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【 CASE REPORT 】

Locked-in Syndrome Due to Meningovascular Syphilis: A Case Report and Literature Review

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Abstract:

We herein report a 46-year-old man presenting with locked-in syndrome secondary to meningovascular syphilis. Brain magnetic resonance imaging (MRI) demonstrated multiple acute infarctions in the left ventromedial pons, right basis pontis, and left basal ganglia. His locked-in syndrome was hypothesized to have been caused by thrombosis of the small paramedian branches of the basilar artery due to syphilitic arteritis. This is a unique case of bilateral ventromedial pontine infarction caused by meningovascular syphilis that presented as locked-in syndrome. Meningovascular syphilis should be included in the differential diagnosis of uncommon stroke, particularly in young men.

Key words: neurosyphilis, meningovascular syphilis, brainstem infarction, locked-in syndrome, young adult, uncommon stroke

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Introduction

Neurosyphilis refers to infection of the central nervous system (CNS) by the spirochete *Treponema pallidum*, and it occurs within the first year after infection, even though CNS involvement is possible at any stage of the disease (1-4). While long-term sequelae of the disease include dementia paralytica and tabes dorsalis, early neurosyphilis can cause meningitis, gumma, and stroke due to meningeal vasculopathy.

Locked-in syndrome is a rare neurological condition characterized by quadriplegia and mutism but with a preserved consciousness (5, 6) wherein patients can sometimes communicate through eye movements or blinking. This catastrophic disorder is typically caused by bilateral infarction of the ventromedial pontine.

We herein report a man with locked-in syndrome due to meningovascular syphilis who was human immunodeficiency virus (HIV)-negative.

Case Report

A 46-year-old left-handed Japanese man was admitted to another hospital for acute-onset right-sided spastic hemiparesis. His recent history included mild fatigue, pyrexia, and severe headaches accompanied by exacerbation of nausea and anorexia for the preceding three weeks. He was transferred to our hospital two days after the onset of right hemiparesis due to increasing muscle weakness, progressive and severe dysarthria, and dysphagia.

On admission, he was slightly febrile (temperature, 37.1°C), blood pressure was high at 200/128 mmHg, and pulse was regular at 61 bpm. He had a smoking habit (15 cigarettes/day for 29 years), and his medical history included gastric ulcer and alcoholism; however, there was no history of hypertension, diabetes, dyslipidemia, atrial fibrillation, or heart disease.

A physical examination revealed the presence of well-demarcated erythematous papules and plaques with scaling on the trunk, the abdomen, and the extremities. On a neurological examination, he was awake, alert, and able to com-

