IMPORTANCE OF INTRAVENOUS INJECTION OF DIPHTHERIA ANTISERUM

A. TASMAN
Ph.D. Leyden

J. E. MINKENHOF
Arts, Amsterdam

H. H. VINK
Vet.D. Utrecht

A. C. BRANDWIJK
M.D. Utrecht

L. SMITH

From the National Institute of Public Health, Utrecht, and the Infectious Diseases Department of the Wilhelmina Hospital, Amsterdam

IT may be regarded as an accepted fact that, when diphtheria is suspected, the patient ought to be injected with antidiphtheritic serum without delay. It is indefensible to wait until the result of the bacteriological examination is known. The bacteriological diagnosis should only confirm the bedside diagnosis made by the practitioner. The desirability of the early injection of antiserum is discussed in more detail by Tasman and Lansberg (1957).

Opinions differ, however, about the best way of injecting antiserum. It is generally deemed advisable to inject it intramuscularly and only to resort to intravenous injection in severe cases (Chatterjee 1954, Gorter 1948, Hyman 1948, Cecil and Loeb 1952, Herderschee 1953, Christie 1955). Lepintre (1954) goes so far as to maintain that the antiserum must never be injected intravenously but should always be injected either intramuscularly or subcutaneously.

Tasman et al. (1954, 1955) have shown that the saliva of diphtheria patients usually contains diphtheria toxin. When purified diphtheria antiserum (obtained from horses) is injected intramuscularly, the antitoxin is sooner or later excreted in the saliva via the salivary glands and will then neutralise this toxin. Actively immunised persons do not excrete the (homologous) antitoxin in their blood through the salivary glands.

We thought it worth while to investigate this excretion of antitoxin through the salivary glands more closely and, more especially, to find if there were any differences in this respect between patients injected intramuscularly and those injected intravenously.

Excretion of Antitoxin in Saliva

The saliva of patients was tested for antitoxin at various times after either intramuscular or intravenous injection of purified diphtheria antiserum. The results are shown in table I.[1]

This excretion of antitoxin in the saliva via the salivary glands is very important. Our investigation showed that, after only 30 minutes, the saliva of all but one of the intravenously injected patients already contained antitoxin and had therefore become atoxic.[2] In other words, from this moment no more toxin would be absorbed from the saliva flowing over the inflamed and damaged membranes of the tonsils and the pharynx. In the intramuscularly injected patients this rapid excretion of antitoxin was exceptional; usually it was a long time before the saliva contained any antitoxin. During this interval diphtheria toxin could be absorbed from the saliva through the inflamed membranes of the tonsils and the pharynx.

TABLE I—EXCRETION OF ANTITOXIN IN SALIVA

Intramuscular injection (average dose 28,000 A.U.)		Intravenous injection (average dose 12,000 A.U.)	
No. of cases	Time taken for antitoxin to reach saliva	No. of cases	Time taken for antitoxin to reach saliva
1	30 min.	12	30 min.
3	4–5 hr.	1	6 hr.
3	10 hr.		
9	1–9 days		

This difference between the two groups of patients is the more striking because the intramuscularly injected patients received on the average 2·5 times as much antitoxin as did those injected intravenously.

Blood-serum Titres

We took samples of blood from patients before they received the antidiphtheritic serum either intravenously or intramuscularly and at set times after its injection, and determined the antitoxin titres. Table II shows the results in a greatly abridged form; the titres are expressed as percentages of the highest titre found.

1. For lack of space the numerical results of this investigation are given only in summarised form here. We hope to publish the full details elsewhere.
2. The first samples of saliva were taken 30 minutes after the serum had been injected. It is by no means inconceivable that antitoxin may appear in the saliva of the intravenously injected patients even before that time.

