laris and ciliary regions, which are supplied by this nerve. In Sigwald's case the limitation of ocular movement as determined by electrical tests was due to paralysis and not to spasm of the eye muscles. In the present case myograms of the facial muscles after the patient had improved showed little change from normal, although the response was more sluggish on the side of the palsy. Gunther and Walker (1943) found that in local tetanus myograms showed action currents present even at rest, with exaggerated summation during contraction and after a noise stimulus.

Baker (1943) reviewed the early literature on the pathological changes; it was extremely conflicting. It may be said in general that no definite nerve lesion had been found except for haemorrhages which might have been due to violent spasms. He recorded the case of a farmer who died of general tetanus after the removal of a wart from the hand. He died with irregular heart and respiratory action not associated with spasms and apparently due to failure of the respiratory and cardiovascular centres in the medulla. Sections of the brain showed destructive changes in the motor nuclei of the fifth nerve and in the dorsal nucleus of the tenth nerve. Minimal changes were found elsewhere, and there was no significant lesion of the anterior horn cells. Baker suggested that selective nuclear damage may often be a cause of death in tetanus, and is usually ascribed to respiratory spasms rather than paralysis. Such localized nuclear changes would certainly account for the bizarre ocular signs found in the present and other reported cases. Baker's patient had no ocular signs, and no damage was observed in any of the ocular nuclei. Reversible damage to the nucleus would account for the temporary nature of the paresis, and the extent of the damage would determine its duration.

Treatment

No matter how mild the initial symptoms may be, every case of cephalic tetanus should be treated as a potential case of general tetanus. Antitoxin should be given, 100,000 units intravenously and 100,000 intramuscularly, as soon as possible after the signs appear. Adequate sedation to prevent spasms, a special nurse, and a darkened room are essential. A high-protein high-calorie diet should also be given, by nasal tube if the trismus is intense.

Tongue-biting may be troublesome and the teeth may have to be wedged with gauze. The general nursing of the patient is often of greater importance than any specific therapy. Mephnesin ("myanesin") has been used to overcome the spasms and it is useful for feeding, but there is a danger of haemolysis (Torrens *et al.*, 1948).

Summary

Cephalic tetanus is defined as tetanus after head or face injury which is accompanied by some cranial-nerve palsy. A case with recovery is described.

The clinical picture involves bizarre palsies of cranial nerves III, IV, VI, VII, IX-X, XII, single or in any combination. Exceptionally, the chorda tympani may be involved. It is suggested that the condition results in transient or permanent damage to the cranial-nerve nuclei.

Prognosis is better than earlier reports suggest if prompt treatment with antitetanic serum and sedation is carried out.

I should like to thank Dr. A. M. Cooke for permission to publish this case and for helpful criticism in connexion with this paper. I should also like to thank Mr. J. P. F. Lloyd, of the Oxford Eye Hospital, for the eye examinations.

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ESTIMATION OF TRYPSIN IN DUODENAL JUICE

BY

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It has been shown (Andersen, 1942) that trypsin is always low or absent from the duodenal juice in fibrocystic disease of the pancreas, and this forms a reliable basis of diagnosis. Andersen has pointed out that the estimation of proteolytic enzyme need not be exact, as there is a fairly wide difference between the amounts found in fibrocystic disease and those found in other conditions, and she therefore adopted a simple modification of Fermi's method (1906) for routine estimation of trypsin. For some time we have been using a modification of the method of d'Este Emery (quoted by Harrison, 1930), which utilizes the gelatin of an x-ray film as substrate for estimating trypsin in duodenal juice and faeces. It has the advantage of ease and simplicity and is accurate enough for diagnosis. A similar method has been described for faeces by Shwachman, Patterson, and Laguna (1949), who appear to have overlooked the prior description.

In this investigation, using the method of James (1951) for aspirating duodenal juice, the normal levels of trypsin have been determined by the x-ray film method; a comparison has been made between this method, the gelatin method of Andersen, and a more accurate method based upon the proteolytic action of trypsin on casein (Northrop, 1924).

