Effect of dexamethasone on experimental brain abscess

KURT A. SCHROEDER, M.D., PAUL E. MCKEEVER, M.D., DENNIS R. SCHABERG, M.D., and JULIAN T. HOFF, M.D.

Sections of Neurosurgery, Neuropathology, and Infectious Diseases, University of Michigan, Ann Arbor, Michigan

Dexamethasone has been used to manage brain edema in patients with intracranial abscess. However, its administration has often been delayed or avoided for fear of adverse effects upon normal host responses to infection. An experimental model of brain abscess in the rat was developed to determine if dexamethasone produced adverse effects on immune competence and collagen deposition in the region of the abscess. Sprague-Dawley rats were inoculated with Staphylococcus aureus and treated intraperitoneally each day with either dexamethasone (0.25 mg/kg) or saline solution. Surviving animals were sacrificed at 4, 8, 12, or 18 days after treatment. The brains were examined grossly for abscess formation and microscopically for intensity of the inflammatory response, abscess diameter, and wall thickness.

There were no differences in mortality rates, abscess production rates, or abscess diameters when groups were compared. The intensity of inflammatory response was similar in both groups. In the group sacrificed 8 days after inoculation, a delay in collagen deposition was apparent, manifested as a thinner abscess wall in the experimental group (mean: 17.8 μ in dexamethasone-treated animals and 85 μ in saline-treated control animals: p = 1.0041). At 12 and 18 days after inoculation, there was no difference in abscess wall thickness between the control and experimental groups. Therapeutic doses of dexamethasone had little effect on mortality rates, incidence of abscess production, or intensity of inflammatory response in the experimental animals. Thus, dexamethasone did cause a delay in collagen deposition in the walls of experimental brain abscesses, but wall thickness 18 days after inoculation was not affected.

Key Words: dexamethasone • brain abscess • Staphylococcus aureus • rat

The introduction of computerized tomography (CT) has modified the management of patients with brain abscess. The CT findings now influence preoperative and postoperative management and frequently affect the timing of operation. Aggressive medical management has supplemented surgical intervention with increasing frequency. Effective management of the patient with brain abscess requires appropriate antibiotic selection to eradicate infection and may require the supplemental use of an agent to control cerebral edema, which is associated with the infection. Dexamethasone is the agent most frequently chosen for the management of edema associated with abscess. However, its use is often delayed or avoided for fear of potential adverse effects on antibiotic penetration, immunocompetence, and abscess encapsulation through collagen deposition.

While experimental evidence has tended to support the detrimental effects of dexamethasone in high doses, dexamethasone has been administered in the management of brain abscess in clinical series with positive results. The current study was undertaken to evaluate the effects of dexamethasone on collagen deposition and inflammatory response in an experimental model of brain abscess in rats. An antibiotic-free design was chosen to remove the variable of antibiotic penetration.

Materials and Methods

Abscess Production

A total of 170 Sprague-Dawley rats, each weighing 250 to 400 gm, were anesthetized with intraperitoneal pentobarbital (40 to 50 mg/kg). Following a modification of the model described by Winn, et al., animal heads were shaved and placed in a stereotaxic frame. The scalp was prepared using povidone paint, and a 1-cm paramedian incision was made over the right calvaria. A 2-mm burr hole was placed 4 mm to the right of the midline just posterior to the coronal suture. A No. 27 needle affixed to a 5-μl syringe was maneuvered with the aid of a micromanipulator. The needle was introduced 3 mm below the dura mater over a
Dexamethasone in experimental brain abscess

period of 10 minutes. An inoculation of 1 μl of Staphylococcus aureus bacterium was made over 30 minutes using a second micromanipulator to depress the plunger of the 5-μl syringe in a controlled fashion. Fifteen minutes elapsed after the termination of injection and the needle was withdrawn over an additional 15 minutes. Wounds were irrigated with povidone and saline solution and then closed.