TABLE II—ANTITOXIN CONTENT OF BLOOD-SERUM OF PATIENTS INJECTED INTRAMUSCULARLY AND INTRAVENOUSLY

Intramuscular injection														Intravenous injection								
Case no.	Dose of antitoxin (A.U.)	Titres expressed as % of highest titre* Days after injection												Case no.	Dose of antitoxin (A.U.)	Titres expressed as % of highest titre found* Days after injection							
		1	2	4	6	8	10	12	14	16	18	20	22			1	2	4	6	8	10	12	14
1	60,000	33	56	100	100	78	67	56	44	33	22	17	11	1	10,000	100	89	55	43	43	43	37	17
2	40,000	44	74	100	96	..	..	..	..	..	..	..	..	2	10,000	..	100	71	43	28	20	14	6
3	20,000	27	38	100	80	..	..	..	..	..	..	..	..	3	10,000	100	..	66	37	30	30	30	20
4	40,000	44	75	100	..	75	50	25	..	19	13	..	5	4	10,000	100	78	56	56	44	44	..	22
5	40,000	100	100	67	..	50	33	..	..	..	..	..	..	5	10,000	100	78	56	44	44	..	11	4
6	20,000	40	60	70	100	80	..	48	..	34	..	..	12	6	10,000	100	82	64	64	..	25	25	..
7	20,000	23	46	92	100	77	53	..	30	23	..	20	18	7	10,000	100	100	..	..	63	33	13	..
8	20,000	100	..	85	63	..	38	..	21	11	8	..	..	8	10,000	100	55	55	37	..	10	..	..
9	20,000	77	100	100	77	50	..	..	43	37	..	..	..	9	10,000	100	64	44	33	22	17	13	..
10	20,000	35	83	100	70	56	50	..	40	..	20	..	..	10	10,000	100	60	40	30	24	20	16	..
11	80,000	30	64	100	64	50	40	24	..	14	10	..	..	11	10,000	100	62	38	25	19	13	7	7
12	20,000	50	80	100	100	70	..	36	..	28	..	26	..	12	10,000	100	..	56	44	28	17	11	5
13	80,000	23	100	85	..	40	34	34	..	18	..	..	9	13	10,000	100	98	98	58	29	29	21	16
14	20,000	60	96	100	64	54	40	24	..	14	..	9	..	14	10,000	100	90	..	..	37	24	..	17
15	20,000	45	77	100	70	54	..	37	..	30	..	..	11	15	10,000	100	100	25	25	25	..	..	10
16	20,000	56	100	46	46	46	..	25	..	25	..	25	..	16	10,000	100	..	67	..	..	30	20	18
17	8000	67	100	85	60	47	32	22	22	15	9	..	..	17	10,000	100	91	40	40	27	18	14	9
														18	10,000	100	50	30	17	10	6	4	3
														19	20,000	100	60	40	34	27	18	12	10
														20	10,000	100	56	46	37	25	19	11	13
	Mean	50	78	90	78	59	44	33	(33)	23	(13)	(19)	(11)		Mean	100	77	53	39	31	23	16	12

* In these calculations the titres before the injection of antiserum have been subtracted from the titres found after the injection because we are here concerned solely with the course of the antitoxin level resulting from the injection of heterologous diphtheria antitoxin

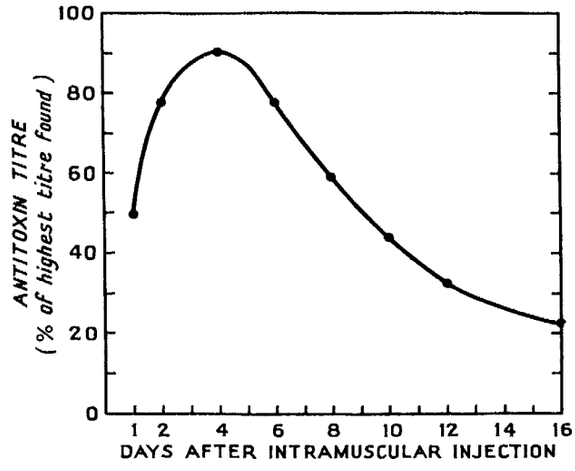

Fig. 1—Mean titres of antitoxin in serum after intramuscular injection.

The absorption and excretion of antitoxin from antiserum injected either intramuscularly or intravenously varied greatly from one patient to another. It is, however, obvious that the antiserum is absorbed much more slowly after intramuscular injection, the highest titre in the serum not being reached until after 2–6 days on the average.

The average titres expressed as percentages of the highest titre found are shown after intramuscular injection (fig. 1) and after intravenous injection (fig. 2).

One is immediately struck by the great similarity between the two curves of antitoxin titres. This is brought out even more clearly when the percentage titres (expressed as percentages of the highest titre found) are plotted on a logarithmic scale and in both cases the

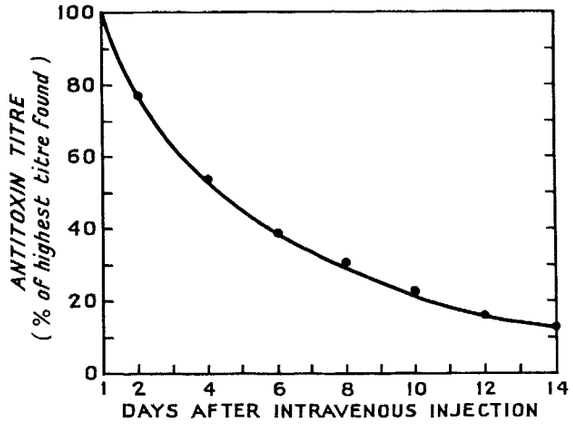

Fig. 2—Mean titres of antitoxin in serum after intravenous injection.

average titre found 4 days after the injection is taken as 100%.[3]

Fig. 3 gives the results of these calculations. Here the two lines practically coincide from the fourth day after the injection of the antiserum. From this we conclude that the antitoxin in the intravenously injected antiserum is not excreted more rapidly than that in the intramuscularly injected antiserum. Hence in this respect there is no reason why the intramuscular injection of antiserum should be preferred to the intravenous. Madsen (1936) reached the same conclusion after a similar investigation.

