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THE PREPARATION OF NONPYROGENIC INFUSION AND
OTHER INTRAVENOUS FLUIDS BY ADSORPTIVE FILTRATION*

REPORT OF 42 MONTHS' TRIAL

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NEW YORK, N. Y.

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PART I—BIOLOGIC ASPECTS

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PART I—BIOLOGIC ASPECTS

IN 1937, there was reported from this laboratory a method of preparing intravenous fluids based on a new principle.^{1, 2} As pointed out in that report, it was Siebert's^{3, 4} extension of Hort and Penfold's⁵ work which laid the foundation for the various methods of preparing intravenous fluids at present practiced in commercial houses and hospitals. Even the method described by Rademaker^{6, 7} and the more recent and mechanically ingenious elaboration by Walter,^{8, 9, 10} stem from Siebert's original principle. The principle, however, suffers from a critical weakness in the assumption that if the water used in the manufacture of the fluids is pyrogen-free, then the resulting mixture must also be pyrogen-free. But should the water become contaminated, say as a result of failure to clean the still, or should pyrogen be present in the chemicals used in making the solutions, the lack of provision for its removal is a notable disadvantage of the method. It is, in fact, the lack of security behind the old working principle which impels manufacturers to test their intravenous fluids on the rabbit. Unfortunately, not even this precaution has entirely eliminated the occurrence of pyrogenic reactions. As pointed out in a previous communication,¹¹ this failure is perhaps due to the inadequacy of the test doses in use.

That chemicals occasionally are contaminated with pyrogen has not heretofore entered into consideration. In previous papers¹² we have reported on the pyrogenicity of samples of inulin as they have come fresh from the manufacturer.

Table I shows the effect on test animals of samples of sodium chloride, of dextrose, and of sodium citrate, all "commercially" and "technically pure," which had been made into solutions with nonpyrogenic water and tested immediately after the containers had been opened. The sodium chloride sample was the more reactive of two found pyrogenic in a test of 38 samples, an incidence of slightly more than 7 per cent. The dextrose was the most reactive of four positive samples of 32 tested, an incidence of 14 per cent, and the sodium citrate was the most reactive of four of 18 tested samples, an incidence of over 22 per cent. It may also be mentioned that sodium citrate

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is usually more heavily contaminated than dextrose which, in turn, is usually more heavily contaminated than sodium chloride. Because of the fact that test animals injected with such large doses of sodium citrate are liable to die in cardiac arrest, perhaps as a result of the blood calcium being thrown out of ionization, it is necessary to add an equivalent dose of calcium chloride to the sodium citrate test solution before it is injected.

TABLE I
EFFECT OF VARIOUS COMMERCIAL SOLUTIONS

Expt. No.	Chemicals	Test Dose Gm. Vol. Cc.	Animal Used	Weight Kg.	Temp. Rise Deg. F.	Leukocyte Count Change x 1000
1	Sodium chloride....	.4 50	Rabbit	3	1.2	
2	Sodium chloride ...	12 1500	Dog	15	2.0	-5.7
3	Dextrose.....	7.5 150	Rabbit	3	2.0	
4	Dextrose.....	75 1500	Dog	15	2.4	-9.4
5	Sodium citrate*....	0.28 11.5	Rabbit	3	2.2	
6	Sodium citrate*....	2.8 115	Dog	15	2.8	-8.2

*These dosages were calculated on the bases that the maximum dose of sodium citrate given to a 60-Kg. animal is 5.6 Gm. (3 liters of blood), and that the rabbit is approximately three times and the dog six times less sensitive to pyrogen than man. An equivalent amount of nonpyrogenic calcium chloride was added to each sodium citrate solution.

The chemicals in these cases were perhaps contaminated by pyrogen in the process of manufacture. Nonpyrogenic chemicals even in the "dry" state can, however, become pyrogenic on standing under unsterile conditions, as has been found by Smith¹³ to be the case with inulin, and by us with dextrose, sodium citrate and heparin. Apparently the better medium the chemical is for bacterial growth, the more easily it becomes pyrogenic. Humidity and a warm environmental temperature seem to accelerate pyrogen production, probably by acceleration of bacterial growth. Among aerial contaminants, Co Tui and Schriit^{14,9} have found *B. proteus vulgaris* and *B. subtilis* fairly prolific pyrogen producers. In the light of these findings, therefore, the assumption that to guard against the occurrence of pyrogenic reactions it is necessary only to keep the water pyrogen-free can be costly as well as unsafe. The problem will perhaps become increasingly acute with the increasing use of blood and plasma transfusions.

