An Eight Year Clinico-Microbiological Retrospective Study on Brain Abscesses in India

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ABSTRACT

BACKGROUND: Intracranial abscesses have been a diagnostic and therapeutic challenge since time immemorial for both the microbiologists and the neurosurgeons. There is paucity of detailed studies documenting the infecting organism causing brain abscesses in South India.

AIMS: The study aimed at identifying and assessing the prevalence of aerobic, anaerobic bacteria and fungi associated with brain abscesses at a tertiary care hospital in South India.

METHODS AND MATERIAL: Eight years data was collected from the records of culture reports from 2007 to 2010 and 2013 to 2018. The corresponding clinical case records were retrieved for the assessment of risk factors. Risk factors of brain abscess development were assessed based on clinical cases records.

RESULTS: Data from 140 brain abscess cases obtained over a period of 8 years were analyzed. Out of the 140 samples, 66 (47.14%) were culture positive in which 33 (50%) had single aerobic/facultative anaerobic bacteria, 20 (30.3%) had mixture of more than one aerobic/facultative anaerobic bacteria, 12 (18.18%) had single obligate anaerobic bacteria and 1(1.5%) sample had Mycobacterium tuberculosis isolated. Among the total 92 isolates, Pseudomonas aeruginosa (21/92, 23%) and Staphylococcus aureus (20/92, 22%) predominated. Bacteroides fragilis group was the most common obligate anaerobe isolated. There were no fungal isolates. As there were various isolates (21/92, 23%) and isolated, hence there is heterogeneity of isolates detected Neuroanatomically, parietal lobe (45/140, 32%) was the most common location. Otogenic infection was the major risk factor for parietal and temporal lobe abscesses (P-value <.05).

CONCLUSIONS: It has become essential for the microbiologists to be aware of unusual isolates from brain abscess and its complex nature. Obscurity and difficulty in their microbiological diagnosis calls for more such detailed studies.

KEYWORDS: Brain abscess, clinico-microbiological profile, mycobacterial brain abscess, anaerobic brain abscess

Introduction

Intracranial abscesses are serious life-threatening condition and include brain abscess, subdural empyema, and intracranial epidural abscess.1 Brain abscess is an intraparenchymal collection of pus. It can occur due to spread of infection from a contiguous focus, hematogenous seeding from a distant source, as a sequelae of head trauma or neurosurgical procedure.2 In 25% of cases, an etiology cannot be identified.2 The incidence of brain abscesses is approximately 8% in developing countries and 1% to 2% in western countries.3 Although several aerobic bacteria such as microaerophilic cocci, gram-negative bacilli and gram-positive bacilli, Streptococcus species, S aureus, and H influenzae were isolated in several instances, the recovery of mainly anaerobic bacteria from the brain abscesses suggests the chronic nature of their infection.2 Brain abscesses due to rare organisms have gained importance in the recent years. There is paucity of detailed studies documenting the infecting organism in brain abscesses in South India and rather new and opportunistic pathogens, are emerging.1 In this study we will be testing the hypothesis whether rare organisms which were once thought to be only part of normal flora like the obligate anaerobes, significantly cause brain abscess.

Material and Methods

A hospital-based retrospective study was conducted at a tertiary care hospital, which is a 1000 bedded hospital (Central government autonomous institute) in South India. The hospital mainly receives patients from Pondicherry, where it is situated and from other 2 neighboring states (Tamil Nadu and Andhra Pradesh). As it is a Central Institute, it also receives patients from North India also. The study commenced after obtaining approval from the institute ethics committee (JIPMER), IEC—NO. RC14568. But we asked for waiver of consent, as it was a retrospective record based study.