Inoculum Preparation

Staphylococcus aureus (ATCC 25923) was inoculated into 10 ml of brain/heart infusion medium and incubated at 37°C for 16 hours. The cultures were centrifuged at 10,000 rpm in an SS34 rotor for 10 minutes and the supernatant was discarded. The pellet was resuspended in 10 ml of 0.85% sodium chloride in water and was again centrifuged for 10 minutes at 10,000 rpm. The supernatant was again discarded and the pellet resuspended in 1 ml of the same salt solution. Final concentrations of viable organisms were routinely assayed by serial dilution with plating on solid media and ranged from 2.4 to 5 × 10⁷ colony-forming units (CFU)/μl.

Experimental Groups

The 170 rats were divided into two large groups. Eighty-five animals were selected randomly as controls and received daily injections of intraperitoneal saline solution. The remaining 85 animals were treated with daily intraperitoneal injections of dexamethasone (0.25 mg/kg) begun the day after inoculation. Each of the two large groups was further subdivided into four subgroups: three groups of 17 animals each to be sacrificed at 4, 8, and 12 days, and one group of 34 animals to be sacrificed at 18 days. The animals were killed by an overdose of pentobarbital.

Mortality and Abscess Rates

Mortality and abscess production rates were evaluated. Early mortality was defined as death within the first 48 hours after inoculation and late mortality as death thereafter and up to the day of sacrifice. Animals surviving to term were sacrificed on their appropriate days and the brain was carefully removed. Brains were initially sectioned in a coronal plane through the inoculation needle track. If no abscess was found at the inoculation site, sequential sections were made at 1.5-mm intervals to assure the absence of an eccentric abscess. Brains without macroscopic abscess were excluded from study if no microscopic abscess was found. Incidence of abscess production (survivors with abscess/total survivors) within each group was calculated.

Pathology of Abscesses

Those brains identified as having an abscess were examined histopathologically. Brains were fixed in 10% formalin for at least 3 weeks before paraffin embedding. Sections of each abscess were stained with hematoxylin and eosin and by Wilder's silver impregnation stain for reticulin. All microscopic studies were performed by one examiner who knew the maturity of the abscess but not the therapy rendered.

Hematoxylin and eosin-stained sections were evaluated for the intensity of the inflammatory response. In five regions of interest the intensity of both polymorphonuclear (PMN) leukocytes and mononuclear (MN) inflammatory cells was assigned a numerical score from 0 (absent) to 4 (maximal). The necrotic center of the abscess, the 500-μ area of brain adjacent to the abscess wall, and the perivascular space of vessels near the abscess were studied. Those sections passing through the ventricle were analyzed for evidence of ventriculitis and inflammation of the choroid plexus, whereas the meningeal perimeter was examined in all sections. Numerical scores were tabulated and a mean intensity of inflammation for both PMN and MN infiltrates was determined within each region of interest in a subgroup of animals.

Microscopic examination of sections stained for reticulin was used to determine abscess diameter and wall thickness. One visually "best" diameter was recorded. Four measurements of wall thickness were made and a mean value was derived. Within an animal subgroup, means and standard deviations were calculated for both wall thickness and abscess diameter.

Statistical Analysis of Data

Data on early and late mortality and incidence of abscess production were analyzed using Fisher's exact test. Two-sample t-tests were used to statistically analyze the abscess diameter data. Data concerning wall thickness were first transformed using a square-root function, then analyzed using two-sample t-tests.

Results

Mortality and Abscess Rates

Early and late death of animals after inoculation is shown in Table 1. Mortality was separated into two groups to reflect potentially different etiologies. Early death was attributable to meningitis, ventriculitis, overwhelming sepsis, or anesthetic complications. Late death is potentially a result of unrestrained infection or of chronic steroid administration. The saline-treated control group and the dexamethasone-treated experimental group failed to show a significant difference in either early or late mortality. Percent of abscess production is shown in Table 1. No differences were detected between the experimental and control groups.