The adsorptive filtration method of preparing intravenous fluids differs from the older method in that it is based on the principle of filtering the resulting mixture of chemicals and distilled water through specially prepared compressed asbestos pads proved to remove pyrogen. This method is efficacious not only for infusion fluids but for all crystalloidal intravenous medications. With colloidal substances such as sera and gum acacia, however, it is ineffective. The method has been in experimental use on the New York University Surgical Division of the Bellevue Hospital continuously since July, 1937 except for an intermission of 16 months. It is the purpose of the present communication to summarize the results of the experiment as well as to describe the practical method of manufacture which has been evolved.

The practical difficulties encountered in the course of this work may be briefly discussed since they are difficulties which many hospitals and similar institutions which undertake the manufacture of their own infusion fluids will have to face.

1. The first problem was the establishment of the criterion of pyrogenicity. With fluids which are frankly pyrogenic, this is an easy matter since the symptom-complex of the pyrogenic reaction is a familiar one. The difficulty was with fluids containing a borderline dose of pyrogen, which would elicit only a slight temperature rise easily missed in the four-hour routine ward temperature readings but might, when administered in doses two or three times larger than the usual one, give rise to a fairly severe reaction.

It was, therefore, necessary during the first six months of this experimental period to subject the fluids made to animal tests, using the rabbit and the dog, according to the technic described elsewhere.¹¹ If the fluids were found nonpyrogenic according to the tests, they were then released for ward use. The temperature readings of all patients receiving the fluid were taken hourly for four hours, both during and after the administration of the fluid. In this way transient temperature rises could be detected.

2. The second problem was the elimination of contamination from external sources, such as the rubber tubings, glass connections and needles used in the dispensing set. Originally, the latter were sterilized in the ward instrument sterilizers in common with contaminated pus basins and surgical instruments. The occurrence of four reactions on patients in the first three weeks of this period led to tests of the water from the sterilizer. Ten and 15 cc. samples of this water gave unmistakable reactions in both rabbits and dogs. This result led to the institution of a method of washing, packing and sterilizing the dispensing sets which will be described in a later section of this paper.

3. A third difficulty was the presence of a yellowish-brown coloration in the solutions after they had been autoclaved. This was first thought to be due to caramelization but has been attributed by Krno¹⁵ to the production of levulinic acid.¹⁰ In our preparation two factors were found responsible for the coloration: (a) The asbestos pads originally used were made for the filtration of fluids which did not have to be autoclaved. They imparted some principle to the filtrate which caused the formation of this coloration on heating. The adoption of a pad specially prepared for infusion fluid use has eliminated this factor; and (b) the second factor was overheating of the solution. This occurred when, after sterilization, the autoclave pressure was permitted to sink so fast that the fluid inside the flasks began to boil over. The film of fluid on the inner wall of the neck of the flasks evaporated and the residue became browned, imparting the coloration to the rest of the solution. The remedy to this was obviously to lower the autoclave pressure so slowly that no boiling over took place.

4. The fourth and most serious difficulty was the presence of shreds and particles in the filtrate. While the injection of solutions containing these particles into numerous experimental animals caused no detectable symptoms,

their presence was none the less objectionable. It was because of this difficulty that the use of these fluids was suspended for 16 months in 1938 and the first part of 1939, until a manufacturer could be found who would cooperate in devising means of eliminating these particles. Jena filters, Chamberland and Berkefeld filters were interposed at the outlet of the asbestos filter without avail. Finally, the difficulty was overcome by both improving the quality of the filter pad and interposing an aloxite fiber eliminator between the asbestos filter and the collecting bottle.

5. Standardization of filter pads: The compressed asbestos pads, as now prepared for our use by the Ertel Engineering Corp., are calibrated with regard to two properties: (a) The speed of filtration; and (b) degree of pyrogen retentiveness.

Speed of Filtration.—This is controlled by three factors: The filtering pressure applied; the filtering area; and the composition of the pad. The filtering pressure is safest at 12 lbs., and the filtering area is limited by the size of filter practicable in hospitals. Our present filter takes pads of 12 inches, with an actual filtering area of 29.5 cm. in diameter. The pads are made of asbestos and pulp. The more asbestos used, the greater the pyrogen retentiveness, but the slower the filtration rate. The interplay of these two factors, therefore, determines not only the rate of filtration but also the efficiency of the pads to remove pyrogen. The pad now in use has a delivery volume of two liters every 2.5 minutes. A six-hour filtration would, thus, yield 288 liters, which would be more than enough to meet the daily requirements of a fairly large hospital. Hospitals using larger amounts of fluid may enlarge the filtering area of the filter by the method to be described later or install two such filters.