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We analyzed 140 brain abscess materials (purulent aspirates and/or tissue/CSF) that were received for culture between 2007 and 2010 and 2013 to 2018 (2011 and 2012 data was not available). Culture reports of all the samples received from cases of brain abscesses were retrieved from the microbiology records maintained as a database in the institute. The demographic and clinical information of the patients were retrieved from the medical records section. Relevant data was collected which included the age and sex of the patients, intracranial location of the abscesses, the probable primary source of the infectious agents leading to the formation of the abscesses and the possible risk factors. Significant association of risk factors was determined by calculating the P value. P value less than .05 was taken as significant. The study was approved by the JIPMER Ethics committee (Institutional Ethics committee).

Specimen processing

Direct microscopy was performed by Gram stain to look for the presence of bacteria and inflammatory cells and potassium hydroxide mounts for the presence of fungal elements in all the specimens. Ziehl Neelsen’s (ZN) stain for acid fast bacilli (AFB) was performed only in cases clinically suspected of Mycobacterial infection. Aerobic and anaerobic bacterial culture and fungal culture was performed for all the samples. Aerobic cultures were performed on 5% sheep blood agar and MacConkey agar and was incubated at 37°C for 48 hours, before being reported as sterile. Anaerobic cultures were performed on 5% sheep blood agar, neomycin blood agar, and phenylethyl alcohol (PEA) blood agar and incubated using Himedia GasPak and Anoxomat automated systems. Robertson cooked meat broth was used as an enriching media and was incubated up to 7 days following which subculture is done on the above mentioned solid medias. Fungal culture was performed on Sabouraud’s Dextrose Agar in 2 sets and incubated at 25°C and 37°C each for a maximum period of 4 weeks before declaring as sterile. All the positive cultures were further identified using either conventional biochemical tests or automated system like Phoenix BD. Antimicrobial susceptibility was performed by Kirby-Bauer disk diffusion method according to CLSI guidelines.

Results

A total of 140 brain abscess cases were analyzed during a span of 8 years. The age of the patients ranged from 1 month to 80 years, with males being affected the most (100/140, 71%). Age distribution of the cases has been depicted in Table 1. Majority of the patients belonged to the age group >40 years (38%). Most of the patients presented with the symptoms suggestive of elevated intracranial pressure such as headache (123/140, 88%) and vomiting (93/140, 66%). Other manifestations included fever, convulsions, neck stiffness, hemiparesis etc. The symptom triad of headache, fever, and vomiting was present in 82 (58.5%) patients. A 6 month old child with congenital heart disease (tetralogy of fallot) presented with the features of hydrocephalus. All the cases except one had a single abscess and cerebrum was the most common location (118/140, 84%). Neuroanatomically, parietal lobe (45/140, 32%) was the most common location. Multiple abscess was observed in a single case involving frontal, temporal, and thalamic regions. Distribution of cases based on their clinical manifestations and location of the abscess has been depicted in Table 1. Majority of the brain abscesses were nontraumatic (137/140, 98%), 3 (2%) cases were posttraumatic (following penetrating head injury). Most common predisposing condition associated with the occurrence of brain abscess was otogenic infections (75/140, 53.6%) which included chronic suppurative otitis media, acute suppurative otitis media, mastoiditis, and sinusitis. All 30 temporal brain abscesses and all 45 parietal brain abscesses were associated with otogenic infection. This association was found to be statistically significant with a P value < .05. No identifiable cause was determined in 23.6% of cases. The distribution of cases according to the probable predisposing factors are represented in Table 2.
Table 2. Distribution of predisposing factors among the cases.

| PREDISPOSING FACTORS       | NUMBER OF CASES (N = 140) | PERCENTAGE (%) |
|----------------------------|--------------------------|----------------|
| Otopgenic infections       | 75                       | 53.57          |
| Preterm birth              | 7                        | 5              |
| Diabetes mellitus          | 7                        | 5              |
| CHD (Congenital heart disease) | 6                     | 4.28           |
| Organ transplant           | 5                        | 3.57           |
| Chemotherapy               | 4                        | 2.85           |
| Trauma                     | 3                        | 2.145          |
| Cryptogenic                | 33                       | 23.57          |