In view of these results the obvious thing would have been to test the presumptive superiority of the intravenous method, compared with the intramuscular, on a large number of patients; but such a plan would meet with many difficulties, the chief obstacle being the small number of diphtheria patients available at present. For this reason we confined ourselves to experiments with laboratory animals.

3. On the average the absorption of intramuscularly injected serum is complete at the end of this period. The subsequent excretion of this intramuscularly injected antitoxin is strictly comparable to that of the intravenously injected antitoxin.

Design and Application of an "Animal Model"

When a guineapig is injected with a certain number of lethal doses of diphtheria toxin it can be saved from death or protected from typical morbid symptoms by injecting antitoxin before or shortly after the injection of toxin. When the quantities of toxin and antitoxin to be injected have been fixed, the interval between the injection of toxin and that of antitoxin will determine the clinical picture. If a short interval is chosen, the guineapigs will show only very slight transient symptoms or

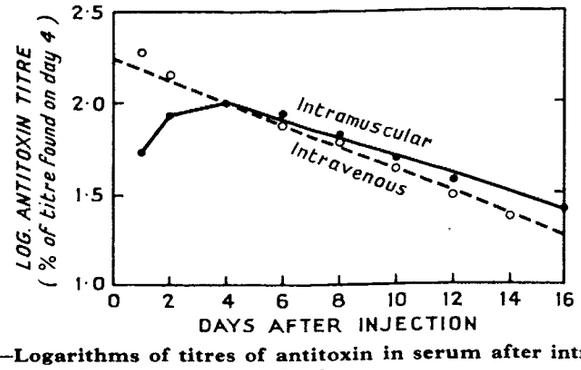

Fig. 3—Logarithms of titres of antitoxin in serum after intramuscular and after intravenous injection.

none at all. If a long interval is allowed, most of the guineapigs will show severe manifestations (much loss of weight, severe cardiac manifestations, and paralyses) or most of them will die.

In other words, therefore, a suitable choice of the three determining factors—amount of toxin, amount of antitoxin, and length of time between the two injections—will make it possible to control the clinical picture so that special attention can be given to different aspects of it.

A well-standardised "animal model" of this kind will also provide an excellent means of ascertaining experimentally the influence of a fourth factor—e.g., the route by which the antitoxin is injected.

After preliminary experiments we used male[4] guineapigs weighing 300–350 g. in all our further investigations. These were injected subcutaneously with 5 M.L.D. of diphtheria toxin at the beginning of each experiment. A certain number of hours afterwards the guineapigs were injected with 1 antitoxic unit of diphtheria antitoxin, half of them receiving this antitoxin intramuscularly and the

4. The intravenous injection of antiserum is technically much simpler in the male guineapig than in the female. The antiserum was injected into a branch of the femoral vein in the subcutaneous tissue of the inside of the thigh.

TABLE III—CHANGES OF WEIGHT OF GUINEAPIGS INTRAVENOUSLY OR INTRAMUSCULARLY INJECTED WITH DIPHTHERIA ANTITOXIN

Days elapsed after the injection or serum	Mean weight of guineapigs on consecutive days (g.)	
	Intravenously injected	Intramuscularly injected
0	306 (*100%*)	307 (*100%*)
1	316 (*103%*)	307 (*100%*)
2	308 (*101%*)	299 (*97·4%*)
3	305 (*99·7%*)	291 (*94·8%*)
4	291 (*95%*)	286 (*93·1%*)
5	294 (*96%*)	281 (*91·5%*)
6	293 (*95·7%*)	275 (*89·6%*)
7	289 (*94·4%*)	271 (*88%*)
8	289 (*94·4%*)	262 (*85·3%*)
9	286 (*93%*)	255 (*83%*)
10	289 (*94·4%*)	255 (*83%*)
11	293 (*95·7%*)	256 (*83·3%*)

other half intravenously. By determining beforehand the time required for the different injections it was possible to ensure that the time between the injection of the toxin and that of the antitoxin was the same for every guineapig in each experiment. In each experiment some guineapigs were injected with toxin only, without a subsequent injection of antitoxin, as controls. These controls were always dead on the 2nd or 3rd day after the injection of the toxin.

Experiment I

Two groups of guineapigs received 5 M.L.D. of diphtheria toxin subcutaneously; after 9 hours 1 A.U. of antitoxin was injected intramuscularly into 15 guineapigs and intravenously into 15 others. Of the intramuscularly injected guineapigs 4 died, after 2, 6, 7, and 9 days, whereas 4 of those injected intravenously died after 6, 7, 8, and 10 days. All the guineapigs were weighed twice daily, after which the mean weight of each group was calculated. Table III gives the results of this experiment, which are graphically represented in fig. 4. The initial weight per group is here taken as 100.