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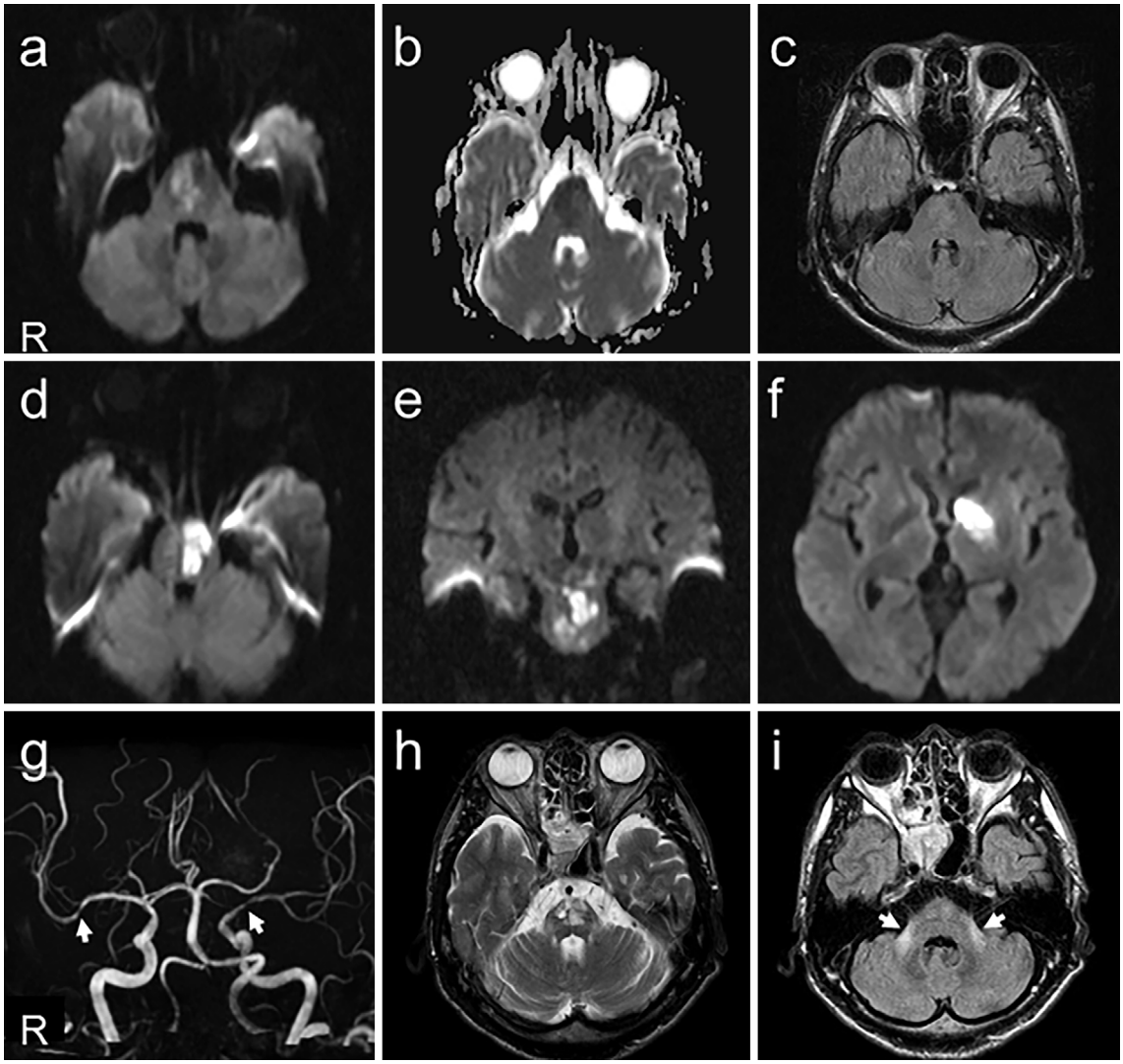

Figure 1. Brain MRI and MRA findings. On the day of admission, brain MRI showed high-intensity lesion in the left ventromedial pons and slight high-intensity lesion in the right ventromedial pons on diffusion-weighted imaging (DWI) (a, d, e), with low-intensity findings noted on an apparent diffusion coefficient (ADC) map (b), high-intensity findings on fluid-attenuated inversion recovery (FLAIR) imaging (c), and high-intensity findings in the left basal ganglia, including the caudate nucleus on DWI (f). MRA showed narrowing of the bilateral middle cerebral artery (MCA) (g, arrows). Eight months after the onset of infarction, follow-up brain MRI showed an old infarction in the bilateral ventromedial pons on T2-weighted imaging (h). High-intensity lesions in the bilateral middle cerebellar peduncles, where the pontocerebellar tract runs, suggested Wallerian degeneration secondary to a pontine infarction (FLAIR) (i, arrows).

municate with a nodding motion but unable to speak. He displayed quadriplegia with preserved full eye movement and blinking, and the muscle strength in his extremities was 1/5. Hyperreflexia was present in all extremities along with bilateral positive Babinski's sign. At best, sensory testing is difficult to perform accurately, and on day 11 of hospitalization, his neurological condition worsened significantly, with only consciousness, breathing, eye movement in all directions, and blinking preserved; no movements in the limbs, mouth, or tongue, including swallowing movements, were observed. Therefore, a nasogastric tube was inserted to deliver nutrients to the patient.

Brain magnetic resonance imaging (MRI) on admission day revealed a high-intensity lesion in the left ventromedial pons, along with a small high-intensity lesion in the right basis pontis and in the left basal ganglia, including in the caudate nucleus, on diffusion-weighted imaging (DWI) (Fig. 1a, d-f). Considering the apparent diffusion coefficient map and fluid-attenuated inversion recovery (FLAIR) imaging, these findings were consistent with acute cerebral infarctions. Magnetic resonance angiography (MRA) showed a normal basilar artery flow but bilateral partial stenosis of the middle cerebral artery (MCA) (Fig. 1e). Brain MRI acquired after neurological deterioration revealed a high-intensity lesion in the right basis pontis. Contrast-enhanced MRI showed no abnormal findings specific to meningovascular

