Case Report

Fungal malignant otitis externa caused by *Alternaria chlamydospora*: first case report

George Korres,¹ Joseph Meletiadis,² Alexander Delides,¹ Anastasia Antoniadou,³ George Petrikos,³ Loukia Zerva,² Ioannis Yiotakis,¹ Maria Siopi,² Eleni Kalogeropoulou,² Ilias Georgantis¹ and Thomas P. Nikolopoulos¹

Correspondence

George Korres
gfkorres@gmail.com

¹Otolaryngology Department, Medical School, National and Kapodistrian University of Athens, Greece

²Clinical Microbiology Laboratory, Medical School, National and Kapodistrian University of Athens, Greece

³Department of Internal Medicine, Medical School, National and Kapodistrian University of Athens, Greece

Introduction: Malignant otitis externa (MOE) is a rare clinical entity, usually observed in diabetic or immunosuppressed patients, with serious morbidity due to associated osteomyelitis, cranial nerve palsies and intracranial infections. *Pseudomonas aeruginosa* is the main pathogen in over 95 % of cases; *Aspergillus* species and *Candida albicans* have also been implicated in fungal MOE.

Case presentation: A 79-year-old male with type 2 diabetes with otalgia, otorrhoea and granulation tissue occupying the right external ear canal was diagnosed with MOE. Direct microscopy of a tissue biopsy specimen taken from the granulation tissue revealed septate branching hyphae and chlamydospores. An olivaceous-black colony cultured within 3 days at 30 °C was identified microscopically as *Alternaria* sp. and molecularly as *Alternaria chlamydospora* using the restriction fragment length polymorphism pattern of the internal transcribed spacer (ITS) region on the basis of the 570 bp ITS amplicon, a *Bst*UI largest band of 578 bp and absence of the *Taq*I 114 bp band. The patient was unresponsive to ciprofloxacin, whereas the pain was relieved after 2 weeks of voriconazole treatment followed by surgical debridement.

Conclusion: We present, we believe, the first reported case of MOE for which *Alternaria* sp. seems to be the causative pathogen. A high index of suspicion was needed in order to reach the diagnosis. We recommend taking tissue cultures when a high-risk patient is not responsive to the initial antibiotic treatment, as fungal MOE could be a repercussion of unsuccessfully treated bacterial otitis externa or it could represent a *de novo* presentation of fungal disease.

Keywords: *Alternaria* sp.; diabetes type II; fungal malignant otitis externa; otalgia; otorrhoea; skull base osteomyelitis; voriconazole.

Received 20 November 2014

Accepted 23 January 2015

Introduction

Malignant otitis externa (MOE) is a rare clinical entity; it is a life-threatening extension of external otitis into the mastoid or the skull base. MOE remains a relatively uncommon disease, frequently overlooked by family physicians, that can lead to serious morbidity. It commonly affects immunocompromised patients or patients with diabetes. The infection concerns primarily the external auditory canal and presents as severe otalgia, predominately during the night-time, and purulent otorrhoea; it can rapidly spread via the ear

canal soft tissue to the temporal bone, resulting in osteomyelitis, subsequent cranial nerve palsies and intracranial infection.

The main criteria for MOE are: pain (worsens during the night-time), purulent discharge, swelling of the external ear canal along with the presence of granulation tissue, immunocompromised patient, presence of *Pseudomonas aeruginosa* in swab culture, elevated acute-phase proteins [C-reactive protein (CRP) and ESR] and a positive bone scan with Tc-99m (Cohen & Friedman, 1987).

Aspergillus species and *Candida albicans* have been implicated in fungal MOE. We present a very rare case of fungal

Abbreviations: CRP, C-reactive protein; ITS, internal transcribed spacer; MOE, malignant otitis externa.

malignant otitis caused by *Alternaria* sp., the first we believe, to be reported to date, diagnosis of which was reached with tissue culture.

Case report

A 79-year-old male with type 2 diabetes was referred by his family physician with otalgia (pain in the ear) that was getting worse through the night, and otorrhoea (purulent discharge from the ear), while he remained afebrile.