From these results we may conclude that the guineapigs injected intramuscularly with antiserum showed a greater and more prolonged loss of weight than did those injected intravenously. Therefore the general condition of the former

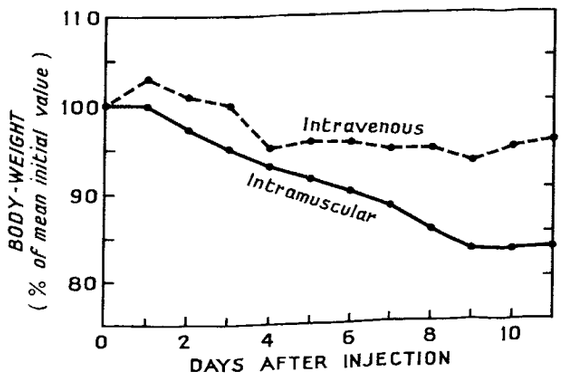

Fig. 4—Weights of guineapigs after intramuscular and intravenous injection of antidiphtheritic serum.

throughout the experiment was considerably worse than that of the latter under otherwise identical conditions.

Experiment II

If the interval between the injection of toxin and that of antitoxin is long, one may expect a greater number of deaths in both groups of guineapigs. To test the difference between the intramuscular and the intravenous injection of antiserum, two groups of 65 guineapigs were subcutaneously injected with 5 M.L.D. of diphtheria toxin. After $9^1/_2$ hours they were given 1 A.U. of antitoxin. One group was injected intravenously with the antitoxin and the other intramuscularly. The results of this experiment are given in table IV.

These figures suggest that guineapigs previously treated with toxin have a considerably better chance of survival when injected intravenously than when injected intramuscularly with the same dose of diphtheria antitoxin.

It may at first sight seem strange that lengthening of the interval between the toxin and antitoxin injections by only 30 min. (from 9 to $9^1/_2$ hr.) should have such a striking effect on the mortality-rate of the guineapigs. In experiment I (9 hr. interval) only 4 guineapigs died in each of the two groups (about 25%), whereas in experiment II ($9^1/_2$ hr. interval) 52% of the guineapigs died in one group and 86% in the other. This result is closely related to the experimental conditions. If we had chosen different doses of toxin and of antitoxin and a different interval we would have obtained results quantitatively different from these. For further general considerations on this " animal model " see below.

Experiment III

It was to be expected that the use of a considerably shorter interval between the injection of toxin and that of antitoxin would lead to fewer deaths and provide information about the effect of the method of injection on the paralyses which appear in a later stage of diphtheria.

TABLE IV—MORTALITY AMONG GUINEAPIGS INTRAVENOUSLY OR INTRAMUSCULARLY INJECTED WITH DIPHTHERIA ANTITOXIN, DIPHTHERIA TOXIN HAVING BEEN INJECTED $9^1/_2$ HOURS BEFORE. NUMBER OF ANIMALS IN EACH GROUP 65; TOXIN DOSE 5 M.L.D.; ANTITOXIN DOSE $\frac{1}{4}$ A.U.

Days elapsed after injection of serum	Deaths in each group of 65 daily	
	Injected intravenously	Injected intramuscularly
1	0	7
2	0	0
3	1	1
4	7	6
5	7	6
6	5	4
7	3	5
8	2	5
9	1	2
10	1	6
11	2	4
12	0	6
13	4	2
14	0	1
15	1	1
Total	34	56

For this purpose two groups of 46 and 48 guineapigs were given 5 M.L.D. of antitoxin subcutaneously and $6^1/_2$ hours later were injected either intravenously or intramuscularly with 1 A.U. of antitoxin. Table V shows the incidence of paralysis: this primarily and chiefly affected the hind legs. Fig. 5 shows these results.

These data show that, under the conditions of the experiment, paralysis came on fairly late: after 16 days among the intramuscularly injected guineapigs, and not until after 21 days among those intravenously injected. 32 days after the beginning of the experiment all the intramuscularly injected guineapigs showed unmistakable signs of paralysis, whereas only 33% of those injected intravenously did so.

It thus appears that the intravenous injection of antiserum very considerably reduces the chance of the development of postdiphtheritic paralysis. These paralyses also appear later than they do after the intramuscular injection of antiserum.

Experiment IV

It is well known that the heart is the organ that suffers most from the harmful effect of diphtheria toxin. Therefore in our efforts to design a well-planned experiment we paid full attention to the heart.