**Microbiological spectrum**

Majority of the samples received were pus (71.4%) followed by tissue bit (22.9%) and CSF (5.7%). Out of the total 140 samples, inflammatory cells and bacteria were observed by direct microscopy in 48 (34%) samples, all were negative for fungal elements by direct potassium hydroxide mount. Sixty-six samples (47.14%) were culture positive, out of which 46/66 (70%) were monomicrobial and 20/66 (30%) were polymicrobial. Majority of the samples yielded aerobes or facultative anaerobes (54/66, 81.8%) among which one of the sample had Mycobacterium tuberculosis. Anaerobic bacteria were isolated as a single pathogen from 12/66 (18.2%) of the samples. Polymicrobial infections constituted of only aerobes/facultative anaerobes. None of the samples yielded fungus on culture. All the culture positive samples yielded a total of 92 isolates. Gram negatives were more compared to gram positive organisms. Among the 92 isolates, *P. aeruginosa* (21/92, 23%) and *Staphylococcus aureus* (20/92, 22%) were the predominant ones. Other organisms, like *Burkholderia pseudomallei* (single isolate) and *Streptococcus oralis* (single isolate, secondary to a monkey bite) were also obtained. Most common anaerobic bacteria isolated were of Bacteroides fragilis group. None of the samples yielded fungus on culture. All the culture positive samples yielded a total of 92 isolates. Gram negatives were more compared to gram positive organisms. Among the 92 isolates, *P. aeruginosa* (21/92, 23%) and *Staphylococcus aureus* (20/92, 22%) were the predominant ones. Other organisms, like *Burkholderia pseudomallei* (single isolate) and *Streptococcus oralis* (single isolate, secondary to a monkey bite) were also obtained. Most common anaerobic bacteria isolated were of Bacteroides fragilis group. None of the samples yielded fungus on culture. The detailed spectrum of organisms isolated is represented in table 3 and Table 4. Table 3 represents the more detailed distribution of the isolates with regards to the cases and whether they were single or mixed.

**Management**

Abscess drainage was performed in 30% (n = 42) of cases to limit the abscess spread. The use of antibiotics was mentioned in 63.5% (n = 89) of the case reports. The bacterial brain abscesses were treated with antibiotics like Penicillins, Cephalosporins and Carbapenems. For anaerobic coverage Metronidazole was added to the regimen. Antibiotic treatments were administered for up to 4 weeks. A variety of antibiotics were prescribed to treat the patients with combination therapies prescribed in 79.7% (n = 66), and a single antibiotic treatment was prescribed in 20.3% (n = 23) of the cases. In most of the combination therapies most commonly prescribed antibiotic regimens were a combination of ceftriaxone, imipenem and metronidazole (23%; n = 20), or the latter 2 combined with vancomycin (23%; n = 20). Moreover, the prescribed antibiotic regimen was changed during the course of infection for 32.9% (n = 33) of the patients; the antibiotic treatment prescribed upon hospital admission was most frequently changed to include metronidazole (30.8%; n = 28), ceftriaxone (19.2%; n = 17), or meropenem (15.4%; n = 14). During the entire course of treatment, the most commonly prescribed antibiotic was metronidazole (64.6%; n = 58) followed by: ceftriaxone (39.2%; n = 35), meropenem (13.9%; n = 13), vancomycin (12.8%; n = 12), piperacillin (10.2%; n = 9), clindamycin (8.7%; n = 10), and ampicillin (2.7%; n = 3). Nevertheless, the excessive use of antibiotics is alarming and favors the emergence of drug resistance especially with vancomycin and carbapenems (meropenem).

Tubercular brain abscess was treated with antitubercular drugs where the ATT (Anti-Tubercular Treatment) regimen was followed. All the 140 cases recovered after treatment. There were no deaths.