Pyrogen Retentiveness of the Filter Pads.—The ability of these pads to remove pyrogen was shown in a previous work¹⁶ to be due to adsorption and not to sieving. This means that the property can be exhausted. It is, therefore, necessary so to standardize the pads that they are not only uniform but also possess such a margin of safety in pyrogen retentiveness that they remove the maximum amount of pyrogen which can occur in the distilled waters and the chemicals in use.

In order to calibrate the pads in this respect it was necessary to secure a sample of pyrogen and use it as a standard. The pyrogen used in this work was isolated from the cell-free Berkefeld filtrate of the supernatant portion of concentrated stock suspension of typhoid vaccine (1 cc. = 88 billion cells). From seven liters of this suspension, four grams of the substance was yielded. The method of isolation as well as the chemical and biologic aspects of this substance will be reported elsewhere. It is necessary, however, to state that the substance is still in the crude stage, containing six per cent nitrogen, which on further purification has been reduced to 1.6 per cent. The biologic (pyrogenic) potency of this crude sample, called "pyrogen-A," has been determined on rabbits, dogs, and men. Table II shows the effect of different doses of this preparation. If the minimum effective dose (MED) is arbi-

trarily defined as that dose, per kilo body-weight, which on intravenous injection will cause a temperature rise of 0.8° to 1.0° F., the MED in rabbits is 0.1 gamma; in dogs, 0.4 gamma; and 0.03 gamma in men. The reason for using this crude substance as a standard instead of the more purified substance is that the process of purification is so tedious that it may be a long time before enough of the purer substance can be available for this purpose. In any case, the biologic calibration of "pyrogen-A" makes it unnecessary to wait for the purer preparation.

TABLE II
EFFECT OF DOSES OF PYROGEN-A IN RABBITS, DOGS, AND MEN

Subject	Weight Kg.	Dose per Kg.	Temp. Change Deg. F.	Leukocyte Count Change x 1000
Rabbit 1.....	3.9	0.12	0.8	
Rabbit 2.....	2.5	0.12	1.0	
Rabbit 3.....	3.0	0.12	0.9	
Rabbit 4.....	3.0	2.0	3.5	
Dog 1.....	15.0	0.3	1.0	-6.1
Dog 2.....	16.0	0.3	1.1	-5.5
Dog 3.....	15.5	0.3	0.8	
Dog 4.....	16.0	2.0	3.2	-10.2
Man 1.....	52.0	0.04	1.2	
Man 2.....	64.0	0.04	0.9	
Man 3.....	72.0	0.04	1.1	
Man 4.....	70.5	2.0	5.0	

Process of Calibrating the Pads.—One milligram of "pyrogen-A" (1000 gammas) is weighed out on a sensitive analytic balance and dissolved in 100 cc. of pyrogen-free distilled water. Ten cubic centimeters of this solution contains 10 gammas of pyrogen. Amounts of from five cubic centimeters up, in multiples of five, are filtered through test pads of a convenient size mounted in convenient filters. The filtrates are then tested in both rabbits and dogs. Table III shows the protocols of some of these tests.

TABLE III
CALIBRATION OF FILTER PADS

Experimental Animal	Weight Kg.	Volume Pre-filtrate Cc.	Total Dose Pyrogen in Pre-filtrate Gamma	Temp. Change Deg. F.	Leukocyte Count Change x 1000
Rabbit 1.....	2.5	30	300	- .2	
Dog 1.....	16.0	30	300	+ .1	- .2
Rabbit 2.....	3.0	35	350	+ .2	
Dog 2.....	15.0	35	350	.25	- .3
Rabbit 3.....	3.0	40	400	+ .15	
Dog 3.....	15.5	40	400	+ .1	+ .1
Rabbit 4.....	2.5	45	450	+ .4	
Dog 4.....	16.5	45	450	+ .2	- .3
Rabbit 5.....	3.0	50	500	+ .4	
Dog 5.....	15.5	50	500	+ .8	-1.2

It will be seen from Table III that the pad removes nearly 500 gammas of "pyrogen-A." Since the pad is 2.5 cm. in diameter, it has a surface of 4.9 sq. cm., and, therefore, each square centimeter removes over 100 gammas of "pyrogen-A." The filtering surface of the four large filter pads used in the hospital filter assembly are each 29.5 cm. in diameter, making a total filtering area of 2734 sq. cm. The amount of pyrogen removable by the entire filter

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assembly is approximately 273,400 *gammas*. Since about 140 *gammas* are required to cause a temperature rise of 5.6° F. in an average man of 70 kilos, this filtering area will remove about 1953 such doses.