On otoscopic examination, a large area of granulation tissue, which seemed to originate from the middle ear, was seen to occupy his right external ear canal. The audiogram showed a sensori-neural hearing loss in his better (left) ear (confirming presbycusis) while mixed hearing loss (presbycusis along with conductive hearing loss) was present in his right ear.

His blood tests revealed a white blood cell count of $6.400 \mu\text{l}^{-1}$ (within normal range), while the ESR was elevated to 44 mm. His first swab results indicated *Staphylococcus epidermidis*; however, it was not considered as the main pathogen. A high resolution computerized tomography scan followed by magnetic resonance imaging of the temporal bone showing opacification of the right mastoid cells along with the middle ear, and oedema of the external ear canal skin. A bone scan with technetium (Tc-99m) was scheduled, which revealed osteomyelitis of the temporal bone.

The main criteria for MOE are pain (worsens during the night-time), purulent discharge, swelling of the external ear canal along with the presence of granulation tissue, immunocompromised patient, presence of *P. aeruginosa* in swab culture, elevated acute-phase proteins (CRP and ESR) and a positive bone scan with Tc-99m (Cohen & Friedman, 1987). Therefore, according to these criteria, the diagnosis of MOE was made, even though the presence of *Pseudomonas* was not documented.

The patient started treatment with intravenous ciprofloxacin 600 mg twice daily, and after 2 weeks the ESR had fallen from 44 to 34, while his clinical status improved. He was discharged from our clinic with oral administration of ciprofloxacin 500 mg twice daily and topical use of drops containing ciprofloxacin and cortisone. A month later, he was re-admitted to our clinic complaining that the symptoms had worsened. In his new blood tests, the ESR was elevated to 60 and his otoscopic image findings included absence of any granulation tissue in the external auditory canal and a small perforation in the antero-inferior quadrant of the tympanic membrane, which seemed opacified by tissue within the middle ear. Otomycosis was evident and *C. albicans* was cultured from swabs of the external ear. Furthermore, the granulation tissue was reduced in size at the upper front quadrant of the tympanic membrane, while there was a small perforation of the tympanum at the lower wall of the external ear canal. After careful consideration, a biopsy was taken from the

granulation tissue originating from the middle ear and sent for microbiological examination. Direct fluorescence microscopy of the tissue specimen using 20 % KOH with 0.25 mg ml^{-1} Blankophor P revealed septate branching hyphae and chlamydospores, whereas optical microscopy showed pigmented hyphae (Fig. 1). Tissue specimens were then inoculated in 10 ml liquid Sabouraud dextrose (SAB) medium and SAB agar and incubated for 4 weeks at both 30 and 37 °C. An olivaceous-black colony was cultured within 3 days at 30 °C (Fig. 2). *Alternaria* sp. was identified microscopically by the presence of golden-brown obclavate multicelled swelling irregular conidia and chlamydospores (Fig. 3). The species *Alternaria chlamydospora* was identified molecularly (de Hoog & Horré, 2002) using the restriction fragment polymorphism pattern of the internal transcribed spacer (ITS) region on the basis of the 570 bp ITS amplicon, a *Bst*UI largest band of 578 bp and the absence of the *Taq*I 114 bp band (Fig. 3). *In vitro* antifungal susceptibility testing was performed according to the Clinical and Laboratory Standards Institute protocol M38-A2 (CLSI, 2008) after 48 h of incubation at 30 °C. The MICs of amphotericin B, voriconazole, posaconazole and itraconazole were 1, 0.5, 0.015 and 0.06 mg l^{-1} , respectively, whereas the minimal effective concentrations of caspofungin, micafungin and anidulafungin were 1, ≤ 0.008 and $\leq 0.015 \text{ mg l}^{-1}$, respectively. Based on these facts, the infectious diseases specialist advised treatment with 200 mg intravenous voriconazole twice daily, while ciprofloxacin was discontinued.