**Discussion and Conclusion**

Brain abscesses increase mortality rates and prolong hospitalization, that pose diagnostic and therapeutic challenge to both neurologists as well as microbiologists. Most of the time the clinical presentation of patients with brain abscess are atypical, only a few presents with the classical triad of headache, fever, and focal neurological deficits. Many a times even radiological features could be misinterpreted as brain tumors. Most of the time, toxic and systemic features are absent owing to the neuroanatomy and blood brain barrier which prevents the spill of the organisms in to the CSF or blood, which adds to the confusion. Microbiological methods for the identification of the pathogen are being evolved over the time. Yield of pathogens from culture has been variable, negative cultures have been reported to range from 9% to 63% in various studies.

The current study with 140 cases in 6 years span exemplifies that brain abscesses still continues to pose a significant problem with an average of 23 cases per year. This is a little higher compared to the average number of cases per year observed in recent Indian studies ranging from 9% to 15%. In an earlier study, the investigators analyzed 715 cases of brain abscess over a span of 7 years with an average of 102 cases per year, documenting a high incidence of brain abscess. The current study revealed that brain abscess can occur at any age ranging from few months old to elderly population (Figure 1). Even though occurrence of brain abscesses in infants is very rare, we came across 11 cases in this age group. First and second decade of age are known to be more commonly affected. But in our study the most affected age group was 40 years and above (38%).
followed by 2 to 20 years (29%) age group. Male preponderance was observed in our study, irrespective of the age group, which is concordant with most of the studies. In this study, cerebrum was the most common location of the abscesses out of which majority were in the parietal lobe (Figure 2). Location of the abscess was mostly affected by the predisposing factors present in the patients. All the cases with a parietal or temporal lobe abscesses were associated with otogenic infections. But unlike many other studies where otogenic infections leads to temporal lobe abscess, in our study majority were In the parietal lobe followed by temporal lobe. Frontal lobe abscess was most commonly seen in patients with congenital heart disease. Cerebellar abscess was present in 16% of the cases, which is concordant with the general trend.

Table 3. Organisms isolated from the positive cultures.

| NATURE OF ISOLATE | ORGANISMS ISOLATED | NO. OF CASES (OUT OF 66) |
|-------------------|---------------------|--------------------------|
| Aerobic Single species | *Staphylococcus aureus* (MSSA) | 11 (out of 16) |
|                    | *Staphylococcus aureus* (MRSA) | 3 (out of 4) |
|                    | *Coagulase negative staphylococcus* | 3 |
|                    | *Streptococcus species* | 5 |
|                    | *Beta hemolytic streptococci group F* | 3 |
|                    | *Streptococcus oralis* | 1 |
|                    | *Pseudomonas aeruginosa* | 4 (out of 21) |
|                    | *Klebsiella pneumoniae* | 2 (out of 5) |
|                    | *Burkholderia pseudomallei* | 1 |
| Total              |                     | 33 |
| Aerobic mixed species | *Pseudomonas aeruginosa + Enterobacter species* | 5 |
|                    | *Klebsiella pneumoniae + P. aeruginosa* | 4 |
|                    | *Pseudomonas aeruginosa + MRSA + Klebsiella pneumoniae* | 3 |
|                    | *Pseudomonas aeruginosa + Escherichia coli + Providencia species.* | 2 |
|                    | *Proteus mirabilis + Enterococcus faecalis* | 2 |
|                    | *Enterococcus faecalis + Escherichia coli + Pseudomonas aeruginosa* | 2 |
|                    | *Proteus mirabilis + Escherichia coli* | 1 |
|                    | *Pseudomonas aeruginosa + MSSA* | 1 |
| Total              |                     | 20 |
| Obligate anaerobes | *Bacteroides fragilis group* | 8 |
|                    | *Peptostreptococcus species* | 3 |
|                    | *Clostridium bifermentans* | 1 |
| Total              |                     | 12 |
| Mycobacterium      | *Mycobacterium tuberculosis* | 1 |

Abbreviations: MRSA, Methicillin resistant *Staphylococcus aureus*; MSSA, Methicillin sensitive *Staphylococcus aureus*.