Method of Intubation

An ordinary duodenal tube containing a magnet in the tip was used as in the method of James (1951). In an infant the 10 o'clock feed was not given; in the older child breakfast was omitted so that intubation was performed with the stomach empty. There was no restriction of fluids. Sedatives were unnecessary; in several instances where one was given a longer time was needed to obtain a satisfactory specimen of duodenal juice, possibly because of the diminished gastric peristalsis or because the patients were more restless when the tube was passed. Whenever possible the tube was passed through the nose, as better control was thus obtained. In most cases a small plotting compass— $\frac{1}{2}$ in. (1.25 cm.)—with a non-dipping needle was used to locate the tip. The point at which maximum oscillation of the needle occurred was found by experience to be always

$\frac{1}{4}$ in. (1.25 cm.) away from the tip as seen by a radiograph. The position of the tip of the tube could not be accurately defined when it lay directly below the rectus muscles unless its position was approached by a deep palpation, with the compass underneath the rectus muscle. In all cases, as soon as the tip was thought to be in the duodenum, its position was confirmed by a radiograph. Of the 48 cases investigated only 2 were unsuccessful at the first attempt, and when repeated no difficulty was encountered. The time taken for intubation varied from 20 minutes to two hours, the majority taking about one hour.

Whenever possible the duodenal juice was allowed to drip out without suction, but it was sometimes found desirable, especially when the fluid was viscous, to apply gentle suction with a syringe to reduce the time of collection. The juice was always collected in approximately 0.5-ml. amounts in separate tubes, thus avoiding the possibility of gastric juice contaminating the whole specimen. The most characteristic specimens of duodenal juice, as judged by colour, clearness, absence of mucus, and the pH, could then be pooled and used for analysis. Estimation of trypsin was performed as soon as possible after the duodenal juice had been obtained, being kept ice-cold in the meanwhile. If the analysis were performed the following day, the specimen was kept frozen. Under these conditions there seems to be no diminution in tryptic activity up to 48 hours.

Estimation of Trypsin

Gelatin Method

Andersen and Early's (1942) directions were followed, except that U.S.P. gelatin was used instead of Knox gelatin.

X-ray-film Method

Serial dilutions of duodenal juice from 1 in 2 to 1 in 4,096 are prepared by adding accurately 0.2 ml. of the fluid to the first of the series of 12 test-tubes—2 by $\frac{1}{4}$ in. (5 by 0.6 cm.)—each containing 0.2 ml. of 1% crystalline sodium carbonate solution. After mixing, exactly 0.2 ml. is removed and added to the second tube, and so on up to the sixth tube, from which 0.2 ml. of the mixture is discarded. To the seventh tube is added 0.2 ml. of duodenal juice already diluted 1 in 32 with 1% sodium carbonate solution, and serial dilution effected as before up to the twelfth tube. In this way the large errors which may occur in carrying out 12 successive serial dilutions were avoided.

With a Pasteur pipette, 1 drop (approximately 0.05 ml.) of undiluted duodenal juice is placed upon a 5-cm. square of unused x-ray film. Then a drop of each of the serial dilutions of the juice are separately placed thereon, starting with the weakest dilution, and finally 1 drop of 1% sodium carbonate solution as a control. The spots are numbered for convenience from 1 (undiluted juice) to 13. Care is taken in delivery that each drop occupies approximately the same area of film. The latter is then transferred to a covered Petri dish containing a moist pad of cotton-wool to prevent drying, and incubated at 37° C. for 30 minutes. The film is then placed in the freezing chamber of a refrigerator for 20 minutes. In all transfers care is taken that the drops are not displaced. Finally the film is washed in a stream of cold water from the tap. If sufficient trypsin is present in any of the drops, a clear circular translucent area is left after the drop is washed off. The control spot remains unaffected, apart from a slight roughening of the surface.

It is essential to place the film in the freezing chamber of the refrigerator before washing in tap water, since the gelatin of the film may be softened by any fluid at 37° C., and if not well cooled may be subsequently washed off in too strong a stream of cold water. The end-point is taken as the highest dilution of duodenal juice giving a completely translucent area. For simplicity the spot number may be reported instead of the dilution. Although used or unused film may be utilized, we have found it preferable to employ unused Kodak film. Other makes give slightly different

results. Duplicate estimations were always performed and there was no difficulty in getting good agreement between duplicates.