For this purpose two groups of 184 guineapigs were subcutaneously injected with 5 M.L.D. of diphtheria toxin. After

TABLE V—INCIDENCE OF PARALYSIS AMONG GUINEAPIGS INTRAVENOUSLY OR INTRAMUSCULARLY INJECTED WITH DIPHTHERIA ANTITOXIN, DIPHTHERIA TOXIN HAVING BEEN INJECTED $6^1/_2$ HOURS PREVIOUSLY. NUMBER OF ANIMALS IN EACH GROUP 46 (INJECTED INTRAVENOUSLY) AND 48 (INJECTED INTRAMUSCULARLY); TOXIN DOSE 5 M.L.D.; ANTITOXIN DOSE 1 A.U.

Days elapsed after injection of serum	Intravenous injection		Intramuscular injection	
	No. of animals with paralysis	Percentage of total number of guineapigs	No. of animals with paralysis	Percentage of total number of guineapigs
15	0	0	0	0
16	0	0	1	2·0
17	0	0	2	4·0
18	0	0	6	12·5
19	0	0	11	23·0
20	0	0	19	39·6
21	3	6·5	24	50·0
22	4	8·8	26	56·2
23	8	17·4	29	60·4
24	10	21·7	31	64·6
25	11	24·0	35	73·0
26	11	24·0	39	81·3
27	11	24·0	39	81·3
28	12	26·1	43	89·6
29	12	26·1	44	91·7
30	14	30·5	47	98·0
31	14	30·5	47	98·0
32	15	32·6	48	100·0

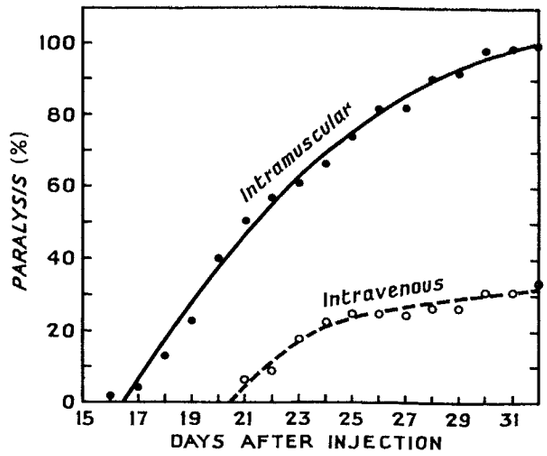

Fig. 5—Incidence of paralysis among guineapigs after intramuscular and intravenous injection of antidiphtheritic serum.

$6^1/_2$ hours they all received 1 A.U. of antitoxin, the first group intravenously and the second intramuscularly. After 1, 3, 6, 9, 12, and 15 days about a sixth of the guineapigs in each group were killed, and their hearts were removed and fixed in 20% aqueous solution of formalin. The guineapigs which had died spontaneously at the times mentioned were regarded as "killed", and their hearts were treated in the same way. Frozen sections were made of roughly the same part of the wall of the left ventricle of each heart. For the study of the myocardial lesions these sections were stained with hæmatoxylin-eosin. For the demonstration of fatty degeneration separate sections were stained with sudan III.

To provide a standard of comparison for the microscopically observed histological changes in the hearts of the treated guineapigs we killed 4 normal non-treated guineapigs per group of killed animals and spontaneous deaths. The hearts of these animals were prepared in the same way.

This pathological study was organised in such a way that the member of our team who performed the dissections and the microscopic examination (H. H. V.) never knew in advance to which category (injected or non-injected) the various guineapigs belonged. Thus an objective appreciation of the material was ensured.

As regards myocarditis, the changes observed ranged from very slight local infiltration with mononuclear cells to strong, practically diffuse, infiltration with many mononuclear cells, sometimes accompanied by leucocytes, lymphocytes, or fibroblasts.

As regards fatty degeneration of the heart, the mild cases showed only a few fat-globules in a few muscle-fibres. In severely degenerated hearts practically all the fibres contained numerous, sometimes fairly large, drops of fat.

In this experiment also some control guineapigs were injected with toxin only and not with serum. All died within 2 days, their hearts always showing considerable fatty degeneration.

The gravity of the lesions microscopically observed in the hearts of the different guineapigs varied greatly and was (more or less quantitatively) classified as ±, +, ++ or +++.

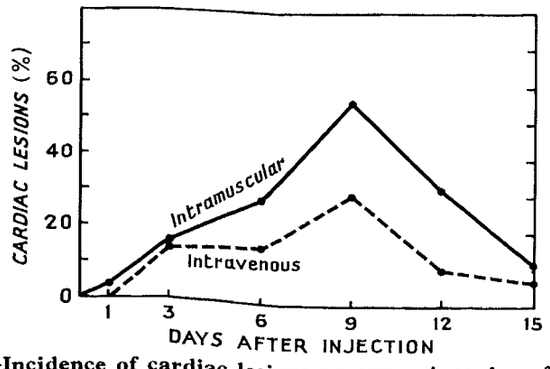

Fig. 6—Incidence of cardiac lesions among guineapigs after intramuscular and intravenous injection of antidiphtheritic serum.

