Preliminary Communications

Experimental Allergic Encephalomyelitis: Comparison of Protective Effects of Prednisolone and Corticotrophin

In some fields of clinical practice corticotrophin (A.C.T.H.) and corticosteroid drugs are used as if they were interchangeable. However, several neurologists (Jönsson, von Reis, and Sahlgren, 1951; von Reis, Sahlgren, and Jönsson, 1953; Paulley, 1957, 1959, 1961; Alexander, Berkeley, and Alexander, 1961) have reported that the therapeutic response of acute episodes of multiple (disseminated) sclerosis to cortisone and prednisone is significantly inferior to that obtained with corticotrophin. In the laboratory both corticotrophin (Moyer, Jervis, Black, Koprowski, and Cox, 1950) and cortisone (Kabat, Wolf, and Bezer, 1951, 1952) have been reported to inhibit the development of experimental allergic encephalomyelitis (E.A.E.) if they are given early enough, while we have demonstrated a possible inhibitory effect of corticotrophin even when the hormone is administered after the onset of the illness (Field and Miller, 1961). In the experimental as in the clinical field, however, doubts have been expressed whether corticosteroids are as effective as corticotrophin, and Ross, Colmant, and Böhm (1958) have gone so far as to claim that the administration of prednisolone actually aggravates E.A.E. In view of the complicating factors which influence the activity of corticotrophin and prednisolone (Bush, 1961) it was decided to compare directly the protective effects of these two substances against E.A.E. in the guinea-pig.

MATERIAL AND METHOD

Ninety-two guinea-pigs (400–600 g.) of both sexes were used in two separate experiments. In the first, 12 control animals received a single injection of human white matter (15 mg.) made up in Freund adjuvant, while 20 experimental animals received the same dose of encephalitogenic mixture but also 0.1 ml. (=2 mg.) of prednisolone intramuscularly. The 12 controls received a similar volume of saline, and the injections of prednisolone or saline were continued daily.

In the second experiment 60 animals were injected with a different (less effective) batch of encephalitogenic mixture. Of these, 20 acted as controls and received daily injections of 0.1 ml. of saline; 20 received 10 I.U. of "acthar" gel intramuscularly each day, divided into 4 I.U. in the mornings and 6 I.U. at night; and 20 animals received 0.1 ml. of prednisolone (=2 mg.) intramuscularly daily. This was the dose claimed by Ross *et al.* (1958) to aggravate the ensuing experimental encephalitis. In every instance the first injection was given on the day the encephalitogenic mixture was injected and continued daily until death or the end of the experiment. Animals received a uniform diet of sows' breeding cubes supplemented with greens and were weighed and examined daily. Histological examination of brain and spinal cord was carried out on all animals whether or not they developed neurological signs.

RESULTS

The results of the first experiment, in which the more potent encephalitogenic mixture was used, are set out in Table I.

TABLE I

	Well	Ill	Died	Total Morbid	Total
Controls	0	3	9	12	12
Prednisolone-treated	5	8	7	15	20

χ^2 test, no significant difference. $P=0.110$.

If the numbers of animals which fell sick are compared for the control and experimental (prednisolone-treated) animals the figures are 12 out of 12 and 15 out of 20 respectively. Because of the 100% morbidity among the control animals Fisher's exact χ^2 test was applied and gave $P=0.077$ —that is, there is a more than 1 in 20 chance that this apparent protective effect was fortuitous. However, when mortality is tested against survival in both groups it would appear that prednisolone does increase the chance of survival ($P=0.02-0.01$). Death is probably due to fortuitous localization of the disease process in vital centres, and this would be more likely to occur when lesions are widespread.

From the first experiment, therefore, it may be concluded that prednisolone has an inhibitory effect upon the development of guinea-pig E.A.E. in so far as it tends to prevent a fatal illness, and that it possibly also has a limited effect in reducing clinical morbidity.

The results of the second experiment, in which a less potent encephalitogenic mixture was used, are set out in Table II. Corticotrophin has a clear-cut sparing

TABLE II

	Well	Ill	Died	Total Morbid	Total
Controls	1	16	3	19	20
Corticotrophin	10	10	0	10	20
Prednisolone	4	14	2	16	20

χ^2 test.

Controls tested against corticotrophin, significant, $P=0.01-0.001$.

prednisolone, not significant, $P=0.171$.

Prednisolone tested against corticotrophin, significant, $P=0.05-0.02$.

effect on the development of E.A.E. ($P=0.01-0.001$), confirming a previous observation (Field and Miller, 1961). Prednisolone, on the other hand, was not effective in reducing morbidity incidence ($P=0.342$; $P=0.171$ with a one-tail probability test). When prednisolone is compared with corticotrophin for efficiency in preventing the onset of disease the latter is found to be more effective ($P=0.05-0.02$).

DISCUSSION

The results described above afford some experimental evidence for the clinical suspicion that corticotrophin may be more effective than adrenocortical substitutes in the suppression of inflammatory demyelinating lesions of the nervous system. Until more is known about the many factors which influence adrenal hormone discharge and about the possible functions of the "amorphous fraction" of cortical extracts it would probably be prudent in this connexion to rely on the physiological adrenal stimulant—corticotrophin—which has been shown to exert a beneficial effect in acute exacerbations of multiple sclerosis (Miller, Newell, and Ridley, 1961) rather than on the putative corticosteroid substitute.