When applied to solutions of sodium chloride or dextrose in distilled water this range of safety is more than ample as the amount of pyrogen contained in any combination of these three substances in the usual dose is tenuous and, further, the filter-pad assembly is made to deliver not more than 288 liters with each filtration. The case of sodium citrate requires special consideration. As shown in Table I, gram for gram, the contamination of pyrogen in sodium citrate is heavier. This, however, is more than offset by the fact that the dose of sodium citrate used per 500 cc. of blood is usually in the neighborhood of 1875 mg. in 75 cc. of water. This, by weight, is only about one-fifth of the dose of sodium chloride in one liter of physiologic salt solution and less than one-twenty-seventh of the dose of one liter of 5 per cent dextrose solution. Therefore, in making sodium citrate solutions, it is only necessary to use two filter pads and restrict the manufacture to not more than 300 doses.

These calculations are admittedly based on biologic effects, and assume that pyrogen from sources other than the typhoid bacilli have the same potency per unit-weight as typhoid pyrogen. This assumption has no direct experimental basis at present. The only justification for applying the figures derived from "pyrogen-A" to the pyrogenic substances found in these fluids, whose exact source is unknown, is that they have so far held true in the samples whose pyrogenic content was roughly determined by the biologic test, then filtered through asbestos pads of the required area and then retested and found negative for pyrogen. In the absence of more exact knowledge, this approximate correspondence for the present can be used as a rough working principle. This method of pretesting before filtration may be used for any intravenous material whose maximum pyrogenic content has not been as thoroughly studied as have those of dextrose, sodium chloride, and sodium citrate.

RESULTS OF 42 MONTHS' TRIAL

During this experimental period, 3867 liters of intravenous fluids prepared by the nonpyrogenic method have been administered. They may be tabulated as follows:

No. of Administrations	Amount Given Liters	Total Given Liters
1815	1	1815
652	2	1304
123	3	492
64	4	256
Totals 2654		3867 liters

In none of these was there a reaction. One patient who complained of a chilly sensation after receiving 1000 cc. did not have a temperature rise. It

is clear that if any pyrogen was present in the solutions, it was in such minute amounts as to be below the MED even in three to four liters.

PART II—MECHANICAL ASPECTS

The parts which comprise the entire filter assembly may now be enumerated and the more important items described. The filtering portion of the assembly is diagrammatically represented in Figure 1. It consists of a compressed nitrogen or air tank, A, which furnishes filtration pressure; the reservoir, B, which holds the prefiltered mixture; the filter itself, C; the aloxite fiber eliminator, D; and the collecting flask, E.

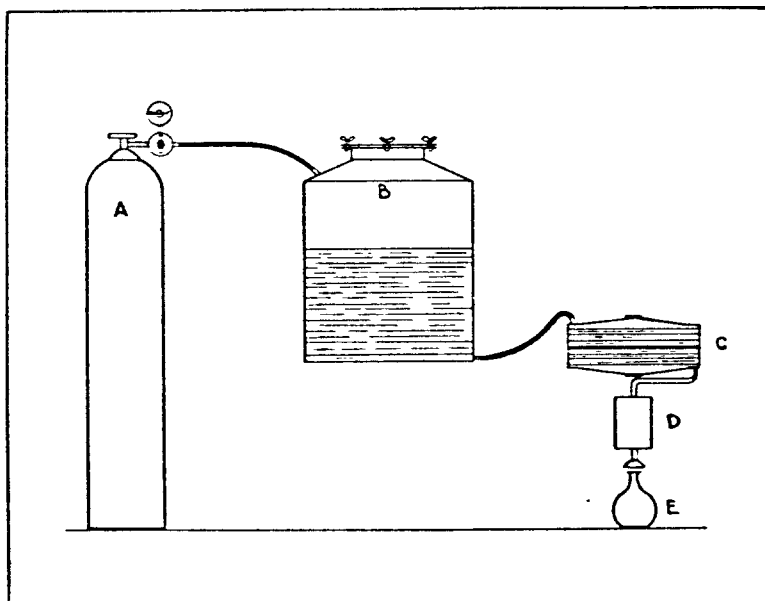


FIG. 1.