They are usually predisposed by middle ear infections, where as in our study majority did not have any identifiable risk factor. Multiple brain abscesses are usually encountered in metastatic abscesses following spread from a primary foci of infection as well as in immunosuppressed individuals. Surprisingly, there was only one patient with multiple abscesses and the predisposing factor present was congenital heart disease.

Most common clinical presentations were fever, headache, and vomiting, symptoms which are consistent with that observed in the literature. Most common risk factor associated with brain abscess were otogenic infections(53.7%) which is similar to many other earlier studies. Next common predisposing factors observed were preterm birth and diabetes mellitus. Congenital heart disease is well known risk factor for
developing brain abscess, especially tetralogy of fallot. People with congenital heart disease are found to be having 10 times higher risk of developing brain abscess. The current study had only 6 patients (4.3%) with congenital heart disease. Organ transplantation and chemotherapy were the other notable risk factors observed. These are commonly associated with fungal etiology than bacterial causes, unlike in our study, where all were bacterial agents. Similarly very few patients had trauma or previous neurosurgical procedures as a predisposing factor, contrary to other studies. Hematogenous spread from a primary foci of infection, most commonly pulmonary infections and endocarditis are well known predisposing factors for the development of brain abscess. Surprisingly, none of the patients in our study had such known foci of infections. But around one-fourth of the cases did not have any notable predisposing factors and majority of these were located in the temporoparietal region. Such cases known as cryptogenic brain abscesses are not uncommon and have an incidence of 15% to 22% in several studies comparable to that of ours.

Direct microscopy plays an important role in Microbiology, as it is the preliminary clue which can be given to the clinician regarding the presence of an infection or the etiology. But in our study, the yield of organisms on direct microscopy was only 48/140 (34%) of the total samples and all these samples were culture positives also. Among the total culture positive samples 18/66 (27.3%) did not show any organism under direct microscopy. A South Indian study has yielded positive direct microscopy findings in 92.7% of cases of brain abscesses, which is very high compared to ours. Even though sensitivity of direct microscopy depends on the load of the organisms present in the sample, better methods are required in this field of technique.

Yield of culture was only 47% in the present study. Most of the studies attribute, prior administration of antibiotics as the major cause of high number of sterile cultures despite all the evidences of brain abscess. Another possibility could be infections due to non cultivable or difficult to cultivate bacteria. Less commonly parasites like Entamoeba histolytica, free living ameba etc. can also cause brain abscess which were not looked into. All the culture positives were bacterial agents, even though we have looked for fungal agents as well. Majority were monomicrobial similar to most of the other studies. However, isolation of multiple pathogens from abscess materials is not uncommon. The polymicrobial nature of brain abscess ranges from 4% to 23%. Facultative anaerobes were predominant agents followed by obligate aerobes, then by obligate anaerobes. Gram negatives are predominant compared to gram positives, contrary to other studies. Pseudomonas aeruginosa (21/92, 23%) and Staphylococcus aureus

Table 4. Spectrum of isolates.

