ACCELERATED CLEARANCE OF RADIOACTIVE CHELATE FROM CEREBROSPINAL FLUID IN EXPERIMENTAL MENINGITIS

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Albumin labeled with ^{131}I or ^{99m}Tc has been used extensively for cerebrospinal fluid (CSF) imaging (*1*). Recently, ^{169}Yb chelated with diethylenetriaminepentaacetic acid (^{169}Yb-DTPA), a gamma-emitting radioactive chelate, has been used for cisternography (*2,3*). Although the molecular weight of these two agents are approximately 70,000 and 600, their kinetic behavior in CSF is similar (*4*). Prockop and associates (*5*) have studied the CSF clearance of ^{131}I-albumin, ^{14}C- or ^{3}H-labeled inulin, and 3-0-methyl-glucose and mannitol in dogs under normal conditions and with pneumococcal meningitis. They observed almost identical clearance rates for albumin and inulin in normal dogs, but the clearance of mannitol was slightly faster and that of 3M-glucose more rapid. Clearance of mannitol was accelerated greatly in meningitis compared to other agents which suggested that it was the result of a nonspecific increase in membrane permeability. Our initial studies also demonstrated the similarities of the behavior of chelate, inulin- and albumin-labeled with gamma-emitting radionuclides in normal dogs (*6*).

We postulated that the behavior of ^{169}Yb-DTPA would be similar to ^{14}C-mannitol but that the gamma-emitting radioactive chelate would have the advantage of permitting measurement of CSF clearance using an external scintillation detector. We therefore undertook the present study with ^{169}Yb-DTPA in dogs with experimentally induced aseptic and bacterial meningitis. Our overall goal is to investigate the feasibility of developing a clinically useful diagnostic test for meningitis.

MATERIALS AND METHODS

Studies were carried out in mongrel dogs of body weight of approximately 15 kg. They were divided into three groups (normal dogs and dogs with aseptic and bacterial meningitis), each consisting of five "successful" experimental animals. Unsuccessful studies were those when the dogs did not develop any clinical symptoms of meningitis or died, or when the intracisternal administration of the tracer dose was technically poor.

Aseptic meningitis was produced by injection into the cisterna magna of varying dosage of streptokinase-streptodornase (SK-SD) between 25,000 and 100,000 units in about 2 ml isotonic saline after withdrawal of about the same amount of CSF. This procedure was similar to that of Baltch, et al (*7*). Bacterial meningitis was produced using a pure strain of pneumococci maintained in sterile blood or chocolate agar culture with serial transfers at least every 2 weeks. Prior to each study an aliquot was transferred to trypticase soy broth with defibrinated blood (1:9) and incubated overnight at 37°C. The inoculum regularly contained 10^{7}–10^{8} organisms/ml. Approximately 1 ml was injected intracisternally.

The dogs were kept under close observation after the injection of enzyme or bacteria, and the isotopic experiments were performed after 18–72 hr depending on the clinical evidence of meningitis. The general signs and symptoms observed in the case of aseptic meningitis were lethargy, cervical rigidity, hypersthesia, apprehensiveness, weakness, partial paralysis of hind quarter and limbs, ataxia, muscle twitching, opisthotonos, etc. In case of bacterial meningitis, along with the above signs, hyperthermia, conjunctivitis, otitis media, rhinorrhea, etc. were also observed. Partial paralysis of hind legs was more frequent with aseptic meningitis than pneumococcal meningitis.

Ytterbium-169-DTPA was prepared according to the method described earlier (*2*) or obtained from 3M Company*, and diluted in isotonic saline to

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obtain a concentration of 500 μCi/ml. A 22-gage needle attached to a 3-ml plastic syringe containing 0.5 ml isotonic saline was inserted into the cisterna magna. If about 0.5 ml CSF could be withdrawn, 0.5 ml of ^{169}Yb-DTPA in isotonic saline (approximately 250 μCi) was injected through the same needle and washed twice by withdrawing about 0.3 ml CSF and at the same time assured of the absence of any trace of blood.

A scintillation probe with a 2-in. crystal and flat field collimation was fixed over the head (vertex, around the site of injection) at a height of about 10 cm. The probe was adjusted for maximum observable initial counting rate which was made equivalent to 100 on the strip chart recorder moving at a speed of 0.5 cm/min. The clearance was recorded for 4–6 hr. Care was taken to keep the dog stationary by administering small amounts of pentobarbital occasionally. The values of counting rates were noted for every 15 min from the strip chart and replotted on a semilogarithmic paper. An index of clearance rate was taken as $0.693/t_{1/2}$, where $t_{1/2}$ was equivalent to the time corresponding to the first 50% clearance. A CSF sample was taken from most of the dogs after the clearance study for routine CSF analysis: protein and sugar contents, cell counts, and bacterial culture.

