INTRATHECAL CHLORAMPHENICOL IN STAPHYLOCOCCAL MENINGITIS RESISTANT TO PENICILLIN AND STREPTOMYCIN

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Chloramphenicol, an antibiotic first isolated from *Streptomyces venezuelae* (Ehrlich *et al.*, 1948), has recently been prepared synthetically (Olive, 1949). It is a crystalline chemical substance with a wide therapeutic range. It is soluble in water to a concentration of 2.5 mg. per ml. at 25° C., and solutions withstand boiling for five hours without measurable loss of potency. The antibiotic shows considerably more activity against Gram-negative organisms than against Gram-positive and acid-fast species. It is seven to thirty times as active as penicillin against tested Gram-negative species, but much less active against *Staphylococcus aureus*. The range of inhibiting concentration for the organism appears to lie between 1 and 16 µg. per ml. The drug may be administered orally or rectally. A review of the literature reveals no record of the use of intrathecal chloramphenicol in the treatment of pyogenic meningitis. We believe this to be the first record of the administration of chloramphenicol by this route.

Case 1

A man aged 44 was operated on for the exploration of a cerebellar tumour on February 14, 1951. A midline cerebellar haemangioblastoma was completely and successfully removed, and routine penicillin was administered in doses of 250,000 units six-hourly. On the third post-operative day the patient complained of increasingly severe occipital headache, worse on the left side. His temperature rose to 103° F. (39.4° C.), and 48 hours later he developed a free discharge of blood-stained pus from the right side of his scalp incision. A direct film stained by Gram's method showed numerous pus cells and large clumps of Gram-positive cocci. Culture of the pus gave a profuse pure growth of coagulase-positive *Staph. aureus*. The antibiotic sensitivity was determined by the disk method. The organism was found to be resistant to penicillin, strepto-

mycin, and sulphadimidine, but sensitive to chloramphenicol, and, when tested on a nutrient agar plate at pH 5.8, highly sensitive to "aureomycin."

Tube titrations with these antibiotics were later performed, and the organism was found to be resistant to 100 units of penicillin per ml. and to 100 µg. of streptomycin per ml. It was sensitive to chloramphenicol in a concentration of 2.5 µg. per ml. and to aureomycin in a concentration of as little as 0.01 µg. per ml. This test was performed in nutrient broth at pH 6.8 and the results were read turbidimetrically (Anderson, 1951). Lumbar punctures performed as a routine on each post-operative day showed a pleocytosis which reached 1,100 per c.mm. on the third day, a figure which is considerably higher than that generally seen. By the eighth day, however, the level had fallen to 80 cells, which was judged to be within the usual limits for a post-operative count. It was therefore considered on clinical evidence that the patient was not suffering from an incipient meningitis.

On the seventh post-operative day, in view of the antibiotic spectrum of the infecting organism, the patient was given aureomycin, 750 mg. six-hourly, by mouth. High cerebrospinal fluid pressures necessitated lumbar punctures twice daily. During the succeeding week the general condition improved and the discharge from the incision became serous and finally ceased on February 28. The cytology of the lumbar cerebrospinal fluid remained within the usual post-operative limits.

On the 16th post-operative day, after some increase in pyrexia, the patient developed a collection of blood-stained fluid deep to the skin flap. This required daily aspiration over the course of four days. Direct films showed masses of Gram-positive cocci, and cultures yielded a pure growth of coagulase-positive *Staph. aureus*.

On the 18th post-operative day the patient presented definite signs of meningitis, and the lumbar cerebrospinal fluid was now turbid, containing 290 leucocytes per c.mm., with 90% polymorphonuclear cells. Direct films of the centrifuged deposit showed several groups of extracellular Gram-positive cocci. Culture yielded a scanty growth of coagulase-positive *Staph. aureus*. This organism proved to have the same antibiotic spectrum as the coccus isolated from the wound infection. Thus the patient had developed a meningeal infection despite the fact that he had been given maximal oral doses of aureomycin for 12 days. Subjectively, he complained of increased headache and drowsiness, and examination revealed some dysarthria in addition to neck rigidity, but no Kernigism. The following day it was decided to give chloramphenicol by mouth in doses of 1 g. six-hourly, increasing in 48 hours to 2 g. six-hourly. On the succeeding three days the leucocyte count remained high in the lumbar fluid, and the infecting organism was still seen in Gram films of the centrifuged deposit. In view of this grave situation it was decided to try to increase the concentration of chloramphenicol in the cerebrospinal fluid by intrathecal administration.