| ORGANISM                           | NUMBER (%) | N = 92 |
|------------------------------------|------------|--------|
| **Aerobic/Facultative anaerobic bacteria** |            |        |
| Pseudomonas aeruginosa            | 21 (23)    |        |
| Staphylococcus aureus             | 20 (21.74) |        |
| Streptococcus species             | 9 (9.78)   |        |
| Escherichia coli                  | 7 (8)      |        |
| Klebsiella pneumoniae             | 5 (5.4)    |        |
| Enterobacter species              | 4 (4.3)    |        |
| Coagulase negative staphylococcus | 3 (3.3)    |        |
| Enterococcus faecalis             | 3 (3.3)    |        |
| Proteus mirabilis                 | 3 (3.3)    |        |
| Providencia species               | 3 (3.3)    |        |
| Burkholderia pseudomallei         | 1 (1.1)    |        |
| Mycobacterium tuberculosis        | 1 (1.1)    |        |
| **Anaerobic bacteria**            |            |        |
| Bacteroides fragilis group        | 8 (9)      |        |
| Peptostreptococcus species        | 3 (3.3)    |        |
| Clostridium bifermentans          | 1 (1.1)    |        |
(20/92, 22%) were the most common isolates in our study. All of them were isolated from patients with otogenic infections (CSOM/ASOM/sinusitis). Streptococcus species were the most common organisms encountered in many of the other studies,1,3,6,8,10 where as in ours study it is the third common cause. Rare organisms like Burkholderia pseudomallei was isolated from a 63 years old man who had met with a road traffic accident and had a penetrating head injury. Melioidosis presenting as brain abscess is rare, with less than 30 cases reported in the last 50 years.13 Streptococcus oralis was isolated from a 2 month old infant, who had acquired infection following a monkey bite over the head. It was a single abscess but multi-loculated and was located in the frontal lobe. An earlier study also documented Streptococcus oralis brain abscess in a 12-year-old girl who was a known case of congenital heart disease.14

Tuberculous brain abscess is an unusual and rare manifestation of tuberculosis of central nervous system, and considered to be a result of altered host response.15 But there are studies where tuberculosis brain abscess predominated compared to pyogenic abscess.1 In this study we have isolated Mycobacterium tuberculosis from only a single case, where the patient was a chronic alcoholic with uncontrolled Type 2 diabetes mellitus. Commonly Mycobacterium tuberculosis gains access to brain through hematogenous route from a distant primary site of infection.16 Tuberculous brain abscess is an unusual and rare manifestation of tuberculosis of central nervous system, and considered to be a result of altered host response.15 But there are studies where tuberculosis brain abscess predominated compared to pyogenic abscess.1 In this study we have isolated Mycobacterium tuberculosis from only a single case, where the patient was a chronic alcoholic with uncontrolled Type 2 diabetes mellitus. Commonly Mycobacterium tuberculosis gains access to brain through hematogenous route from a distant primary site of infection which was not present in this case.1 But, immunocompromised individuals as well as HIV patients has shown higher incidence of TB brain abscess even without a primary focus of infection.15

Obligate anaerobes are well known agents causing brain abscess. There are studies which has shown obligate anaerobes outnumbering aerobes and facultative anaerobes.6 In this study, Bacteroides fragilis group (8/12) were the most common obligate anaerobes isolated followed by Peptostreptococcus. These 2 organisms were commonly isolated from many of the earlier studies as agents of anaerobic brain abscess.1,7

Fungal brain abscesses are rare, but extremely fatal especially in immunocompromised patients. A good clinical suspicion as well as a meticulous diagnostic approach is required for reducing the morbidity and mortality.16 Agents reported in earlier studies include Aspergillus species,1 Zygomycetes,5,17 Cladophialaphora bantiana,18 Candida species,19,20 Scedosporium apiospermum21 etc. But in this study, none of the samples grew fungus in culture.

The current study highlights that, brain abscess is a significant central nervous system infection in developing countries. The microbiological diagnosis of the brain abscesses has become challenging because of the emergence of a variety of new etiological agents like anaerobes which were once a upon a time thought to be a part of normal flora. Role of direct microscopy and culture for the identification of causative agents cannot be underestimated, but the high rate of culture negative cases implies the need for advanced and more sensitive diagnostic methods. The organisms causing brain abscess are difficult to isolate by conventional techniques and hence, there is a need to implement PCR based molecular typing and NGS sequencing for the identification and molecular charac-
terization of the brain abscess associated pathogens.

Author Contributions
Conception of idea: MC, BK. Study design: MC, BK. Drafting: MC, SS, GMS. Drafting of the manuscript: KR.

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