Occasionally the non-injected controls (neither toxin nor antiserum) also showed slight lesions of the myocardium, classifiable as ± or +. This was the case in 15 of the 24 normal non-treated controls. Fatty degeneration of the heart was not found in any of the non-treated controls.

Some of the treated guineapigs also showed such slight lesions. It seemed only logical to leave these slight lesions, also found in non-treated controls, out of account. Consequently table VI includes only the grave lesions (myocarditis and fatty degeneration taken together). The data are also shown in fig. 6.

More or less severe lesions of the heart (myocarditis and fatty degeneration) were found in both groups. This is understandable because all the guineapigs had been exposed to the injurious action of 5 M.L.D. of diphtheria toxin for $6^1/_2$ hours before they were injected with antitoxin.

Under the conditions of the experiment these lesions reached their climax after 9 days, recovery then setting in at the same time in both groups. Of course this last circumstance may have been due to chance. It is not impossible that, if the experiment were arranged differently, the times of recovery would not coincide.

Finally we tried to divide the observed lesions into myocarditis and fatty degeneration.

The numerical data and graphs relating to this part of the investigation are not shown here, owing to shortage of space. It will suffice to state that the guineapigs injected intramuscularly with antidiphtheritic serum showed more cases of

TABLE VI—DISTRIBUTION OF SERIOUS LESIONS OF THE HEART FOUND AMONG THE VARIOUS GROUPS OF GUINEAPIGS GIVEN 5 M.L.D. OF TOXIN SUBCUTANEOUSLY, FOLLOWED BY INJECTION OF 1 A.U. OF ANTITOXIN INTRAMUSCULARLY OR INTRAVENOUSLY AFTER $6^1/_2$ HOURS

No. of days between serum injection and killing	Intravenously injected		Intramuscularly injected	
	No. killed	No. of heart lesions	No. killed	No. of heart lesions
1	30	0	30	1 (3·3%)
3	35	5 (14·3%)	38	6 (15·9%)
6	32	5 (15·6%)	31	9 (29·0%)
9	29	9 (31·0%)	30	17 (56·6%)
12	29	3 (10·3%)	30	10 (33·3%)
15	29	2 (7·0%)	25	3 (12·0%)

myocarditis and fatty degeneration of the heart than did the intravenously injected ones.

As regards the observed lesions of the heart, there again emerges a clear difference between the results of the two methods, which is strongly in favour of the intravenous injection of antiserum. This is easily explained. After the injection of intravenous antitoxin the serum of the guineapig is practically immediately "up to titre"—i.e., the total amount of antitoxin is available immediately for neutralisation of the toxin still circulating freely at that moment. This toxin can then no longer cause a lesion of the heart. Possibly part of the toxin which has already penetrated into the myocardium is also neutralised.

The effect of the intramuscular injection of antiserum is the same, but slower, because of the comparatively slow absorption of the antitoxin thus injected. The toxin still circulating freely is rendered inactive more slowly or can act longer on the myocardium, with all the consequences of this.

The difference between the antitoxin titres in the guineapigs injected intravenously and intramuscularly with antiserum will, of course, be greatest in the first few hours. This is undoubtedly responsible for the large difference in the incidence of cardiac damage.

Summary of Results of Experiments

Groups of guineapigs were subcutaneously injected, 2 at a time, with a constant dose of diphtheria toxin (5 M.L.D.). After a certain number of hours they all received a constant dose of antitoxin (1 A.U.), half of them being injected intravenously and the other half intramuscularly. The intravenous injection of antiserum

had a considerably greater therapeutic effect than had the intramuscular. The guineapigs injected intravenously with serum showed less loss of weight and more rapid recovery of weight, fewer deaths, less frequent and later development of paralysis, and fewer lesions of the heart (myocarditis and fatty degeneration) than did guineapigs injected intramuscularly.

Discussion

The experiments were made under strictly standardised conditions. The doses of toxin and of antitoxin injected were the same for all the guineapigs, which were all of the same sex and had the same weight in each pair. The time between the injection of toxin and that of antitoxin was the same for each pair. In this way we found a quantitative difference between the therapeutic effects of the intravenous and the intramuscular injection of antiserum. Had the criteria of these experiments (amount of toxin, amount of antitoxin, and interval between the two injections) been different, we would no doubt have seen the same qualitative differences, but the absolute figures would not have been the same.

Obviously the quantitative results obtained with this " animal model " cannot be quantitatively transferred to the therapy of human diphtheria patients. The criteria for the latter are different from those applied in the experiment on guineapigs.

If, however, the pathogenesis of human diphtheria is determined by the formation of toxin by the diphtheria bacillus infecting the patient and by the effects of this toxin, especially on the heart and the nervous system, we may indeed apply the qualitative results of these laboratory experiments to man.