On the 23rd post-operative day 100 µg. of chloramphenicol in 1 ml. of sterile distilled water was cautiously given intrathecally; this was repeated on the following day. At the same time 200 µg. was injected into the collection of cerebrospinal fluid deep to the scalp flap. These injections were well tolerated. The considerable nausea experienced by the patient at this stage was thought to be due to the large dosage of aureomycin. The intrathecal chloramphenicol was increased gradually until a maximum of 750 µg. was reached after 19 days. The cell counts in the lumbar fluid and the chloramphenicol levels during treatment are shown in Fig. 1 and the Table. The patient gradually improved, apart from an increase in cells on the 35th post-operative day which may have been due to an escape of infective material

FIG. 1.—Total cell counts in cerebrospinal fluid of Case 1, showing response to chloramphenicol therapy.

Table Showing Chloramphenicol Levels in Cerebrospinal Fluids Related to Daily Intrathecal Dosage

Case 1			Case 2		
Days Post-operative	Daily Intrathecal Dose in μ g.	Cerebrospinal Fluid Levels in μ g. per ml.	Days Post-operative	Daily Intrathecal Dose in μ g.	Cerebrospinal Fluid Levels in μ g. per ml.
23	100		14	600	
24	100		15	600	
25	400		16	600	40
26	400		17	700	40
27	400	10	18	700	
28	400		19	750	
29	400		20	—	20
30	400	10			
31	400				
32	400				
33	400	20			
34	500	20			
35	500	20			
36	600	30			
37	600				
38	700				
39	700				
40	700				
41	750	40			

from a focus deep to the flap, although at this time there was no collection requiring aspiration.

One positive culture was obtained from the lumbar fluid on the 30th post-operative day, but all subsequent cultures were sterile. Aureomycin was discontinued owing to severe nausea.

Intrathecal chloramphenicol was discontinued on the 41st post-operative day, after 19 days of treatment, but oral therapy was continued until April 6. The cerebrospinal fluid cell counts fell gradually to normal and the patient's general condition was much improved. He was allowed up, and examination six weeks after the cessation of all treatment showed him to be symptom-free.

Methods

Pure crystalline chloramphenicol was used for the preparation of intrathecal solutions. It must be emphasized that only the pure substance should be employed for intrathecal use in order to avoid the introduction of foreign material and also to ensure accuracy in preparing the solutions. The drug was dissolved in distilled water to give a final concentration of 100 μ g. per ml. This was dispensed in ampoules of 1-ml. and 5-ml. amounts. These solutions were subsequently compared with solutions which had not been autoclaved. Each was titrated against the patient's organism, and there was no detectable loss of potency after sterilization in the autoclave. The seven chloramphenicol assays on the cerebrospinal fluid were carried out on specimens taken immediately before the next intrathecal injection. Unit volumes of 1 ml. were employed, making doubling dilutions with nutrient broth. A six-hour broth culture of the staphylococcus isolated from the patient was employed throughout as a test organism, 0.02 ml. being added to each tube from a dropping pipette. Control titrations with known chloramphenicol concentrations were set up in each case.

The first assay was performed when the patient was still receiving aureomycin by mouth, and was therefore complicated by the possible presence of this antibiotic in the cerebrospinal fluid. In view of the relatively high sensitivity of the test organism to aureomycin (0.01 μ g. per ml.), and the instability of that antibiotic to heat, it was felt that a combined assay might be attempted. One sample of fluid was titrated in the normal way, whilst a second sample was boiled and cooled before titration. It was thought that this treatment would destroy any aureomycin present, whilst

leaving the extremely stable chloramphenicol unchanged. In reading the aureomycin titration it was necessary to make the somewhat unwarrantable assumption that no synergistic action existed between the two antibiotics. The approximate levels so obtained were 0.64 μ g. of aureomycin per ml. and 10 μ g. of chloramphenicol per ml. After this assay, aureomycin was discontinued, and subsequent estimations were uncomplicated by the presence of two antibiotics.

Case 2

A man aged 47 was admitted for exploration of an acoustic neuroma which was completely removed by an occipital approach on March 16. He subsequently made satisfactory progress, his total cerebrospinal fluid cell count being consistently below 100 per c.mm.