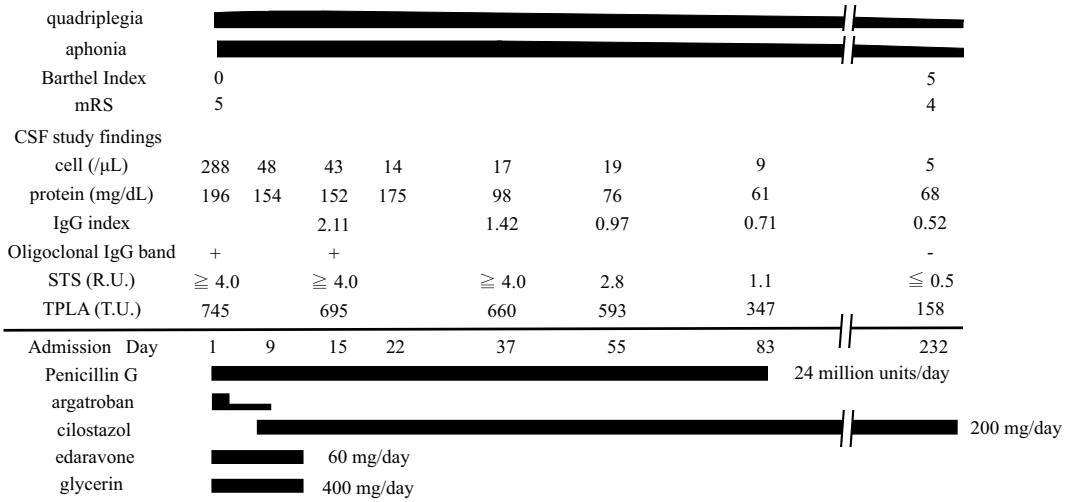

Figure 2. Clinical course of the patient. mRS: modified Rankin Scale, CSF: cerebrospinal fluid, STS: serological test for syphilis, TPLA: *Treponema pallidum* latex agglutination

syphilis, except for findings consistent with the natural course of cerebral infarction in the left basal ganglia. MRA showed a normal basilar artery flow similar to the previous findings. Consciousness was clear during the clinical course. Although an electroencephalogram test was performed, it was not useful for evaluating the background activity because of the inability to exclude artifacts. An electrocardiogram and chest X-ray findings were unremarkable, and a serum test for HIV antibodies was negative. Screening for hepatitis B virus (HBV) serology displayed the following pattern: hepatitis B surface antigen (HBsAg)-negative, anti-hepatitis B surface antibody (anti-HBsAb)-positive, antihepatitis B core antibody (anti-HBcAb)-positive, and HBV-DNA not detectable by real-time detection polymerase chain reaction.

Routine laboratory evaluations revealed an elevated white blood cell count (17,900/μL; neutrophil 88.8%) and C-reactive protein levels (0.80 mg/dL). A serological test for syphilis [STS; cut-off point: 1 rapid plasma reagin units (R.U.)] and *T. pallidum* latex agglutination [TPLA; cut-off point: 10 titer units (T.U.)] test were positive at 64 R.U. and 14,347 T.U., respectively. A cerebrospinal fluid (CSF) analysis was remarkable for the presence of cells (288/μL), mainly lymphocytes, with high levels of protein (196 mg/dL), glucose (30 mg/dL), STS (8 R.U., cut-off: 1 R.U.), and TPLA (745 T.U., cut-off: 10 T.U.). Both serum and the CSF were positive for the fluorescent treponemal antibody-absorption test. An elevated CSF immunoglobulin G (IgG) index (2.11; cut-off <0.7) with positive oligoclonal IgG bands (OCB) was also detected, suggesting intrathecal production of anti-treponemal antibodies. Polymerase chain reaction for herpes simplex virus and varicella zoster virus was negative, and no findings indicative of cardiogenic cerebral embolism were present. Based on the above, the patient was diagnosed with meningovascular syphilis, and Fig. 2 shows the clinical course of the patient.

He was treated with intravenous penicillin at a dose of 4 million units, 6 times a day, for 12 weeks and was prescribed intravenous argatroban, edaravone, and glycerin for cerebral infarction, followed by oral cilostazol tablets. We did not administer steroids due to concerns of deep-vein thrombosis complications and HBV reactivation. Both the IgG index and OCB normalized gradually and in parallel with improvements in CSF parameters.