The reservoir, usually of 250 liter capacity, is made of stainless steel, and may be equipped with an electric stirrer.

Figure 2 illustrates the essential parts of the filter as well as the path taken by the solution passing through it. It may be seen that each filter pad is held between two perforated Monel metal plates or frames, the three forming a pad-frame unit. Two such units form a compartment. The fluid flows from the solution inlet, A, into a conduit system, B, which splits it into two streams. One stream filters through one pad downward and the other upward to meet in the collecting chamber, C, situated between the two pad-frames. From C the solution is conducted into the second compartment to have the process repeated, after which the conjoint stream from the second collecting chamber, D, passes through the fiber eliminator into the collecting flask. Both the latter are to be described in a subsequent section.

Since provision is made in the present filter to accommodate two additional compartments, the filter has a still wider range of usefulness than has been calculated. Any one of the three factors—capacity, speed of filtration, or retentiveness—can be increased two-fold by this doubling of the filtration area. The stream in this case would be split in four instead of two as in the filter illustrated in Figure 2.

Figure 3 is a photograph of the filter ensemble. A is the spindle with which the pads are loaded and unloaded and the filtering system rendered watertight. B is the filtering system, C is the pressure gauge, D the housing for the aloxite fiber eliminator, and E the outlet. The self-autoclav-

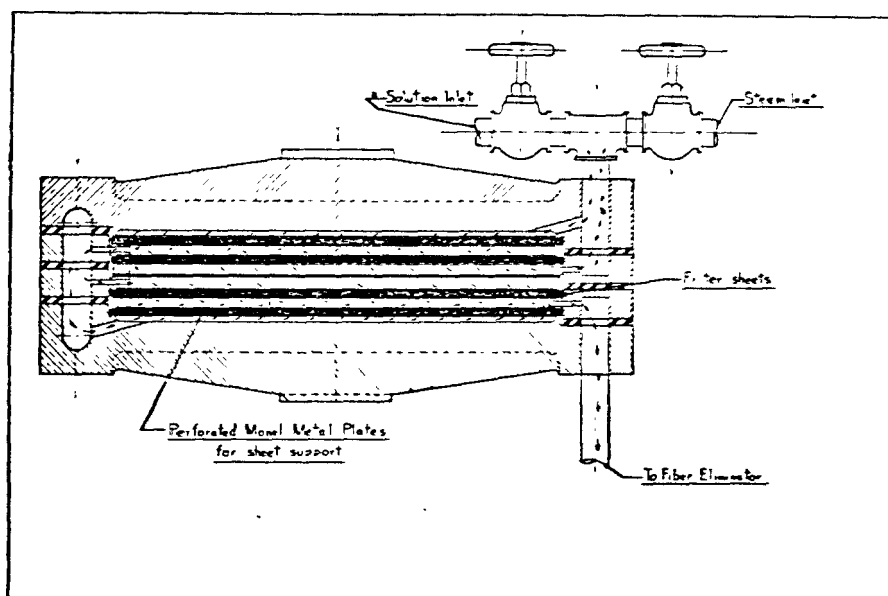


FIG. 2.

ing system, F, consists of a steam generator in which the source of steam used is pyrogen-free distilled water. It may be heated electrically, by gas, by house-steam, or by an oil burner. The steam generated is led into the interior of the filter by the flexible hose, G. The course of the steam through the filter is identical with that of the fluid which follows it and whose course has been described in the foregoing paragraph.

The aloxite fiber eliminator is a thick, finely porous filter made of carborundum. Because of its composition it is sturdy and may be heated to a temperature high enough to destroy any pyrogen which may have developed in the interstices of the stone.

The dispensing part of this system, mainly the collecting and storing flasks and accessories, is essentially the same as that described by Walter.^{8, 9, 10} It is the most satisfactory one available at present, made to withstand hard

usage, and in three years of use has suffered the minimum amount of breakage. It has, however, the slight inconvenience of having too many parts.