The claim of Ross *et al.* (1958) that prednisolone intensified E.A.E. contradicts all previous reports on the activity of corticosteroids in this disorder. These authors invoked species differences, the use of different corticoid preparations, and anomalies of dosage to explain their findings. Their six prednisolone-treated

animals (given 2 mg. on the day preceding and on the day of the encephalitogenic injection) were reported as manifesting an early onset and unusual severity of neurological disturbance, while the disease was also thought to be histologically intensified. In a much larger series of prednisolone-treated animals (40) we could not substantiate these findings, though it must be conceded that we administered 0.1 ml. (=2 mg.) daily on the lines employed in clinical trials.

SUMMARY

Evidence is presented that corticotrophin is more effective than prednisolone in mitigating the experimental encephalomyelitis induced in guinea-pigs by the inoculation of brain emulsion with Freund adjuvant.

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A three-stage campaign against cholera is to be carried out in Hong Kong. The first stage will be among the people who live permanently afloat in junks and sampans. Inoculating teams will be operating from launches for two weeks in the typhoon shelters and junk anchorages of Hong Kong, Kowloon, and the New Territories. The campaign will first be directed at the urban population, and it is expected that this phase will be completed by the end of March. Inoculations will be available at all Government clinics, dispensaries and inoculation centres. In addition, a number of temporary centres will be opened. The last stage of the campaign will begin in April when mobile teams will cover the rural areas of the New Territories. Inoculation will be free, and only one injection is required. During the recent outbreak of cholera in Hong Kong 129 cases were confirmed and 15 people died. Between August 14 and August 25 more than 1,500,000 people—roughly half the population—were inoculated against cholera. A report on the outbreak by Dr. D. J. M. MacKenzie, Director of Medical and Health Services, recommended that an inoculation campaign should be carried out, aimed at protecting 70% or more of the population.

Medical Memoranda

Liver Flukes in Common Bile-duct

Human infection with the liver fluke *Fasciola hepatica* has only rarely been reported in Britain, although Facey and Marsden (1960) reported a small outbreak in Ringwood, Hants. In the case reported five adult flukes were found in the common bile-duct at operation.

CASE REPORT

A married woman of 28 had lived on a farm in south-east Dorset for the previous nine years, and had always been healthy; but about July, 1959, she started to get attacks of pain in the epigastrium (no radiation). The pain was sharp and fairly severe, lasting several hours, and only partially relieved by tablets given by her doctor. There was slight vomiting and slight jaundice, but no urticaria. The attacks kept her from work only for two to three days. They recurred every month or two.

On admission to hospital on December 29, 1960, she complained of a constant right hypochondrial pain, which was severe at times, causing her to vomit, but was continually present as an ache. On examination she was found to be slightly jaundiced. C.N.S., alimentary system, and cardiovascular system N.A.D.

At operation the stomach, duodenum, pancreas, and liver appeared intact. The gall-bladder was thin-walled, and no stones were felt. The common bile-duct was dilated. This was opened and found to be thick-walled, with a granulomatous lining, and five live flukes of *F. hepatica* were removed. The gall-bladder contained biliary mud blocking the cystic duct. Cholecystectomy was performed, and the duct drained by the usual T tube.

The patient made good progress for nine days after operation, during which fluid from her T tube was examined for ova and flukes. Very scanty *F. hepatica* ova were present in some of the specimens (mostly degenerate forms). On January 12, 1961, there was onset of peritonitis, with shock-like condition, severe electrolyte disturbance, and a very low serum sodium and chloride. Gastric suction and intravenous saline were instituted without avail. The usual steps were taken to combat this condition, but, despite all, the patient deteriorated gradually and died on January 21.

Investigations.—Blood examination on January 4 demonstrated eosinophilia (11%), and examination of the faeces by the formol-ether concentration method disclosed a moderate number of *F. hepatica*. E.C.G. showed a normally functioning heart.

Necropsy.—The liver weighed 52 oz. (1,475 g.). Its surface was irregularly mottled with pale and haemorrhagic areas. Cut surface showed subcapsular broad wedge-shaped areas of congestion studded with crumbling beads of calcium, varying in size from a pin's head to a pea. They were far more numerous in the right lobe. The bile-ducts appeared to be free of residual liver flukes. Histological report: Numerous granulomatous lesions composed of central amorphous and partly calcified areas of variable size surrounded by thick hyalinized capsules, which are sparsely infiltrated by round cells and eosinophils. The liver parenchyma was grossly necrotic. Bile-duct: Granulomatous inflammation of the walls with epithelial loss. Pancreatic duct normal, unobstructed. Spleen normal. Peritoneal space: Localized biliary peritonitis in epigastrium bounded anteriorly by abdominal wall, posteriorly by anterior wall of stomach and omentum. *Cause of death:* Fulminating bronchopneumonia following cholecystectomy for fascioliasis (*F. hepatica*).