Four months after the onset, he required full assistance for his daily activities, and he was transferred to the Department of Rehabilitation Medicine after penicillin therapy. During follow-up at 8 months after the diagnosis and treatment for neurosyphilis, the CSF parameters, including CSF IgG index (0.52), had reverted to normal values, except for a slight elevation of the protein concentration. Follow-up brain MRI was remarkable for old infarctions in the bilateral ventromedial pons on T2-weighted imaging (Fig. 1h) and bilateral symmetrical hyperintense lesions at the middle cerebellar peduncles, which suggested Wallerian degeneration secondary to ventromedial pontine infarction (Fig. 1i) (7, 8). No abnormal signals were detected in the medulla oblongata or corticospinal tract. During the course, he did not need gastrostomy or mechanical ventilatory support. Intensive rehabilitation slightly improved his Barthel index score from 0 to 5, and although he was able to pronounce vowels, he could not speak in sentences. The muscle strength in the right upper extremity improved gradually, such that he was able to hold a spoon with a self-help device and managed to bring prepared soft food to his mouth. His left upper extremity and lower extremities remained in contracture, and he was unable to independently maintain a seated posture due to brainstem infarction.

Discussion

We encountered a 46-year-old man with locked-in syndrome due to meningovascular syphilis, which was caused by multiple cerebral infarctions, including brainstem lesions,

Table. Case Reports of Brainstem Infarction Due to Meningovascular Syphilis.

Reference	Age/ sex	Symptoms	HIV status	Lesion on CT/MRI	Vascular findings
17)	35/M	Flaccid right hemiplegia	+	Left basis pontis region of the upper pons	ND
	38/M	Dysarthria, left hemiparesis	+	Right basis pontis	ND
18)	27/M	Intermittent numbness in the left limbs and difficulty in speaking	-	Right anterior tegment of the pons	Marked narrowing and irregularity of the basilar artery
10)	43/M	Left hemiparesis, seizures	+	Right basis pontis	High-grade narrowing of the basilar artery
	48/M	Left hemiparesis	-	Right basis pontis	Severe stenosis of the right supraclinoid internal carotid artery, decreased caliber of the ipsilateral proximal middle cerebral artery, arteritis of the right internal carotid artery, vasculitis involving the distal basilar artery with marked irregularity
19)	33/M	Right-sided weakness, dysarthria, right facial droop	+	Left pons, left thalamus	Occlusion at the distal basilar artery†
20)	43/M	Vertigo, left occipital headache with vomiting	-	Bilateral medial PICA territories, left dorsal medulla	Absence of flow-related signal in a branch of the PICA, normal flow signals in the vertebral arteries
21)	40/M	Left-sided weakness and a soft voice	-	Medial aspect of the right pons	Mid-basilar artery stenosis
22)	54/M	Right hemisensory loss, right inferior visual-field loss	ND	Right crus of the midbrain, left thalamus, left pons (lacunar infarctions)	No evidence of arterial narrowing
11)	35/M	Left hemiparesis, right facial drooping, slurred speech, depress level of consciousness	-	Left cerebellar hemisphere, right side of the pons	Proximal basilar artery occlusion†, ‡
23)	22/M	Left-sided numbness on the face and body	+	Right middle pons	No evidence of arterial narrowing
24)	33/M	Left abducens paresis	-	Posterior aspect of the pons adjacent to the anterior wall of the fourth ventricle	ND
25)	45/M	Occipital headache, right-sided weakness, dysarthria, and right facial droop	-	Left cerebellar hemisphere, both sides of the pons	Absence of flow signal within the distal basilar artery, aneurysm in the middle cerebral artery
26)	50/M	Double vision (Parinaud syndrome)	-	Right paramedian dorsal midbrain	Multiple consecutive stenosis of the right vertebral and basilar artery
27)	51/M	Left-sided weakness	+	Right pons (first episode), right putamen (second episode)	ND
28)	31/M	Bilateral limbs weakness and numbness	-	Bilateral frontal lobe, centrum semiovale, right lateral ventricle, and pons	Stenosis of the bilateral ACA, MCA, right vertebral artery, and basilar artery
Present case	46/M	Quadriplegia and aphonia with eye blinking and eye movements preserved	-	Bilateral ventromedial pontine infarction Left caudate nucleus	Stenosis of the bilateral middle cerebral artery No evidence of basilar arterial narrowing

ND: not described, ACA: anterior cerebral artery, MCA: middle cerebral artery

All reported cases revealed pleocytosis, elevated protein concentration, serological test for syphilis, and venereal disease research laboratory in the CSF.