Inflammatory Response

Table 2 lists the mean scores assigned for intensity of the MN and PMN leukocyte infiltrates in the five regions of interest. The initial intense PMN response seen in the necrotic centers of the abscesses of the animals studied 4 days after treatment was seen to decline in intensity over time as the response came to involve more MN cells. Brain adjacent to the abscess
TABLE 1
Mortality rate and incidence of abscess production

<table>
<thead>
<tr>
<th>Animal Group</th>
<th>No. of Rats</th>
<th>Early Mortality</th>
<th>Late Mortality</th>
<th>Survivors To Term</th>
<th>Survivors with Abscess</th>
</tr>
</thead>
<tbody>
<tr>
<td>4-day saline</td>
<td>17</td>
<td>5</td>
<td>29</td>
<td>0</td>
<td>12</td>
</tr>
<tr>
<td>4-day dexamethasone</td>
<td>17</td>
<td>3</td>
<td>18</td>
<td>0</td>
<td>14</td>
</tr>
<tr>
<td>8-day saline</td>
<td>17</td>
<td>5</td>
<td>29</td>
<td>0</td>
<td>12</td>
</tr>
<tr>
<td>8-day dexamethasone</td>
<td>17</td>
<td>3</td>
<td>18</td>
<td>0</td>
<td>14</td>
</tr>
<tr>
<td>12-day saline</td>
<td>17</td>
<td>1</td>
<td>6</td>
<td>1</td>
<td>15</td>
</tr>
<tr>
<td>12-day dexamethasone</td>
<td>17</td>
<td>2</td>
<td>12</td>
<td>0</td>
<td>15</td>
</tr>
<tr>
<td>18-day saline</td>
<td>34</td>
<td>8</td>
<td>24</td>
<td>2</td>
<td>24</td>
</tr>
<tr>
<td>18-day dexamethasone</td>
<td>34</td>
<td>6</td>
<td>18</td>
<td>2</td>
<td>26</td>
</tr>
<tr>
<td>all saline</td>
<td>85</td>
<td>19</td>
<td>22</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>all dexamethasone</td>
<td>85</td>
<td>14</td>
<td>16</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

TABLE 2
Mean intensity scores of inflammatory response

<table>
<thead>
<tr>
<th>Animal Group</th>
<th>Central Abscess</th>
<th>Exterior to Wall</th>
<th>Perivascular</th>
<th>Ependymal</th>
<th>Meningeal</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>MNL PMNL</td>
<td>MNL PMNL</td>
<td>MNL PMNL</td>
<td>MNL PMNL</td>
<td>MNL PMNL</td>
</tr>
<tr>
<td>4-day saline</td>
<td>0.5 4.0</td>
<td>2.8 0.3</td>
<td>2.2 0.0</td>
<td>1.1 0.3</td>
<td>1.7 0.0</td>
</tr>
<tr>
<td>4-day dexamethasone</td>
<td>0.3 3.7</td>
<td>2.8 0.2</td>
<td>2.3 0.2</td>
<td>1.3 0.4</td>
<td>0.8 0.2</td>
</tr>
<tr>
<td>8-day saline</td>
<td>0.7 3.4</td>
<td>2.4 1.1</td>
<td>2.3 0.5</td>
<td>0.3 0.8</td>
<td>1.2 0.7</td>
</tr>
<tr>
<td>8-day dexamethasone</td>
<td>0.5 3.7</td>
<td>3.0 0.7</td>
<td>1.8 0.4</td>
<td>no section</td>
<td>0.7 0.5</td>
</tr>
<tr>
<td>12-day saline</td>
<td>0.9 3.6</td>
<td>3.0 0.0</td>
<td>2.2 0.0</td>
<td>1.6 0.3</td>
<td>1.7 0.6</td>
</tr>
<tr>
<td>12-day dexamethasone</td>
<td>0.5 3.5</td>
<td>2.7 0.0</td>
<td>2.8 0.0</td>
<td>1.7 0.5</td>
<td>1.0 0.0</td>
</tr>
<tr>
<td>18-day saline</td>
<td>2.5 2.0</td>
<td>2.0 0.0</td>
<td>0.8 0.0</td>
<td>1.5 0.2</td>
<td>0.0 0.0</td>
</tr>
<tr>
<td>18-day dexamethasone</td>
<td>1.6 1.8</td>
<td>2.4 0.2</td>
<td>1.7 0.1</td>
<td>0.3 0.0</td>
<td>1.5 0.0</td>
</tr>
</tbody>
</table>

* MNL = mononuclear leukocytes; PMNL = polymorphonuclear leukocytes. Scale: 0 (absent) to 4 (maximal).
† No section through ventricle.

wall and perivascular spaces of vessels near the abscess showed a mild and persistent MN infiltrate. Ependymal and meningeal surfaces showed a mild MN infiltrate with an even less intense PMN infiltrate. The intensity of inflammatory infiltrate of saline-injected control animals and dexamethasone-treated animals was not significantly different.

