Some Facts About Pneumocystis jirovecii

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ABSTRACT

Pneumocystis Jirovecii is a unique yeast-like fungus *(*previously named *Pneumocystis carinii*). It has been demonstrated as a predominant cause of intrapulmonary and rarely extra-pulmonary infection and a significant cause of morbidity and mortality in immunocompromised patients with and without AIDS. *Pneumocystis jirovecii* was classified previously as protozoan endoparasite, and then reclassified as a fungus, based on nucleic acid analysis and biochemical characteristics. No successful cultural method for the organism had been preceded yet. Direct microscopic identification of the organism in biopsies or inspired materials is the method of choice for diagnosing the organism. Advanced molecular techniques offer a high sensitivity and specificity in *P. Jirovecii* diagnosis. Sulfa-based medicines have been shown effectiveness in *P. Jirovecii* infections treatment.

Aim of this review to provide an updated knowledge on the role of *P. jirovecii* as a human pathogen.

Keywords : Immunocompromised patients , pathogenic Fungi , *Pneumocystis jirovecii* , *Trypanosoma spp .*

بعض الحقائق عن المتكيسة الرئوية الجؤجؤية

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الخلاصة

Pneumocystis Jirovecii "المتكيسة الرئوية الجؤجؤية" كائن فطري استثنائي, شبيه بالخميرة ،سمي سابقا ب *Pneumocystis carinii* عرف كمسبب مرضي شائع للانتانات الرئوية وخارج الرئوية احيانا, يمكن ان يسبب الموت للاشخاص منقوصي المناعة سواء بسبب الاصابة بالايدز او لاسباب اخرى. صنف سابقا على انه طفيلي داخلي وحيد الخلية, واعيد تصنيفه لاحقا على انه كائن فطري بالاعتماد على تحليل الاحماض النووية والخصائص الكيموحيوية. لم يتم التوصل الى وسط زرعي كفوء لتنمية هذا الكائن في الزجاج حتى الوقت الحاضر. الطريقة المثالية لتشخيص الاصابة بهذا الكائن تعتمد على الرؤية المجهرية المباشرة للكائن المجهري في خزعة نسيجية او المحتوى المسحوب من منطقة الاصابة. الطرق الجزيئية الحديثة يمكن ان تكون ذات دقة وحساسية عاليتين في تشخيص الاصابة بهذا الكائن. المستحضرات الدوائية المعتمدة على الكبريت اثبتت كفاءة عالية في علاجه.

الهدف من مقالة المراجعة هذه توفير معرفة محدثة عن دور المتكيسة الرئوية الجؤجؤية كممرض بشري

الكلمات المفتاحية : مرضى منقوصي المناعة، فطريات مرضية، المتكيسة الجؤجؤية، جنس المثقبيات.

INTRODUCTION

History of Pneumocystis

Pneumocystis spp. was originally identified by Carlos Chagas in 1909 as a lungs pathogen of rats and guinea pigs. Chagas's theory depends on the morphological features of *Pneumocystis* species, in addition to its resistance to antifungal agents and susceptibility to antiprotozoal therapy[1]. Antonio Carini and Delanöes noted cysts of *Pneumocystis* in rats infected with Trypanosomiasis but they assumed that its influence to be disparate to *Trypanosoma cruzi* organism[2]. Pierre and Marie Delanoës ordered *Pneumocystis* as a different species in 1912, and then still assumed to exist as a *Trypanosoma cruzi*[3]. Vanek and Jirovec found a new disease of interstitial plasma cell pneumonia that ensued in epidemics in untimely and starving infants in

packed European orphanages throughout and subsequently World War II[4]. In the 1960s and 1970s *Pneumocystis* was establish as an significant source of fatal pneumonia, *Pneumocystis pneumonia* (PCP), in patients who has crucial immunodeficiency complaints or that receiving immunosuppressive agents in cases like cancer, organ transplantation and other disorders[5]. *Pneumocystis jirovecii*, remained relatively infrequent source of sickness till the discovery of acquired immune deficiency syndrome (AIDS) in the early 1980s, when PCP was the furthermost mutual proven manifestation of this new disease[6].

Investigations showed that *Pneumocystis* is a necessitate extracellular fungal pathogen of low virulence and be existent in cyst and trophozoid stages. *Pneumocystis* has been fixed in fungi kingdom of Ascomycota phylum, belongs to opportunistic yeast-like fungi with separate genus and a new species with a distinctive tropism for the lungs, referred as *Pneumocystis jirovecii*. It was alleged that there was merely one strain belong to *Pneumocystis* species which is accomplished of infecting diverse mammalian hosts[7].