†Tissue-plasminogen activator administration.

‡Endovascular recanalization.

secondary to symptomatic syphilitic meningitis. The pathology underlying meningovascular neurosyphilis is endarteritis of the medium and large arteries (Heubner's arteritis) and of the small arteries and arterioles (Nissl-Alzheimer's arteritis) (9-11), wherein hyperplasia of the subintimal fibrous tissue results in a narrowed lumen that leads to thrombotic occlusion and ischemic infarction (9). The MCA is the most commonly involved artery, followed by the basilar artery (9). In our patient, the basis pontis infarction that led to incomplete locked-in syndrome was assumed to have been caused by thrombosis of small paramedian branches of the basilar artery due to Nissl-Alzheimer-type syphilitic arteritis. In addition, multiple cerebral infractions, including in the left caudate nucleus, implied CNS vasculitis secondary to

neurosyphilis. Furthermore, an elevated CSF IgG index with OCB pointed toward a specific CNS immune response to the infectious agent (12-15), so the pathogenesis of multiple vascular lesions in this case likely involved both vasculitis and direct invasion by *T. pallidum* (3, 9-11).

A study of imaging findings in neurosyphilis by Brightbill et al. reported that cerebral infarctions were seen in 8 (23%) of 35 patients with neurosyphilis and that brainstem lesions were observed in 2 of 35 cases (10). In contrast, in a report by Peng et al., cerebral infarctions were seen in 6 (43%) of 14 patients, whereas neurosyphilis and brainstem lesions were found in 2 of 14 cases (16). Table shows the details of previous reports of brainstem infarction in meningovascular syphilis (10, 11, 17-28). The patient age ranged from 22 to 54 years old, all were men, and 6 were HIV-positive. Pontine infarctions were seen in 15 (88.2%) of 17 patients, and some reports have also described the presence of multiple cerebral infarctions, as seen in our case. Only a few patients with acute stroke due to meningovascular syphilis who had occlusion of the vertebrobasilar system were treated with intravenous tissue plasminogen activator and subsequently with antibiotics (11, 19).

Locked-in syndrome was first described by Plum and Posner in 1966 (29), and its causes include hemorrhaging, tumor, central pontine myelinolysis, heroin abuse, trauma, pontine abscess, brainstem encephalitis, post-infective polyneuropathy, and air embolism (30). To our knowledge, this is the first report of meningovascular syphilis as the cause of locked-in syndrome. On the ventral side of the pons run the cortical spinal and corticobulbar tracts, a bundle of efferent fibers starting from the motor cortex of the cerebral cortex. The ascending reticular activating system for alert consciousness, named the brain stem reticular formation, has not yet been clarified, but brainstem lesions will not produce coma unless they pass the pontomesencephalic junction in the rostral direction and affect the tegmentum bilaterally (6, 31). Brain stem infarction in the territory of the proximal basilar artery bilaterally shows clinical findings of quadriplegia (bilateral cortical spinal tracts), bifacial paralysis, dysarthria, and tongue and mandibular weakness (bilateral corticobulbar tracts) (31). Locked-in syndrome often leads to severe disability or death (5, 32), and among cases with the former, the prognosis for non-vascular etiologies, such as trauma, central pontine myelinolysis, tumors, and encephalitis, is better than that for vascular etiologies (5). In addition, a young age, absence of hypertension, and vertebrobasilar insufficiency are considered relatively good prognostic factors (30). Despite our patient's young age and lack of severe stenosis of the basilar artery, he experienced severe sequelae, such as becoming bedridden, despite antiplatelet therapy and extended penicillin treatment, probably because the vascular etiology (i.e. infarctions in the bilateral ventromedial pontine, where the corticospinal and corticobulbar fibers run) led to minimal recovery.