Summarising the results of experiments I–III we may conclude that, for the treatment of human diphtheria also, the intravenous injection of diphtheria antiserum is much to be preferred to the intramuscular route for the following reasons:

(1) After the intravenous injection of antiserum the patient's serum is almost immediately " up to titre "—in other words, the total amount of diphtheria antitoxin is transported very quickly through the blood to the organs concerned. Intramuscularly injected antiserum is absorbed only slowly; on the average this absorption is not completed until after 4 days, but it may take as much as 6 days. The serum titres of the patients were determined 24 hours at the earliest after the injection of antitoxin; hence we cannot give any figures for the serum-antitoxin levels within a shorter time after the intramuscular or the intravenous injection. It can, however, safely be assumed that the relative and absolute differences will be large in the first few hours after the injection.

(2) As a non-specific protein, the intravenously injected antitoxin is not more quickly broken down and excreted than intramuscularly injected antitoxin.

(3) Very shortly after the intravenous injection of antiserum some of the antitoxin is excreted into the saliva through the salivary glands. From this moment the saliva is atoxic; henceforth no toxin can be absorbed from this saliva through the inflamed and damaged tonsils and wall of the pharynx. After the intramuscular injection of antiserum it usually takes many hours, sometimes even days, for this excretion of antitoxin through the salivary glands to get going. During this interval the saliva contains toxin which can be absorbed through the inflamed tonsils and wall of the pharynx.

(4) Lesions of the heart (myocarditis and fatty degeneration) will very probably be less common among intravenously injected than among intramuscularly injected patients. These lesions will presumably also be less serious among the former than among the latter.

(5) As a group, intravenously injected patients will very probably show fewer cases of paralysis than will intramuscularly injected patients.

(6) Under comparable conditions the mortality among intravenously injected patients is likely to be lower than among intramuscularly injected patients.

There is no danger attached to the intravenous injection of the antiserum, provided that certain precautions are taken. The skin test for hypersensitivity to the antiserum is especially important and should not be omitted.

For this test 0·2 ml. of the antiserum, diluted to a tenth with saline solution, should be injected intracutaneously on the flexor aspect of the forearm. It is absolutely imperative that a weal should form during this injection; if it does not, the intracutaneous injection must be considered unsuccessful and will have to be repeated.

If no redness or swelling shows within an hour after the intracutaneous injection, the intravenous injection can safely be given. Our experience with many hundreds of patients is that there are then never any serious reactions. Some hours after the intravenous injection of serum (enzymatically purified and radically despeciated horse-serum) a few of the patients had slight pyrexia, sometimes accompanied by a rigor. This type of reaction proved harmless.

If, however, the skin reaction is positive (redness and/or swelling), the serum must be injected intramuscularly and in very small portions at intervals of half an hour or an hour, under the supervision of an experienced person who, if necessary, can administer adrenaline (0·1–1·0 ml. of adrenaline hydrochloride 1 : 1000, according to the patient's age) if the patient shows obvious allergic phenomena.

Parish et al. (1957) propose a hypersensitivity test in which 0·2 ml. of undiluted serum is injected subcutaneously. If there is no general reaction within 30 minutes, the whole dose of serum can be injected either subcutaneously or intramuscularly without special precautions. This refers to the injection of a prophylactic dose of antitetanus serum (generally 1500 A.U. in 1 ml.).

We do not recommend this procedure for diphtheria patients. We believe that the risk of serious allergic reactions is much greater after the subcutaneous injection of 0·2 ml. of undiluted antidiphtheritic serum (4000 A.U. per ml., the protein content generally being greater than that of prophylactic antitetanic serum) than after the test injection described by us.

The doses of antidiphtheritic serum which we have given in the last few years have been 10,000–20,000 A.U. per patient according to age. The results of the determination of antitoxin titres after the intravenous injection have convinced us that an additional intramuscular injection of antidiphtheritic serum would be superfluous.

Provided that the precautions described above are observed, there is not the slightest objection to the intravenous injection of antiserum in all cases where an injection may be required. Consequently all these patients should be injected not intramuscularly but intravenously with diphtheria antitoxin. The intravenous injection of antiserum in patients suspected of diphtheria ought therefore to become the rule and should not be reserved for " serious " or toxic cases, as is still usually done at present.

Moreover it is by no means always possible to predict with certainty the course of the diphtheria. During an epidemic there have repeatedly been cases which initially seemed mild but became toxic within a few hours and rapidly fatal. Some of these patients would probably have been saved if they had been given an intravenous injection of antidiphtheritic serum at the earliest opportunity.