Casein Method

The method of Northrop (1924) was used. To approximately 17 ml. of distilled water is added 1 g. of soluble casein B.P. and the mixture is well shaken to dissolve. Then 6 ml. of 2% crystalline sodium carbonate solution is added, followed by 0.5 ml. of duodenal juice, and the solution made up to 25 ml. in a volumetric flask with distilled water. After inverting several times to mix, 1 ml. is at once removed as a blank, placed in a test-tube containing 2 ml. of water, and 2.5 ml. of 10% trichloroacetic acid solution is added. After standing for five minutes the precipitated protein is filtered off. The remainder in the volumetric flask is incubated at 37° C. for 45 minutes, after which 1 ml. is removed and treated as before. From both these filtrates 3 ml. is removed for the estimation of nitrogen by the micro-Kjeldahl method. From the difference between the two results the number of milligrams of non-protein nitrogen produced by the action of 1 ml. of duodenal juice on 4% casein solution under these conditions is calculated.

Material

Duodenal juice was obtained from 48 infants and children. The total included 15 normal children aged 3 weeks to 2 years; 8 with fibrocystic disease of the pancreas, aged 3 to 14 months; 20 with symptoms suggestive of coeliac disease or fibrocystic disease, aged 7 months to 2½ years; and 5 marasmic infants aged 14 months to 3 years. All specimens of juice except two were golden yellow due to bile, the two exceptions being light green. The specimens from cases of fibrocystic disease were thick viscous fluids, whereas all the others were thin and mainly clear. The pH ranged from 7 to 8.3, except for one which was pH 5.8, although this gave a normal level of trypsin.

Results

The results of the estimation of proteolytic enzyme by the x-ray-film method in the duodenal juice from 48 infants are shown in the Chart. In 7 of the 48 specimens no trypsin could be detected. One further case showed a very low amount of trypsin, giving an end-point in a dilution of 1 in 2 only (spot 2). All these were diagnosed as fibrocystic disease and treated as such. The remainder showed end-points at spots 6 to 11 inclusive, with the largest number at spot 7. There is a wide margin, therefore, between the cases of fibrocystic disease and the others, and this result is also obtained with Andersen's gelatin method as shown in the Table. There were no cases of coeliac disease, marasmus, or possible fibrocystic disease (other than proved cases) in which the amounts of proteolytic enzyme fell below the lowest level found in normal infants.

Chart showing trypsin levels of duodenal juice in 48 infants.

Comparison of X-ray-film Method, Andersen's Method, and Casein Method

In a preliminary experiment, using serial dilutions of pancreatin (B.P.) containing from 200 to 0.2 mg./ml., parallel results were obtained by all methods. The amount of trypsin in the duodenal juice from 14 infants was estimated by

Trypsin in Duodenal Juice of Infants. Comparison of the Three Methods

Specimen No.	X-ray-film Method Spot No.	Gelatin Method (Andersen) Tube No.	Casein Method mg.N/ml.
1	Nil	Nil	Nil
2	"	"	"
3	2	9	4
4	6	4	52
5	6	3	67
6	7	4	67
7	7	2	85
8	7	2	96
9	7	2	115
10	7	2	124
11	8	4	54
12	8	3	83
13	8	3	88
14	10	4	93

all three methods (see Table). Although the results given for the x-ray-film method and Andersen's gelatin method show a close relationship, they are not exactly parallel. Of the three cases of fibrocystic disease, two showed no trypsin by both methods, and the third gave the same low result. However, among the normals, specimens of duodenal juice which gave identical levels of trypsin by the x-ray-film method gave levels that differed by as much as three tubes by Andersen's method. These differences are to be expected, since neither method can be considered accurate as compared with the casein method. For example, duodenal juice with an end-point at spot 7 can give a range of 67 to 124 mg. of nitrogen by the casein method and an even wider range is obtained with specimens of duodenal juice giving an end-point at spot 8. A similar wide variation is shown with Andersen's method. Results are, however, generally parallel with all three methods.