In 1999, the organism that caused human Pneumocystis pneumonia (PCP) is named *Pneumocystis jirovecii;* in honor of the Czech parasitologist Otto Jirovec who was the first described the microorganism in humans. In addition, he distinguished the organism in human from physiological variants of *Pneumocystis* spp. in animals[8].

By means of molecular performances, it takes remained that there are many different species belong to the genus *Pneumocystis* organism. This could be due to the fact that each verified primate counting humans seems to have their particular species of *Pneumocystis* that is unqualified to cross infected with other host species and has coevolved with every mammal species[6].

Pneumocystis spp. ought to official termed with *P. jirovecii* from human, *P. murina* from mice, *P. wakefieldiae*, and *P. oryctolagi* from rats, and *P. oryctolagi* from rabbit [9,10]. *Pneumocystis jirovecii,* species specific to human has been shown capable of infecting human only[11]. Facts were achieved by polymerase chain reaction (PCR) technique as no amplification was detected when primers from non - human with human clinical isolates were tested [8].

Pneumocystis Jirovecii Life Cycle

Over years, the life cycle of *Pneumocystis* has been a significant challenge. This might be due to the circumstance that conservational reservoir has still not been distinct. *P.* jirovecii transient proliferation was achieved especially in lung epithelial tissues [12]. Its lifetime phase is consist of asexual and sexual stages however, there has been reported of at least two forms exist in *Pneumocystis* life cycle, trophic form and cyst [13].

The small (1–4μ) haploid forms "trophic forms" or "spores" heterogonous in contour and is walled only by a plasma membrane with no inflexible cell wall reproduce asexually by double fission [14] . During sexual life stage, two trophozoite forms companion and eventually change into a mature (5–8μ) "cyst" or "ascus" with eight nuclei. The nuclei either transform to trophozoite or spore stage to, which has the ability for excystation or restart the organism life cycle [15] .

β-1,3 glycan with of chitin made the cyst wall to be rigid, and more uniform in shape than the trophic form and contains up to eight intracystic bodies[8]. The third form precystic stage (sporozoite) has infrequently encountered which represents an intermediate stage between cystic and trophic form [16] . P. jirovecii cystic and trophic forms have developed a unique mechanism which allows them to evade immune recognition, then mimic and maintain its existence in the host [17].

Aerosolized particles containing trophic form can transmit from infected host to host, via infected air droplets from respiratory samples which represent the main source of infection with P. jirovecii [18]. Morris and Coworker, 2012 [19] showed that another source of infection can occur throughout childhood, by reactivation during periods of immune suppression by detection anti-*Pneumocystis* antibodies with positive PCR test of normal healthy immunocompetent child at age 3 or 4 years supports the theory of early exposure to this organism.

Lungs has been found to be the main site for P. jirovecii infection; when inhaling a trophic form they reside in lung alveolar epithelial cells and rarely disseminate to other organs. However extra pulmonary dissemination has been recounted [20] .

What Is Pneumocystis Pneumonia?

Pneumocystis pneumonia (PCP) refers to the infection that is caused by P. jirovecii, which occurs in different forms according to Stringer and Coworkers 2002[8] which include asymptomatic infection, pneumonia in immunocompromised patients, and extra pulmonary infection. In 1980, PCP had been denoted as the AIDs defining disease in patients with HIV infection in the United States. It has also been described as the leading origin of death in such patients. It occurs mainly when there is a defect in T-helper cell (CD_4) especially when its count is less than 200 cell per cubic millimeter, PCP can occur in any patients with non-AIDs immunosuppression state [21,22] .

Pathogenesis of P. Jirovecii:

Infections caused by P. jirovecii is more frequent in patients with defects exist mainly in cellular immunity. When breathe P. jirovecii in trophic form it is attached to the lung cells and starts high rate replication and gradually fills the alveoli [23]. Gas exchange disruption may occur due to direct attachment of the microorganism to alveolar epithelial cells [20]. In addition, cell debris is associated with frothy exudate in the alveolar lumen, and both may form an insulating layer. As the infection develops, the interface of P. jirovecii through lung epithelial lining may block lung restoration cells and inhibiting epithelial proliferation [24]. When pneumonia becomes more severe, fluid can accumulate and tissue scaring may ensure [25]. These changes have been found to result in decreased respiratory function associated with lower level of oxygen in blood. The two stages of P. jirovecii were found in the infected lung tissue simultaneously [23] .