It should be borne in mind that, in cases of suspected diphtheria, the first object of the antiserum therapy is the quickest and completest possible elimination of any further absorption of diphtheria toxin (e.g., from the saliva) and the binding of the toxin still circulating. The investigations here described will have shown that not only the promptness of the injection but also its route can have a decisive effect. Not until this has been done will the treatment be directed to combating the damage already done to the heart, kidneys, and nervous system.

Summary

An investigation is described which clearly brought out the advantages of the intravenous injection of antitoxic serum in patients suspected of diphtheria, compared with the intramuscular injection still commonly used.

The intravenously injected antitoxin immediately brings the patient's serum "up to titre", whereas absorption of the intramuscularly injected antitoxin is slow, usually not being complete until after about 4 days, and sometimes 6 days.

Intravenously injected antitoxin is not excreted more rapidly than intramuscularly injected antitoxin.

Very shortly after the intravenous injection of antitoxin a small portion of it is excreted through the salivary glands into the saliva, which it renders atoxic. Consequently from this moment no more toxin will be absorbed from the saliva through the inflamed and damaged tonsils and wall of the pharynx.

The results of experiments on guineapigs indicate that patients intravenously injected with antidiphtheritic serum may be expected to show fewer lesions of the heart (myocarditis and fatty degeneration), fewer post-diphtheritic paralyses, and a lower mortality than patients intramuscularly injected with antiserum.

With due observance of the necessary precautions—e.g., a skin test for hypersensitivity (which must be negative)—diphtheria antitoxin should be injected not intramuscularly but intravenously whenever such an injection is required, even in mild cases of diphtheria.

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"The only advantage that other animals can be supposed to have over man is, that being excluded by their nature from all mental enjoyments, they are also secured from all the pains and disquietudes that proceed from the same source; but to acquire an exemption from disquietude at the expense of being equally exempted from all the delicate feelings of the mind and affections of the heart, is a purchase which I hope no honest mind will ever be willing to make."—JOHN MOORE. *Medical Sketches*; pp. 223-4. London, 1786.

EPIDEMIOLOGY OF STILLBIRTHS AND INFANT DEATHS DUE TO CONGENITAL MALFORMATION

W. J. R. ANDERSON
M.B. Aberd., M.R.C.O.G.
LATELY REGISTRAR, ABERDEEN MATERNITY HOSPITAL

D. BAIRD
M.D., B.Sc. Glasg., F.R.C.O.G., D.P.H.
REGIUS PROFESSOR OF MIDWIFERY IN THE UNIVERSITY OF ABERDEEN

A. M. THOMSON
M.B., B.Sc. Glasg., D.P.H.
HONORARY DEPUTY DIRECTOR, OBSTETRIC MEDICINE RESEARCH UNIT (MEDICAL RESEARCH COUNCIL), UNIVERSITY OF ABERDEEN

THE cause of most fœtal malformations is obscure. Some types, both animal and human, are inherited and appear in accordance with the mendelian laws. The environment of the early embryo may be responsible for other types or may bring out latent tendencies; this has been proved experimentally in primitive animals and some mammals through changes of temperature and pH, mechanical injury, anoxia, dietetic deficiencies, and exposure to X-rays and chemical substances.

It is not known whether these factors cause malformations in man, but indirect evidence suggests that fœtal defects may sometimes be brought about by noxious agents or by an inadequate intrauterine environment. Abnormal implantation of the ovum, as in tubal pregnancy, is associated with a high incidence of malformations (Mall 1908). The incidence is increased if bleeding has occurred in the early months of pregnancy (Burge 1951, Turnbull and Walker 1956) and in diabetic pregnancies (Peel and Oakley 1949). Gregg's (1941) observation is now generally accepted, that infection with rubella in early pregnancy is often followed by the birth of a deformed child.

The incidence of congenital malformations is difficult to estimate. The best study based on a defined population is that of Record and McKeown (1949) dealing with Birmingham. Several reports based on hospital records are available; but, though the data are no doubt accurate, hospital patients are not necessarily representative of the population from which they are drawn. National statistics refer only to deaths certified as due to deformity, and are by no means comprehensive. Scottish vital statistics give much information about malformed stillbirths but very little about neonatal or infant deaths. Those for England and Wales give no information about the causes of stillbirths and little about infant deaths due to malformations.

This study deals with stillbirths and infant deaths associated with malformations. The main group used for analysis consists of 37,585 single births to booked patients delivered in the Aberdeen Maternity Hospital in 1938–55. Lest the trends found were produced by selection of patients for hospital confinement, these were compared with those in all Aberdeen births in 1949–55. (Records for 1938–48 were not available on a city-wide basis.) These local statistics are compared with certain national data derived from the annual reports of the Registrar-General for Scotland.

Method

Group 1.—The records of the Aberdeen Maternity Hospital for 1938–55 were searched for fœtal deformity in booked cases. The present analysis has been restricted to cases of stillbirth or of infant death. The records of infant deaths taking place some time after mother and child were discharged from hos-