Three options should be considered during treatment of cerebral infarction due to meningovascular syphilis: antibiotics against pathogens (duration of penicillin therapy), standard treatment including antiplatelet therapy for cerebral infarction, and immunotherapy for vasculitis. Penicillin administration for 10-14 days is recommended by the practice guidelines for neurosyphilis in the United States (33), the United Kingdom (34), and Europe (35). As a previous case report indicated that a single course of pharmacotherapy for cerebral infarction and of penicillin failed to prevent relapse of syphilitic vasculitis (28, 36), we administered penicillin for 12 weeks until a clear improvement in inflammatory findings and intrathecal antibody production were confirmed. Furthermore, penicillin and antiplatelet treatment alone would be insufficient to improve the prognosis after cerebral infarction. Although we did not add prednisolone to his drug regimen, prednisolone (20-60 mg daily for 3 days) starting the day before anti-treponemal treatment has been recommended in order to prevent syphilitic vasculitis and acute cerebral infarction via the Jarisch-Herxheimer reaction (3, 28). Incidentally, a previous report also suggested the need for a second course of penicillin and corticosteroid therapy in syphilis-associated cerebral vasculitis, as despite corticosteroid and antibiotic treatment for one month, relapse occurred after treatment discontinuation (36). However, it should be noted that there is no evidence at present that prophylactic antiplatelet therapy and corticosteroid therapy for infectious cerebral infarction are effective, especially in syphilitic vasculitis (28, 36, 37). As our patient showed an elevated CSF IgG index with positive OCB, the efficacy of the combination of three approaches for the treatment of cerebral infarction due to meningovascular syphilis should be evaluated.

In summary, we describe a unique cause of bilateral ventromedial pontine infarction, namely meningovascular syphilis, which presented as locked-in syndrome. As brainstem infarction either is fatal or has extremely severe sequelae, meningovascular syphilis should be included in the initial diagnostic workup in young men presenting with uncommon stroke.

The authors state that they have no Conflict of Interest (COI).

References

1. Ghanem KG, Ram S, Rice PA. The modern epidemic of syphilis. New Engl J Med **382**: 845-854, 2020.
2. Ropper AH. Neurosyphilis. New Engl J Med **381**: 1358-63, 2019.
3. Berger JR, Dean D. Neurosyphilis. Handb Clin Neurol **121**: 1461-1472, 2014.
4. Golden MR, Marra CM, Holmes KK. Update on syphilis: resurgence of an old problem. JAMA **290**: 1510-1514, 2003.
5. Patterson JR, Grabois M. Locked-in syndrome: a review of 139 cases. Stroke **17**: 758-764, 1986.
6. Bauer G, Gerstenbrand F, Rumpl E. Varieties of the locked-in syndrome. J Neurol **221**: 77-91, 1979.
7. Uchino A, Sawada A, Takase Y, Kudo S. Symmetrical lesions of the middle cerebellar peduncle: MR imaging and differential diagnosis. Magn Reson Med Sci **3**: 133-140, 2004.
8. Shen Y, Jian W, Li J, Dai T, Bao B, Nie H. Bilateral wallerian de-