Epidemiological Features and Risk Factors:

Pneumocystis can infect humans and animals with numerous strains have been identified even in a single species. *Pneumocystis* DNA has been observed in air, water, and soil samples[24]. *Pneumocystis* spp. may persist in the atmosphere extended enough to pass on the disease to a susceptible host. *Pneumocystis* human to human transmission can occur; evident by outbreaks in oncology and transplant units[6]. The microorganism can be transmitted via air born droplets from respiratory samples and reside in alveolus as the main site of infection. Some centers have advocated isolating the patients with PCP to prevent transmission to other susceptible patients [25] .

Anciently, *Pneumocystis* infection was attained throughout childhood and then infections occurred by recrudescence of the dormant *Pneumocystis* infection when the host's immune system failed to kill the microorganism. This theory has been accepted for decades. In addition, it has been reported that people who do not show signs of infection may be asymptomatic carriers of the organism [26]. *Pneumocystis* mechanism of transmission has importance in clinical and public health implications. The infection is either asymptomatic or revealed as a slight respiratory infection in immunocompetent host. *Pneumocystis* infection has shown a global distribution. This has evident by serological studies that have shown that about 80% of children developed antibodies to *pneumocystis* organism[13].

Cisse and Coworkers, 2012 [28] showed that genome sequencing of *Pneumocystis* naked a deficiency of virulence genes and the absence the metabolic enzymes and initiate as a free-living form dissimilarity to other human fungal pathogens, which grow leisurely and noninvasively in its accommodating host. This pointed out that *Pneumocystisspp.* has been modified to survive and disseminate inside lung of human and other mammalian hosts [13].

Risk factors for *Pneumocystis* colonization and subsequent infection include pulmonary diseases such as obstructive pulmonary diseases, cigarette smokers, pregnancy, young children with respiratory symptoms, human immunodeficiency virus infection (HIV), cancer, organ transplantation, autoimmune diseases [29].

Methods for Diagnosing PCP Infections:

Success diagnosis of PCP infections is almost depending on different diagnostic methods, some of them are essential; others are subsidiary, as shown below:

1- Clinical manifestations and radiology:
Immunocompromised patients with PCP can be presented with progressive dyspnea, non-productive cough, fever, respiratory distress with hypoxia, and tachycardia [30]. Chest X-ray for immunocompromised patients with PCP showed a bilateral peri-hilar interstitial infiltrate that has become increasingly homogenous and diffused as the disease progressing[31]. However, chest radiograph finding was found normal in one third of the cases[29]. In addition, higher-resolution computed tomography, has supplementary delicate other than chest radiography [32].

2-Pulmonary function test:
This test should be obtained as part of the initial workup in patients with suspected PCP [33].

3-Enzymatic methods:
Estimation of the serum level of *Lactate dehydrogenase test* LDH has represented the most useful test during active infection. High serum levels up to around 460 IU/L in patients with PCP have been tested as part of the initial workup in the diagnosis of PCP[29]. Lactate dehydrogenase is non-specific also raised serum levels of LDH have been come in other condition counting bacterial pneumonia, and tuberculosis[32].

4-Microscopical examination (direct identification):
A traditional method for morphological detection of the microorganisms that could not cultivated in the culture media. The stain used for microscopical examination of *P. jirovecii* depends on cyst or trophic form present in clinical specimens[33]. If sample is obtained from upper respiratory tract, only few organisms can be found in comparison

with induced sputum and broncho-alveolar lavage (BAL) obtained from lower respiratory tract[30]. Trophic form can be inspected by Gram-Weigert or Giemsa stain, while cyst form can be detected with Grocott's methenamine silver stain (GMS), toluidine blue O, and Gram-Weigert, These stains colored the wall of the cyst. However Giemsa, polychrome methylene blue, and Gram-Weigert stain the internal contents of the cyst and the trophic forms[18].

5-Immunodiagnosis:
Immunofluorescent stains include two methods, direct and indirect fluorescent antibody stains. Monoclonal antibody detection is considered basic for direct florescent which has a higher sensitivity in staining[34]. Cyst and trophozoite stages could be stained by direct fluorescent technique. Otherwise, indirect antibody technique depending on polyclonal antibodies, in which detection of *Pneumocystis* spp. is easy, rapid, and sensitive immunofluorescence assay[35],[36].