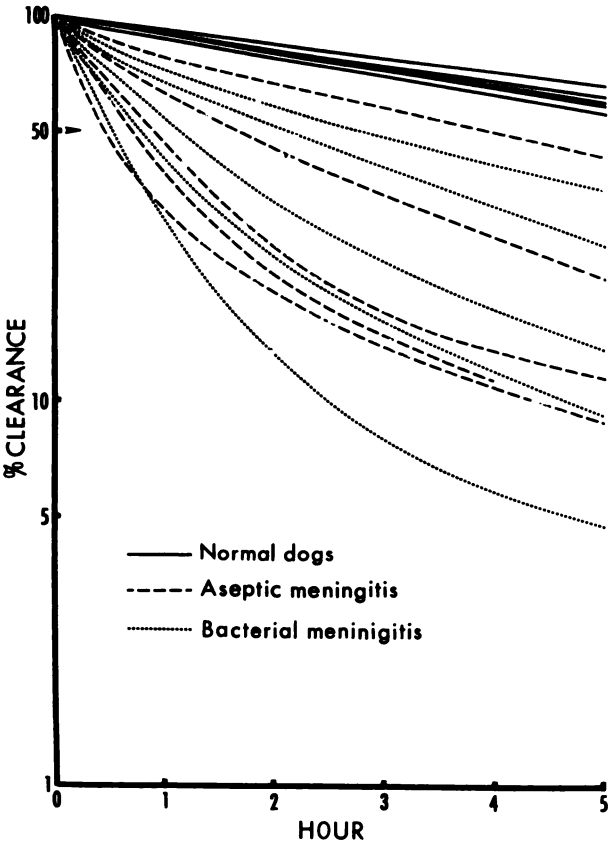

FIG. 1. Accelerated clearance of radioactive chelate from cerebrospinal fluid in experimental meningitis.

RESULTS

The clearance of ^{169}Yb-DTPA from the head of normal dogs after injection into the cisterna magna approximated to a single exponential rate with clearance half-times ranging between 5.6 and 7.8 hr. The calculated clearance rate constants were between 0.15 and 0.21%/min (0.18 $\pm$ 0.02%/min). The clearance pattern in experimental meningitis deviated from a single exponential time function. The 50% clearance times for all these dogs ranged between 27 min and 3 hrs, 50 min, which were considerably less than in normal dogs, indicating increased rates of clearance of the chelate. The indices of clearance rates for aseptic meningitis ranged between 0.3 and 2.6%/min (1.25 $\pm$ 0.89%/min) and those for bacterial meningitis between 0.4 and 2.2%/min (1.13 $\pm$ 0.74%/min). Aseptic meningitis could not be differentiated from bacterial meningitis on the basis of clearance studies. Figure 1 summarizes the clearance of the radiochelate for all the dogs.

Results of analysis of chemical and cellular changes in the CSF did not correlate well with observed clearance rates. These values for normal dogs were 10–18 mg% for protein, 70–76 mg% for glucose, 0–10/cu mm for WBC, and 0–2/cu mm for RBC. In aseptic meningitis CSF protein and WBC counts were significantly increased (20–139 mg% and 35–180/cu mm, respectively) with little variation in glucose content. In bacterial meningitis variable values of protein and glucose contents and WBC counts were found (0–90 mg%, 30–80 mg%, and 0–300/cu mm, respectively). CSF samples with pneumococcal meningitis showed bacterial growth in culture but not those with aseptic meningitis.

DISCUSSION

Considerable work has been carried out on the CSF clearance of radioiodinated albumin and other agents in bacterial meningitis but not in aseptic meningitis. Prockop and associates suggested a nonspecific increase in membrane permeability in pneumococcal meningitis (*5*). They observed a rapid clearance of mannitol, with a relative block in the bulk CSF flow of large molecules in the subarachnoid space and/or arachnoid villi. Rapid clearances of ^{169}Yb-DTPA from the CSF in the present study with experimental meningitis are attributable to increased diffusion of the chelate due to nonspecific increase in membrane permeability. The wide varia-

tion in clearance rates in meningitis was roughly correlated with the degree of meningitis on the basis of clinical observations.

The deviation of clearance pattern from single exponential type in most of the meningitic dogs might be explained on the basis of relatively severe alterations in membrane permeability around the cisterna magna. As the enzyme or the bacteria was introduced into the cisterna magna, it was likely that the changes in the surrounding membrane were more severe than elsewhere. If the permeability of the membrane were altered uniformly, the clearance pattern might be expected to approximate a single exponential type. However, if the permeability of the membrane around the cisterna magna were altered considerably as compared to the rest of the region, the initial rate of clearance would be fast due to passive diffusion from the cisterna magna.

The present technique of in vivo measurement of clearance of a gamma-emitting chelate from the CSF is potentially useful for the clinical evaluation of meningitis. However, intracisternal injection of any agent is not usually favored in patients but the radiochelate might be injected into the lumbar subarachnoid space at the time of lumbar puncture. Ytterbium-169-DTPA has a long physical half-life of 32 days but the biological half-life with respect to CSF is less than 1 day (*4*). However, other short-lived chelates, like ^{111}In-DTPA (*8*) or ^{99m}Tc-DTPA (*9*), could be used for the same purpose.

SUMMARY

Studies were carried out in dogs after 18–72 hr following intracisternal injection of SK-SD or pneumococci to produce aseptic or bacterial meningitis, respectively. Approximately 250 μCi of ^{169}Yb-DTPA was injected intracisternally and its clearance from CSF was measured using a scintillation probe at a distance of about 10 cm. Clearance rates were calculated using 50% clearance time and the values were found to be 0.15–0.21, 0.3–2.6, and 0.4–2.2%/min, respectively, for normal dogs and for aseptic and bacterial meningitis. The results suggested an increased diffusion of the radioactive chelate due to nonspecific increase in membrane permeability. Its potential usefulness in the clinical diagnosis of meningitis is under investigation.

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