On the thirteenth post-operative day the space deep to the skin flap was aspirated because loculation of cerebrospinal fluid in that site was suspected. The fluid was clear

FIG. 2.—Total cell counts in cerebrospinal fluid of Case 2 during chloramphenicol therapy.

and contained 59 leucocytes. Overnight the patient complained of increasing headache and nausea, and his temperature rose steeply to 101° F. (38.3° C.). Lumbar puncture now showed a frankly purulent fluid containing 2,200 leucocytes per c.mm. (Fig. 2). The patient had developed a pyogenic meningitis within 12 hours of aspiration. As the nature of the infecting organism was at that time unknown, treatment with penicillin was begun at once, 500,000 units being administered intramuscularly six-hourly, and 40,000 units injected intrathecally. Immediate examination of the fluid showed large numbers of pus cells and several clumps of extracellular Gram-positive cocci. After 18 hours' incubation, cultures on blood agar yielded a moderate growth of coagulase-positive *Staph. aureus*. In order to provide information about the antibiotic sensitivity at the earliest possible moment, a thick lawn plate was prepared with the appropriate disks and examined by reflected light at the end of six hours' incubation. At this time it was possible to report that the organism was resistant to penicillin and streptomycin, but sensitive to chloramphenicol. Further incubation and tube titrations subsequently confirmed these findings.

All penicillin therapy was immediately discontinued, and, in view of the failure to eradicate the organism in the previous patient with oral aureomycin, it was decided to treat the case with large doses of chloramphenicol, both orally and intrathecally. The patient was accordingly given 2 g. six-hourly by mouth and 600 μ g. intrathecally daily, rising to 750 μ g. on the sixth day. Within 24 hours there was a marked symptomatic improvement: headache disappeared, and the patient was no longer nauseated or drowsy. Within 72 hours the cell count in the lumbar fluid had fallen to 75 per c.mm. (Fig. 2). By this time

the patient was symptomless and was, in fact, making persistent demands for permission to get up. Intrathecal chloramphenicol was discontinued after six days, and oral therapy after 12 days. The results of three assays performed on the lumbar fluid are shown in the Table. The patient maintained uninterrupted progress and was allowed to get up.

Discussion

It is difficult to explain why Case 1 developed meningitis while receiving full doses of aureomycin by mouth. The infecting organism was shown to be markedly sensitive, and the isolated assay performed while the patient was still taking the drug suggested an adequate concentration to be present in the cerebrospinal fluid. Two possible explanations present themselves. First, it seems likely that the cerebrospinal fluid was subjected to repeated and heavy contamination from the collection of infected fluid deep to the operation flap. This is supported by the findings of numerous cocci in Gram films prepared from several samples of fluid, and also by the isolated positive culture and cell peaks seen in Fig. 1, all of which suggest repeated contamination. Secondly, aureomycin is reported to reach the cerebrospinal fluid in relatively small concentrations.

Werner *et al.* (1950), investigating five patients receiving aureomycin by mouth, were unable to detect the antibiotic in the cerebrospinal fluid. Even with serum concentrations of 7.6 $\mu\text{g.}$ per ml. Bryer *et al.* (1948), working with dogs, failed to detect any measurable amounts in the spinal fluid after parenteral administration. However, Harned *et al.* (1948) found that intravenous administration in dogs resulted in levels up to 0.8 $\mu\text{g.}$ per ml. Owing to the fact that solutions of aureomycin have a final pH of 4.5 this antibiotic has so far been considered unsuitable for intrathecal use. Spinal fluid concentrations in patients treated with oral chloramphenicol seem to be more encouraging. Ross *et al.* (1949) have shown that 30% to 50% of chloramphenicol present in the blood may appear in the spinal fluid. Two hours after an oral dose of 3 g. one might therefore expect levels between 14 and 24 $\mu\text{g.}$ per ml. to be present.