Abscess Diameter and Wall Thickness

At 8 days, abscess wall thickness was less in sacrificed animals that were treated with dexamethasone (Table 3). Mean wall thickness in 8-day saline-treated control animals was 85 μ, while dexamethasone-treated animals had a mean wall thickness of 17.8 μ (p = 0.0041) (Fig. 1). By 12 and 18 days this difference in collagen deposition was no longer apparent, indicating a lag in collagen deposition but no decrease in final wall thickness. Abscess diameters were not significantly different.

Discussion

Although successful nonoperative treatment of brain abscess preceded the introduction of CT, widespread use of the imaging capabilities of CT has allowed more frequent nonsurgical management of patients with brain abscess. The CT findings often allow assessment of the abscess' stage and improve the precision of medical management. Newer microbiological techniques have resulted in more complete identification and characterization of the bacterial spectrum responsible for brain abscess, leading to better choices for antibiotic therapy. Newer antibiotics with greater penetration into the central nervous system and newer methods of delivery of antibiotics to achieve higher tissue levels may have also improved outcome in patients with brain abscess.

However, in spite of significant progress in the imaging, microbiology, and antibiotic treatment of brain abscess, much controversy still surrounds the treatment of abscess-associated edema. With the exceptions of ventriculitis and septic thrombophlebitis, it is the effect of the expanding mass of the abscess and surrounding edema that most often represents the threat to the patient's life. Both dexamethasone and osmotic diuretics have been recommended to control abscess-associated edema, but dexamethasone is most frequently used. Experimental evidence documenting edema reduction supports the clinical observations of often dramatic neurological improvement in patients treated with corticosteroids.

Dexamethasone has the potential to produce adverse
Dexamethasone in experimental brain abscess

**FIG. 1.** Left: Photomicrograph of the abscess wall in an 8-day saline-treated control animal. The abscess center is at the left of the illustration with the reticulin and collagen deposition in the center. Reticulin, × 240. Right: Photomicrograph of the abscess wall in an 8-day dexamethasone-treated animal. Region of collagen deposition and reticulin formation is in the center of the field and is contrasted with that shown at left. The abscess center is at the left. Reticulin, × 240.

Effects on antibiotic penetration, immunocompetence, and collagen deposition. Although abscesses may not be cured in spite of high antibiotic levels within the abscess, experimental evidence suggests that certain antibiotics (benzylpenicillin, cefazedon, lymecycline), but not others (gentamicin, metronidazole) show decreased tissue levels in the brain after dexamethasone treatment. It is postulated that these decreased tissue levels are a direct result of decreased permeability at the blood-brain barrier brought about by dexamethasone, which may explain the mechanism of edema modification. Antibiotics that are more freely permeable at the blood-brain barrier and with higher lipid solubilities may not be as affected by dexamethasone.

Evidence from experimental brain abscess research has generally supported the contention that high-dose corticosteroids interfere with the inflammatory response of the host. Adverse effects on PMN, macrophage, and glial elements have been reported. These same studies and others have documented altered collagen deposition which may delay abscess encapsulation. Enzmann, et al., considered that once the capsule was formed there should be little hindrance of further collagen deposition and that inhibition of collagen deposition took place when corticosteroids were administered from the onset of the inoculation of bacteria. However, edema is greatest early in the development of the abscess, and at that time corticosteroids will most likely be necessary to control mass effect of the abscess and its associated edema.

The current study used an antibiotic-free design to remove the potentially confounding variable of antibiotic penetration and allow direct evaluation of dexamethasone on collagen deposition/abscess encapsulation and immunocompetence/inflammatory response. Like Neuwelt, et al., we required more potent in-oculi to achieve high rates of abscess production. This certainly contributed to an increased mortality rate. The direct-injection method of abscess production has the advantage of one-step abscess production without the potential artifacts of the septic embolic model or potential foreign-body reactions associated with the concomitant injection of nutrient medium.