Figure 4 illustrates the part of this system which consists of: A, pyrex flasks of specially rugged construction of one to two liters capacity, preferably graduated; B, a one-hole rubber bushing to fit the flask; C, a special stainless steel stopper with a partially grooved stem; D, a two-way vent tube for the administration of the fluids and for the admission of air into the flasks to displace the fluids.

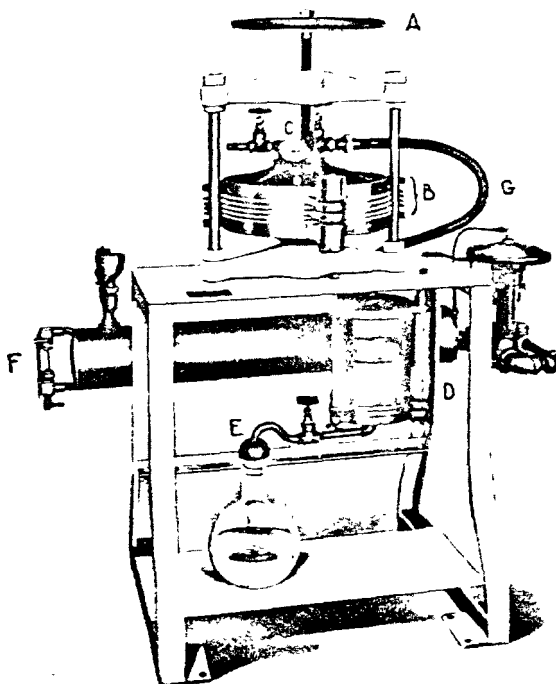


FIG. 3.

The bushing, B, is of special rubber which can stand 500 sterilizations without shrinking. It is provided with a hole in the center for the insertion of the two-way vent tube when fluid is being administered, and for the insertion of the steel stopper when the flask is not in use. The stopper is provided with a partially grooved stem which is fitted partly into the hole of the bushing while sterilization takes place. The groove allows equalization of inside pressure with outside pressure. When sterilization is completed, the stem of the stopper is pushed all the way in, as in E, the umbrella of the stopper serving as an hermetic seal for the rubber bushing.

The original Fenwal-tube is short, so that the flask end is submerged in the fluid when the flask is inverted for administration. There are two objec-

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tions to this feature: First, when the infusion is being given, the air which enters the flask to displace the fluid has to be bubbled through the fluid. This, in effect, means the washing of 1000 to 2000 cc. of unsterile air in the infusion fluid. Bacteriologic cultures of ten samples of fluid subjected to this kind of air-washing gave three positive cultures. By the simple device of

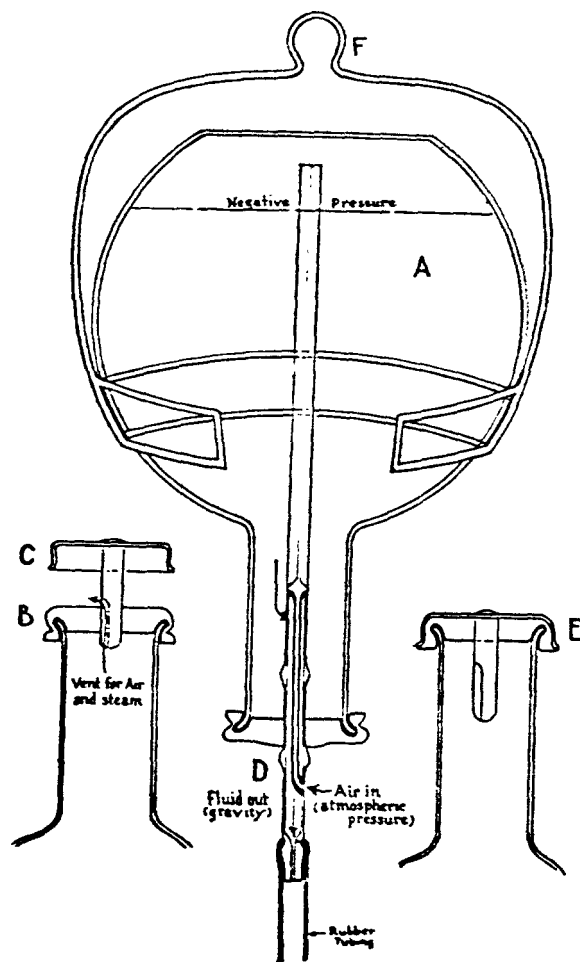


FIG. 4.

lengthening the tube so that the flask end extends above the surface of the fluid when the flask is inverted, this washing effect is avoided.