In Case 1, at the beginning of intrathecal treatment, 10 $\mu\text{g.}$ per ml. was present in the fluid, the level rising gradually with increasing dosage to a maximum of 40 $\mu\text{g.}$ per ml. In Case 2, in which a large initial intrathecal dose was given, the first estimation showed 40 $\mu\text{g.}$ per ml. to be present, and this fell abruptly to 20 $\mu\text{g.}$ per ml. within 24 hours of stopping intrathecal chloramphenicol, although oral administration was continued. The response to treatment was immediate and spectacular, doubtless owing to the high spinal fluid levels achieved at the onset of meningitis, and also to the fact that this case was not complicated by loculated infected fluid. Both cases indicate that maximal titres in the cerebrospinal fluid were not achieved by oral dosage alone.

It would appear, therefore, that an intrathecal dosage of 750 $\mu\text{g.}$ daily, in combination with oral administration, is capable of rapidly raising the cerebrospinal fluid level to a figure in the region of 40 $\mu\text{g.}$ per ml. Such dosage is well tolerated, and to date there have been no undesirable effects.

Summary

Two cases of staphylococcal meningitis resistant to penicillin and streptomycin are described.

Possible reasons for failure to control the infection with aureomycin in Case 1 are examined, and the methods and results of treatment with intrathecal chloramphenicol are discussed.

We are indebted to Mr. Murray Falconer for permission to publish the details of these cases, and to Professor R. Knox for advice and encouragement during the bacteriological investigations. Our thanks are also due to the Department of Clinical Pathology, Guy's Hospital, for giving us details of the cell counts and protein estimations on the fluids, and for their co-operation in other ways; also to Parke, Davis and Co., Middlesex, for supplies of crystalline chloramphenicol.

ADDENDUM.—Since the preparation of our paper, chloramphenicol has been introduced into the lumbar theca in doses as high as 3 mg. without undesirable results. Where intrathecal therapy has been continued for a week or more, two side-effects have been noted in patients so treated.

1. Depression with tearfulness has been seen in all patients. Some have occasionally responded to lumbar puncture and clinical examination with outbursts of weeping.

2. Where a cerebellar tremor has been present due to a pre-existing lesion or to operative interference, this tremor has been markedly accentuated by intrathecal chloramphenicol.

Both these side-effects have disappeared with the cessation of intrathecal therapy.

REFERENCES

- Anderson, K. F. (1951). *J. clin. Path.* In press.
 Bryer, M. S., Schoenbach, E. B., Bliss, E. A., and Chandler, C. A. (1948). *Ann. N.Y. Acad. Sci.*, **51**, 254.
 Ehrlich, J., Gottlieb, D., Burkholder, P. R., Anderson, L. E., and Pridham, T. G. (1948). *J. Bact.*, **56**, 467.
 Harned, B. K., Cunningham, R. W., Clark, M. C., Cosgrove, R., Hive, C. H., McCauley, W. J., Stokey, E., Vessey, R. E., Yuda, N. N., and Subba Row, Y. (1948). *Ann. N.Y. Acad. Sci.*, **51**, 182.
 Olive, T. R. (1949). *Chem. Engng.*, **56**, 107.
 Ross, S., Bischoff, H., Preisser, W., and Orr, W. (1949). *J. clin. Invest.*, **28**, 1050.
 Werner, C. A., Knight, V., and McDermott, W. (1950). *Proc. Soc. exp. Biol., N.Y.*, **74**, 261.

Medical Memorandum

Calcification of the Peritoneum

While it is not very uncommon to find, on radiographic examination, calcification in the pleura and in the pericardium as a result of past tuberculous infection, empyema, or post-traumatic haematoma, extensive calcification of the peritoneum, as distinct from abdominal glands and tumours, would seem from the literature to be rare enough to warrant publication of the following case.

Case Report

A railway worker aged 15 was examined at his home by a tuberculosis officer on November 22, 1947, because of abdominal swelling of three weeks' duration. He had fallen on his chest and abdomen six weeks before and had developed a swelling in his left groin a fortnight later. He now complained of lassitude, anorexia, diarrhoea, night sweats, and loss of weight. His general condition was poor: he was thin and pale, but had no tachycardia or abnormal signs in the chest. His abdomen was distended, fairly tense, with a few visible veins on the abdominal wall. There was deep tenderness and dullness in the left flank, but no fluid thrill. There was a small left inguinal hernia. Tuberculous peritonitis was diagnosed, and he was placed on a sanatorium waiting-list. He remained at home, however, and from April, 1948, reported at three-monthly intervals to the tuberculosis clinic. A radiograph in April showed no abnormality in the chest.