Following 2 weeks of treatment, the patient reported that he was free of pain, while his ESR was reduced to 30. It was then decided that surgical debridement would be the next course of action. The patient was taken to the operating room, where atticotomy–mastoidectomy was performed.

Altogether, the patient received intravenous treatment for 1 month (200 mg twice daily) followed by oral treatment

Fig. 1. Direct fluorescence microscopy of tissue specimen treated with 20 % KOH and 0.25 mg ml^{-1} Blankophor P revealed septate branching hyphae and chlamydospores, whereas optical microscopy showed pigmented hyphae. (Magn. $\times 400$.)

Fig. 2. Macroscopic (surface and reverse) and microscopic pictures of *Alternaria* sp. cultured from biopsy specimen.

(200 mg twice daily). After 2 months of treatment with voriconazole, the patient was free of symptoms, while his ESR was 3 (within normal range).

One year later he was free of symptoms and disease.

Discussion

In 1959, Meltzer reported a case of pseudomonal osteomyelitis of the temporal bone. In 1968, Chandler

was the first to define MOE as a distinct clinical disease, while he also discussed the clinical characteristics of malignant external otitis (Chandler, 1968). He observed an aggressive clinical behaviour, along with poor treatment outcome and a high mortality rate; he described this external otitis as malignant.

Over the years that followed, the development of effective antibiotics for treating pseudomonal infections improved the treatment outcomes for patients with MOE. However, during the past decade the number of cases for which treatment has failed has increased. Multidrug-resistant *Pseudomonas* (Loh & Loh., 2013) and the presence of methicillin-resistant *Staphylococcus aureus* (MRSA) (Hobson *et al.*, 2014) in tissue cultures have been identified as possible reasons for the increasing mortality rate.

In the present case, the patient was not responsive to the first line of treatment (ciprofloxacin), and this became a matter of serious concern. According to current clinical practice (Grunstein *et al.*, 2008; Carney, 2008), *Aspergillus* is an aetiological organism of malignant otitis externa, especially when the disease is thought to originate from the middle ear or the mastoid. In cases of *Aspergillus* MOE, the disease is commonly confined to the middle ear rather than the outer ear (Tarazi *et al.*, 2012). *Aspergillus niger*, *Aspergillus flavus* and *Aspergillus fumigatus* are the most common pathogens implicated in fungal MOE (Parize *et al.*, 2009). Therefore, amphotericin B, itraconazole and voriconazole are at present used as the first line of treatment. The use of voriconazole is increasing nowadays in cases of invasive aspergillosis (Walsh *et al.*, 2008); the most commonly reported side-effects are abnormal renal and liver function, electrolyte abnormalities, and visual disturbances (Thompson & Lewis, 2010). Diagnosis of fungal MOE requires formal tissue sampling from the external ear canal or the middle ear in order to identify the causative organism (Tarazi *et al.*, 2012; Halsey *et al.*, 2011; Hamzany *et al.*, 2011; Gordon & Giddings, 1994).

It is not known whether fungal MOE is a repercussion of unsuccessfully treated bacterial otitis externa or if it represents a *de novo* presentation of fungal disease (Walton & Coulson, 2014). There have been 33 previously reported cases of fungal MOE, usually occurring in patients with some form of immunosuppression – typically diabetes, acquired immunodeficiency, or malignancy. A high index of suspicion was needed in order to reach the diagnosis, as, to our knowledge after reviewing the English literature, this is the first reported case of MOE for which *Alternaria* sp. seems to be the causative pathogen. We should therefore recommend taking tissue cultures when a high-risk patient is not responsive to the initial antibiotic treatment.

Acknowledgements

The authors declare no conflict of interest.

Fig. 3. Agarose gel electrophoresis of restriction fragment length polymorphism (RFLP) pattern using the restriction enzymes *TaqI* and *BstUI* to cut the PCR-amplified ITS region. Lanes: 1 and 6, DNA ladder; 2, negative control; 3, RFLP (ITS-*TaqI*); 4, RFLP (ITS-*BstUI*); 5, ITS amplicon.

