of antigen suspension to each circle.
- Place the slide on the mechanical rotator. Rotate the slide for 4 mins at 180+_2rpm.
- Immediately after rotation, read the test.
- If the highest dilution tested (1:8) is reactive, then continue as follows-

1. Prepare a 1:8 dilution of the test specimen in a test tube.
2. Add 0.1 ml of serum to 0.7 ml of 0.9% saline.Mix thoroughly.
3. Place 50 µl of 0.9% saline into paraffin rings 2, 3 and 4. Prepare additional serial dilutions for strongly reactive specimens.
4. Add 50 µl of the 1:8 dilution of the test specimen to paraffin rings 1 and 2.
5. Prepare serial twofold dilutions beginning with ring 2.

Test Procedure for Cerebrospinal fluid (VDRL-CSF)
Preparing the Sensitized Antigen Suspension
Prepare the VDRL antigen suspension as described for the VDRL slide tests on serum. Add one part of 10% saline to one part of VDRL antigen suspension. Mix by gently rotating the bottle or inverting the tube. Allow the mixture to stand for at least 5 minutes. The sensitized VDRL-CSF antigen suspension is good for only 2 hours after preparation.
The test can be performed both quantitatively and qualitatively

Qualitative test
Holding the VDRL-CSF sensitized antigen suspension dispensing needle (21-or 22-gauge) and syringe in a vertical position, dispense exactly 1 free-falling drop (10 µl) of sensitized antigen suspension to each slide concavity that contains 50 µl spinal fluid. Rotate the slide for 8 minutes at 180 ±2 rpm. Read under a microscope. Report the results as follows.

Reading Report
Definite clumping of any degree- Reactive (R)
No clumping or very slight roughness- Nonreactive (N)

Quantitative Test
Test each specimen undiluted and in 1:2, 1:4, and 1:8 dilutions or more.
People who have a high risk of syphilis may want to consider getting routine screening tests about every 3 months.
Risk factor- VDRL test is safe and convenient. Only minor risk involved with having blood taken.
Other risks includes - Excessive bleeding, Fainting or feeling lightheaded, Multiple punctures to locate veins, Hematoma (blood accumulating under the skin), Infection (a slight risk any time the skin is broken)

Result
This is sensitivity to detect syphilis nears 100% during the middle stages; it is less sensitive during the earlier and later stages.
Medium or large clumps - Reactive (R)
Small clumps - Weakly reactive (W)
No clumping -Nonreactive (N)

- Negative test - Normal (No antibodies to syphilis have been seen in given blood sample), Early and late-stage syphilis (Confirm the condition with another test)
- Positive test - Secondary and latent stages of syphilis
- False-positive results: HIV, Lyme disease, malaria, pneumonia (certain types only), systemic lupus erythematosus, IV drug use, tuberculosis.
- False-negetive results: In some cases, if body may not produce antibodies even if patient has been infected with syphilis. So the VDRL test will be inaccurate.

Cerebrospinal Fluid
A reactive VDRL test on CSF, free of blood or other contaminants, usually suggests past or present syphilis infection of the central nervous system. A biologic false-positive VDRL test result for syphilis is rare in spinal fluid. A nonreactive VDRL test on CSF may indicate that the patient does not have neurosyphilis. However, a negative result may occur in some serum from neurosyphilis patients.

Precaution
Should be taken some standard laboratory precautions like sterilization of required materials and about complete disposal of biomedical materials.

Interpretation
How to check on patient
Based on clinical sign and symptoms of Phirang (Bhavaprakash, Siddhant nidana) [6, 7]

- Primary stage - Prathama Dasha/ Masat/ Ardha masat/ Pakshat Dasha, Bahya Phirang
- Secondary stage - Dvitiya Dasha, Abhyantar and Bahya abhyantar Phirang
- Tertiary stage - Treetiya Dasha, Bahya abhyantar Phirang
- Complication - Upadrava Avastha

This is sensitive to detect syphilis nears 100% during the middle stages (Abhyantar and Bahya abhyantar Phirang/ Dvitiya Dasha); it is less sensitive during the earlier and later stages (Masat/ Ardha masat/ Pakshat/ Prathama Dasha and Treetiya Dasha or Bahya Phirang).

- Negative test Normal (no antibodies to syphilis have been seen in given blood sample) Early and late-stage syphilis (Masat/ Ardha masat/ Pakshat/ Prathama Dasha and Treetiya Dasha or Bahya Phirang) confirm the condition with another test.
- Positive test - Secondary and latent stages of syphilis (Abhyantar and Bahya abhyantar Phirang or Dvitiya Dasha)

Discussion

Phirang is firstly described by Bhavamishra on 16th century in Bhavaprakash. Bhavaprakash madhyam khand 59, defines Samprapti (pathogenesis), Bheda, Lakshana, and Upadrava (complication) of Phirang. This disease is named Phirang because occurrence of this disease is more in Europoean country (Phirang desh). In Bruhattrayee absence of this disease shows before 16th century Phirang was not prevailing in Bharat. Features of Phirang rog is similar to Syphilis. Syphilis is a sexually transmitted disease, caused by the bacterium *Treponema Pallidum*. Finding and treating the infection early can also prevents from spreading of Syphilis. RPR (Rapid Plasma Reagin), VDRL (Venereal Disease Reasearch Laboratory Test), FTA-ABS (Fluorescent Treponemal Antibody Absorption), TP-PA (*Treponema Pallidum* Particle Agglutination assay), Darkfield microscopy. Above these only RPR and VDRL test is usually included under screening test for Syphilis. VDRL can be done on blood and spinal fluid. It is relatively quick and easy to perform. The VDRL test doesn't detect the presence of bacteria in blood, it checks the antibodies in patient which is present in reponse to antigens produced by cell damaged by the bacteria. In VDRL test presence of symptoms of Syphilis is not necessary only infection of Syphilis which initiates antibody production is required. In this study the role of VDRL test is evaluated in Phirang Rog. This is sensitive to detect Syphilis/Phirang nears 100% during the middle stages or Abhyantar-Bahya abhyantar or Dvitiya Dasha of Phirang; it is less sensitive during the earlier and later stages or Masat/ Ardha masat/ Pakshat/Prathama Dasha and Treetiya Dasha as Siddhant Nidana and Bahya Phirang as Bhavaprakash.

Conclusion

By this study we can correlate the features of Phirang and Syphilis. According to Acharya Bhavamishra Bahya Phirang is more similar to Primary stage of Syphilis and Abhyantara, Bahya-Abhyantara Phirang is more similar to Secondary and Tertiary stage of Syphilis. Features of Upadrava of Phirang is as like as Tertiary stage of syphilis. According to Kaviraj Gananath Sen Phirang is more similar to Syphilis. Finally we can conclude that VDRL is easily be a tool of investigation in Phirang diagnosis. By using this test we can prevent the spreading of disease and the development of irreversible tissue damage.

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