6-Moleculer biology:
Polymerase chain reaction (PCR); this method is depending on detection of *Pneumocystis* DNA after amplifying various genetic loci[37.] It is often used to insure the sensitivity of BAL fluid and induced sputum than other noninvasive oral washes. Multi copy gene target (such as the Msg or the mitochondria large-subunit rRNA [mtLSU] gene) are increase sensitivity of the method. However, detection sensitivity was shown to increase by employing two rounds of PCR[32].

7-Cultural methods:
Although many researchers have attempted to cultivate and grow the primary isolates of *P. jirovecii* that was extracted from mammalian hosts by using monolayer cell tissue cultures and artificial media, only a limited replications could be provided following inoculation. Moreover, the growth of the organism has declined after a few passage in these systems [36] .

Treatment

1-Antimicrobial agents:
Initiation of systemic antimicrobial treatment of *P. jirovecii* pneumonia influenced by clinical signs and symptoms. Treatment courses can be ranged from 14-21 days which depends on the intensity of the infection and the immune status of the patients.
The agent of choice for treating PCP, regardless of underlying situations, is Trimethoprim-sulfamethoxazole (TMP-SMX) [37]. TMP-SMX has been documented to be effective superiorly to other lines and has comparable potency to parenteral pentamidine with minimal adverse effects [38] . Oral dose of 160 mg TMP and 800 mg SMX three times a day has been recommended for mild form of the disease. However, intravenous route of administration is favored for the severer form of the disease. A dose of 15-20 mg TMP and 75-100 mg SMX per kilogram body weight every 6 - 8 h has been used successfully for such cases[٣٩]. When clinical steadiness has been achieved, treatment can be converted to oral therapy [40].
What has been concluded from reviewing the literature is that TMP-SMX combination is the most likely to result in treatment cessation due to serious adverse effects such as fever, pancytopenia and dysfunction of the renal system [41]. More serious toxic effects were commonly observed in HIV patients and may necessitate switching to another agent[17]. Better patients' tolerance has been reported with dapson-TMP, pentamidine inhalation and atovaquone [39].
Intravenous clindamycin with primaquine taken orally is also prescribed for patients with moderate to severe PCP when TMP-SMX is intolerable [8,42]. In 2020, a meta-analysis systematic review study has been conducted by Butler-Laporte and his colleagues [43] and found that the lower doses of TMP-SMX had a remarkably lower incidences of adverse effects resulting in treatment termination. Therefore, lower doses of TMP-SMX may consider as an optimal option. Inhaled pentamidine advantageously taken monthly has a common side effect of causing cough and respiratory spasm [39]. Moreover, Bactrim (co-trimoxazole) has been recommended for moderate cases in patients with good oral absorbance. Clindamycin at doses of 600-900 mg given intravenously with primaquine (15 to 30 mg by mouth per day) is an extra alternate for moderate to severe cases[44]. Atovaquone may be considered for patients with PCP in dose of 750 mg orally given twice daily.
Although no studies have looked at pregnancy and breastfeeding with PCP, the most acceptable regimen for pregnant patients is TMP-SMX. Benefits are thought to overcome risks of side effect (<1 % congenital malformation) associated with TMP-SMX [39]. Intravenous pentamidine, clindamycin-primaquine, atovaquone, and dapsone-TMP could be considered as alternative approach in pregnancy. If dapsone and primaquine have simultaneous with glucose-6-phosphate dehydrogenase deficiency, hemolytic anemia can occur in the newborn and patients with G6PD [45, 46]. More insight into the treatment trial of PCP in pregnancy and breastfeeding should be considered by regulatory agencies for better outcome. Newer agents with antifungal effects have been studied as treating or prophylactic against PCP[47]. For instance, echinocandins has

demonstrated activity against the cyst stage but not the trophozoite [21].

2- Corticosteroids, The anti-inflammatory characteristics of the corticosteroids make it beneficial to use them adjuvant with other medications in patients with PCP. In HIV-infected patients with PCP, hypoxemia and subsequently mortality had been shown to reduce in response to using corticosteroids[48]. However, in an unselected population with no respiratory failure, the use of corticosteroids might be detrimental and connected with amplified death toll[49]. Tapered dosing of prednisolone can be given orally starting with 40 mg twice a day for 5 days followed by reducing the dose to 40 mg once a day for another five days and then half it to 20 mg per day for eleven days [39].

CONCLUSION

Pneumocystis jirovecii is an opportunistic fungus that has recently discovered to have global distribution. P. jirovecii has been documented as a considerable agent of pulmonary infections in immunocompromised individuals, particularly those with AIDS. Developing of rapid successful diagnostic and treatment strategies for this infectious disease is a future prospective.

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