Although differences exist among species regarding corticosteroid immunosuppression sensitivity (man is relatively resistant, rat relatively sensitive), we purposely chose an animal likely to demonstrate a deleterious effect from dexamethasone. The corticosteroid dose chosen was intended to reflect a dose analogous to that frequently employed in man in the clinical management of abscess-associated edema. Treatment was initiated 1 day after inoculation and was continued through to sacrifice to reflect timing when dexamethasone is most likely to be used to combat maximal edema. This is before and during early abscess encapsulation and is a time when dexamethasone is most likely to have its detrimental effects.

**TABLE 3**

<table>
<thead>
<tr>
<th>Animal Group</th>
<th>Mean Abscess Diameter (μ)</th>
<th>Mean Abscess Wall Thickness (μ)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4-day saline</td>
<td>1267 ± 549</td>
<td>none</td>
</tr>
<tr>
<td>4-day dexamethasone</td>
<td>1213 ± 512</td>
<td>none</td>
</tr>
<tr>
<td>8-day saline</td>
<td>1174 ± 600</td>
<td>85 ± 81.6</td>
</tr>
<tr>
<td>8-day dexamethasone</td>
<td>1003 ± 630</td>
<td>17.8 ± 37.6</td>
</tr>
<tr>
<td>12-day saline</td>
<td>1260 ± 595</td>
<td>276 ± 97.8</td>
</tr>
<tr>
<td>12-day dexamethasone</td>
<td>1064 ± 561</td>
<td>231 ± 61.5</td>
</tr>
<tr>
<td>18-day saline</td>
<td>1018 ± 797</td>
<td>285 ± 95.3</td>
</tr>
<tr>
<td>18-day dexamethasone</td>
<td>1094 ± 823</td>
<td>257 ± 116.5</td>
</tr>
</tbody>
</table>

*Data are means ± standard deviations.
pharmacological doses for experimental brain abscess, has little effect on early and late mortality, incidence of abscess production, or intensity of the inflammatory response. Collagen deposition was delayed by dexamethasone in our experiments, but encapsulation proceeds and final abscess wall thickness and abscess size are not affected. Our experiments lend support to the favorable outcomes without adverse effects described in patients with brain abscess treated with corticosteroids.

References
38. Rosenblum ML, Hoff JT, Norman D, et al: Decreased mortality from brain abscesses since advent of comput-
Dexamethasone in experimental brain abscess

treatment of brain abscesses in selected high-risk patients.
40. Rotheram EB Jr, Kessler LA: Use of computerized to-
mography in nonsurgical management of brain abscess.
Arch Neurol 36:25–26, 1979
41. van Alphen HAM, Dreissen JJR: Brain abscess and sub-
dural empyema. Factors influencing mortality and results
of various surgical techniques. J Neurol Neurosurg Psy-
42. Wallenfang T: Das entzündliche Hirnödem und der re-
geonale Hirnstoffwechsel beim experimentellen Hirnab-
43. Wallenfang T, Bohl J, Kretzschmar K: Evolution of brain
abscess in cats: formation of capsule and resolution of
44. Wallenfang T, Bohl J, Schurmann K: Etude expérimen-
tale de l'évolution des abcès du cerveau. (Oedème céré-
bral et corrélation neuropathologique.) Neurochirurgie
26:145–152, 1980

45. Wallenfang T, Reulen HJ, Schindling H: Investigation on
the prognosis of brain abscess, in Wültenweber R, Brock
alus. Advances in Neurosurgery, Vol 4. Berlin: Springer-
Verlag, 1977, pp 296–299
46. Winn HR, Mendes M, Moore P, et al: Production of
experimental brain abscess in the rat. J Neurosurg 51:
685–690, 1979
(Letter)
vascular proliferation on angiographic appearance and
encapsulation of experimental traumatic and metastatic

Manuscript received September 24, 1985.
Accepted in final form August 4, 1986.
Address reprint requests to: Kurt Schroeder, M.D., 5182
East Farness Drive, Tucson, Arizona 85712.