The results obtained in this investigation show that the x-ray-film method is entirely adequate for the differentiation of fibrocystic disease from other conditions. It is especially suitable as a routine clinical laboratory procedure; it is simple to perform, less juice is required, and it is less time-consuming than other methods, since results can be obtained within an hour. The use of James's method of intubation in conjunction with the x-ray-film method of estimation of proteolytic enzyme makes the investigation of suspected cases of fibrocystic disease of the pancreas relatively easy.

Summary

The amounts of trypsin in the duodenal juice of 48 infants and children, including 8 suffering from fibrocystic disease of the pancreas, have been determined by a method utilizing the gelatin of x-ray film as substrate. This method, which has advantages of ease and simplicity, has been compared with the gelatin method of Andersen as well as with a more accurate casein method, and has been shown to be accurate enough for the diagnosis of fibrocystic disease of the pancreas.

We would like to thank Dr. A. H. James for originally demonstrating his method of duodenal intubation, and Dr. W. F. Young and Dr. A. H. James for obtaining some of the specimens of duodenal juice.

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On the day of the total eclipse of the sun a million Hindu pilgrims bathed in the sacred lakes at Kurukshetra (East Punjab). *The Times* special correspondent (February 26) pays special tribute to the excellent public health arrangements. All pilgrims were inoculated compulsorily against cholera, all water (including the sacred lakes) chlorinated, and D.D.T. sprayed throughout the pilgrim camps, and no outbreak of disease has been reported.

ACUTE IDIOPATHIC PAROXYSMAL COLD HAEMOGLOBINURIA OF NON-SYPHILITIC TYPE IN A CHILD

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Haemoglobinuria, in contrast to haematuria, is a clinical sign which is so seldom seen in paediatric practice that its appearance is a matter of unusual interest and significance. In the great majority of cases there is either clinical or serological evidence of congenital or acquired syphilis, although in the last decade this disease has become increasingly rare.

The syndromes of paroxysmal cold haemoglobinuria associated with cold haemagglutination and paroxysmal cold haemoglobinuria as found in syphilitics were first clearly distinguished by Stats and Wasserman (1943). Cases of paroxysmal haemoglobinuria induced by exposure of the body to cold in which clinical and serological evidence of syphilis is lacking have been described by Stats and Wasserman (1943), Stats, Wasserman, and Rosenthal (1948), Malley and Hickey (1949), and Ferriman *et al.* (1951). These last observers, in an analysis of the reported cases, could find only 12 in which the aetiology is unknown. All were in the later age groups, only one (aged 38) being under 50.

Case Report

A boy aged 3½ was admitted to hospital with a history that on three days during the previous fortnight he had been out with his father (a general practitioner) on his rounds and, on returning home, complained of a headache and vague abdominal pain, had what appeared to be a rigor, and immediately afterwards passed dark urine "like burgundy." On each occasion very low environmental temperatures were present. His symptoms cleared completely in a matter of minutes and his urine became clear in about 12–18 hours. It had also been noticed that the child developed a dusky blue discoloration and puffiness of the hands and ears on exposure to cold, suggestive of Raynaud's phenomenon.

On admission the child was of normal size and weight, active, and of good nutrition. He was pale, with a slight icteric tinge. Neither liver nor spleen was palpable. His pulse was 70 and regular and the blood pressure 110/70. Pulsation of arteries in the upper and lower limbs was normal. There was no history of any previous illness. The family history was irrelevant.

Investigation of Urine.—During a paroxysm the urine was the colour of burgundy and of high specific gravity (1026–1030). Albumin was present. The benzidine test was positive and spectroscopy showed the presence of oxyhaemoglobin. No red cells were seen on microscopical examination. Immediately after an attack a trace of bilirubin was detected and urobilinogen was found in an increased amount. In the intervals between attacks spectroscopy did not show any oxyhaemoglobin and albumin was absent. Haemosiderin was present on two occasions only (Prussian-blue reaction).