References

- Carney, A. S. (2008).** Scott-Brown's Otorhinolaryngology, 7th edn, vol. 3, p. 3337. Edited by M. Gleeson Hodder Arnold. CRC press, United States.
- Chandler, J. R. (1968).** Malignant external otitis. *Laryngoscope* **78**, 1257–1294.
- CLSI (2008).** Reference Method for Broth Dilution Antifungal Susceptibility Testing of Filamentous Fungi, Approved standard, 2nd edn, M38-A2, Wayne, PA: Clinical and Laboratory Standards Institute.
- Cohen, D. & Friedman, P. (1987).** The diagnostic criteria of malignant external otitis. *J Laryngol Otol* **101**, 216–221.
- de Hoog, G. S. & Horré, R. (2002).** Molecular taxonomy of the *Alternaria* and *Ulocladium* species from humans and their identification in the routine laboratory. *Mycoses* **45**, 259–276.
- Gordon, G. & Giddings, N. A. (1994).** Invasive otitis externa due to *Aspergillus* species: case report and review. *Clin Infect Dis* **19**, 866–870.
- Grunstein, E., Santos, F. & Selesnick, S. H. (2008).** Current Diagnosis and Treatment, Otolaryngology Head and Neck Surgery, 2nd edn, p. 631. Edited by A. K. Lalwani, FL: Lange. Mc Graw Hill publications, New York, United States.
- Halsey, C., Lumley, H. & Luckit, J. (2011).** Necrotising external otitis caused by *Aspergillus wentii*: a case report. *Mycoses* **54**, e211–e213.
- Hamzany, Y., Soudry, E., Preis, M., Hadar, T., Hilly, O., Bishara, J. & Nageris, B. I. (2011).** Fungal malignant external otitis. *J Infect* **62**, 226–231.
- Hobson, C. E., Moy, J. D., Byers, K. E., Raz, Y., Hirsch, B. E. & McCall, A. A. (2014).** Malignant otitis externa: evolving pathogens and implications for diagnosis and treatment. *Otolaryngol Head Neck Surg* **151**, 112–116.
- Loh, S. & Loh, W. S. (2013).** Malignant otitis externa: an Asian perspective on treatment outcomes and prognostic factors. *Otolaryngol Head Neck Surg* **148**, 991–996, doi:10.1177/0194599813482107.
- Meltzer, P. E. & Kelemen, G. (1959).** Pyocutaneous osteomyelitis of the temporal bone, mandible and zygoma. *Laryngoscope* **169**, 1300–16.
- Parize, P., Chandesris, M. O., Lanternier, F., Poirée, S., Viard, J. P., Bienvenu, B., Mimoun, M., Méchai, F., Mamzer, M. F. & other authors (2009).** Antifungal therapy of *Aspergillus* invasive otitis externa: efficacy of voriconazole and review. *Antimicrob Agents Chemother* **53**, 1048–1053.
- Tarazi, A. E., Al-Tawfiq, J. A. & Abdi, R. F. (2012).** Fungal malignant otitis externa: pitfalls, diagnosis, and treatment. *Otol Neurotol* **33**, 769–773, doi:10.1097/MAO.0b013e3182565b46.
- Thompson, G. R. & Lewis, J. S. (2010).** Pharmacology and clinical use of voriconazole. *Expert Opinion Drug Metab Toxicol* **6**, 83–94.
- Walsh, T. J., Anaissie, E. J., Denning, D. W., Herbrecht, R., Kontoyannis, D. P., Marr, K. A., Morrison, V. A., Segal, B. H., Steinbach, W. J. & other authors (2008).** Treatment of aspergillosis: clinical practice guidelines of the Infectious Diseases Society of America. *Clin Infect Dis* **46**, 327–360.
- Walton, J. & Coulson, C. (2014).** Fungal malignant otitis externa with facial nerve palsy: tissue biopsy AIDS diagnosis. *Case Rep Otolaryngol* **2014**, 192318, doi:10.1155/2014/192318.