generation of the middle cerebellar peduncles secondary to pontine infarction: a case series. J Neurol Sci **388**: 182-185, 2018.
9. Holmes MD, Brant-Zawadzki MM, Simon RP. Clinical features of meningovascular syphilis. Neurology **34**: 553-556, 1984.
10. Brightbill TC, Ihmeidan IH, Post MJ, Berger JR, Katz DA. Neurosyphilis in HIV-positive and HIV-negative patients: neuroimaging findings. AJNR Am J Neuroradiol **16**: 703-711, 1995.
11. Feng W, Caplan M, Matheus MG, Papamitsakis NI. Meningovascular syphilis with fatal vertebrobasilar occlusion. Am J Med Sci **338**: 169-171, 2009.
12. Vartdal F, Vandvik B, Michaelsen TE, Loe K, Norrby E. Neurosyphilis: intrathecal synthesis of oligoclonal antibodies to treponema pallidum. Ann Neurol **11**: 35-40, 1982.
13. Prange HW, Moskophidis M, Schipper HI, Müller F. Relationship between neurological features and intrathecal synthesis of IgG antibodies to Treponema pallidum in untreated and treated human neurosyphilis. J Neurol **230**: 241-252, 1983.
14. Link H, Tibbling G. Principles of albumin and IgG analyses in neurological disorders. III. Evaluation of IgG synthesis within the central nervous system in multiple sclerosis. Scand J Clin Lab Invest **37**: 397-401, 1977.
15. Jones HD, Urquhart N, Mathias RG, Banerjee SN. An evaluation of oligoclonal banding and CSF IgG index in the diagnosis of neurosyphilis. Sex Transm Dis **17**: 75-79, 1990.
16. Peng F, Hu X, Zhong X, et al. CT and MR findings in HIV-negative neurosyphilis. Eur J Radiol **66**: 1-6, 2008.
17. Johns DR, Tierney M, Parker SW. Pure motor hemiplegia due to meningovascular neurosyphilis. Arch Neurol **44**: 1062-1065, 1987.
18. Gállego J, Soriano G, Zubieta JL, Delgado G, Villanueva JA. Magnetic resonance angiography in meningovascular syphilis. Neuroradiology **36**: 208-209, 1994.
19. Han JH, Lee CC, Crupi RS. Meningovascular syphilis and improvement with tissue-plasminogen activator (T-PA). Am J Emerg Med **22**: 426-427, 2004.
20. Umashankar G, Gupta V, Harik SI. Acute bilateral inferior cerebellar infarction in a patient with neurosyphilis. Arch Neurol **61**: 953-956, 2004.
21. Flint AC, Liberato BB, Anziska Y, Schantz-Dunn J, Wright CB. Meningovascular syphilis as a cause of basilar artery stenosis. Neurology **64**: 391-392, 2005.
22. Asdaghi N, Muayqil T, Scozzafava J, Jassal R, Saqqur M, Jeerakathil TJ. The re-emergence in Canada of meningovascular syphilis: 2 patients with headache and stroke. CMAJ **176**: 1699-1700, 2007.
23. Chen MH, Lin CC, Lin TY, Sung YF. Neuro-syphilis, HIV infection and brainstem infarction. QJM **106**: 1039-1040, 2013.
24. Khamaysi Z, Bergman R, Telman G, Goldsher D. Clinical and imaging findings in patients with neurosyphilis: a study of a cohort and review of the literature. Int J Dermatol **53**: 812-819, 2014.
25. Yin-Dan Y, Wen-Ke H, Li-Feng G. Meningovascular syphilis with basilar artery occlusion: case report and literature review. Neurol India **62**: 213-214, 2014.
26. Bäuerle J, Zitzmann A, Egger K, Meckel S, Weiller C, Harloff A. The great imitator - still today! A case of meningovascular syphilis affecting the posterior circulation. J Stroke Cerebrovasc Dis **24**: e1-e3, 2015.
27. Chen TC, Wang JH, Tsai TC. Repeated episodes of acute stroke as manifestation of neurosyphilis in a well-controlled human immunodeficiency virus-infected patient - Successful treatment with ceftriaxone. J Formos Med Assoc **116**: 725-726, 2017.
28. Shi M, Zhou Y, Li Y, et al. Young male with syphilitic cerebral arteritis presents with signs of acute progressive stroke: a case report. Medicine **98**: e18147, 2019.
29. Plum F, Posner JB. The Diagnosis of Stupor and Coma. 1st ed. Davis, Philadelphia, 1966.
30. McCusker EA, Rudick RA, Honch GW, Griggs RC. Recovery from the ‘locked-in’ syndrome. Arch Neurol **39**: 145-147, 1982.
31. Sparaco M, Ciolli L, Zini A. Posterior circulation ischaemic stroke - a review part I: anatomy, aetiology and clinical presentations. Neurol Sci **40**: 1995-2006, 2019.
32. Haig AJ, Katz RT, Sahgal V. Mortality and complications of the locked-in syndrome. Arch Phys Med Rehabil **68**: 24-27, 1987.
33. Workowski KA, Bolan GA. Sexually transmitted diseases treatment guidelines, 2015. MMWR Morb Mortal Wkly Rep **64**: 1-137, 2015.
34. Kingston M, French P, Higgins S, et al.; Members of the Syphilis guidelines revision group 2015. UK national guidelines on the management of syphilis 2015. Int J STD AIDS **27**: 421-446, 2016.
35. Janier M, Unemo M, Dupin N, Tiplica GS, Patel R. 2014 European guideline on the management of syphilis: giving evidence priority. J Eur Acad Dermatol Venereol **30**: e78-e79, 2016.
36. Mageau A, Rodriguez-Régent C, Dion J, et al. Treatment of syphilis-associated cerebral vasculitis: reappearance of an old question. Am J Med **131**: 1516-1519, 2018.
37. Carod Artal FJ. Clinical management of infectious cerebral vasculitides. Expert Rev Neurother **16**: 205-221, 2016.

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