No positive cultures were found in ten fluids subjected to this system of air displacement. If it is desired to admit absolutely sterile air into the flask, the air vent may be modified in such a way as to accommodate a small sterile Berkefeld candle. This was originally used by us but later discontinued because of inconvenience.

The second objection to the short original Fenwal-vent-tube is that unless special care is taken, the fluid is as liable to flow through the air vent as through the fluid vent.

The split ring bracket, F, is simply a wire basket for suspending the flask in an inverted position when the flask is in use.

Some reference must be made to the still which delivers distilled water for making the solutions. Since the still is the heart of all other processes, much emphasis has been laid on it. While it is our opinion that the undue concentration on the still has lulled to rest suspicion of other possible sources of pyrogen, we believe that the still must be cleaned periodically so that it adds no pyrogen to the distilled water to lower the factor of safety of the filter. Moreover, it must be large enough to deliver the necessary amount of distilled water to be used for cleansing as well as to make the solutions themselves. A still that delivers 10 gallons of water per hour (40 liters), will be sufficient to meet the needs of the average hospital. The periodic cleaning also does away with incrustations which lower the output of the still.

CARE OF THE PARTS

The Reservoir: The reservoir when new must be rinsed first with soap and water and then copiously with distilled water. Subsequent care after every use consists in rinsing it out thoroughly with distilled water and then drying it.

The Filter: When new, the conduit system of the filter is rinsed thoroughly with distilled water. Thereafter, the same rinsing process is repeated after each use.

The Aloxite Fiber Eliminator: After every use, the surface of the stone fiber eliminator is simply flushed thoroughly in order to wash away the particles and shreds which may be caught on its surface. After washing, it is placed in an electric oven at a temperature of 120° C. for one hour to dry. This temperature sterilizes the stone so that pyrogen production within the interstices of the stone is halted.

Fenwal-Flasks, etc.: The initial cleaning and subsequent care of the Fenwal-flasks, rubber bushings, steel cages, and the modified vent tube have been described by Walter and need no extended treatment here. Suffice it to say, that the flasks are washed and brushed with hot soapsuds and rinsed first in tap and then in distilled water until the walls run homogeneously clear. The same treatment should be applied to the other glassware, namely, the vent tube and the drip as well as to the steel stopper. An occasional treatment of the glassware with a cleaning mixture of 1.5 per cent solution of sodium dichromate in concentrated sulfuric acid may have to be resorted to in order to get rid of spots in the glassware difficult to eliminate otherwise. The rubber bushings when new are first scrubbed with soap and water until no further powdery substance is given off, then rinsed with

PREPARATION OF INFUSION FLUIDS

distilled water. The rubber tubings when new are likewise rinsed well with hot soapsuds and then with distilled water. Immediately after each washing, the rubber tubings, the vent tubes, the drip and the needles are wrapped and packed in a convenient container. We have used for this purpose a specially constructed copper cylinder, one foot long and $2\frac{1}{2}$ inches in diameter, provided with a detachable cap as well as holes in the wall to allow penetration of the steam in the autoclave. After sterilization, the holes are closed by a special sliding device and the set stored for future use.

Preparation for Filtration: The flasks without the rubber bushings in place are sterilized in the hospital autoclave. The rubber bushings and the steel stoppers are also sterilized but separately from the flasks.

The filter with pads mounted on the frames is sterilized by its own self-autoclave for 30 minutes at 18 pounds pressure. After that it is allowed to cool to a temperature convenient for handling.

The solution is mixed in the reservoir. The reservoir is then connected, on the one hand, with the nitrogen tank and, on the other hand, with the inlet of the filter. The collecting and storing flask is placed at the outflow tube of the filter and filtration is begun by turning on the gas gradually to 12 pounds. After discarding the first four liters, the flasks are then filled one by one.

After collection, the flasks are partially stoppered with the stainless steel stopper in such a way that the groove in the stem of the stopper allows communication of the inside of the flask with the outside (Fig. 4). The flasks are then put into the autoclave and sterilized at 250° F. for 30 minutes. After sterilization, special care is taken to have the pressure in the autoclave gradually diminished so as not to cause boiling. The stem of the steel stopper may then be pushed home, in which case a partial vacuum is created as the solution cools. This partial vacuum, which helps retain the stopper in place and causes a water-hammer effect when the bottom of the flask is struck with the fist, may be used to indicate whether the stopper is leaking after storage.

After the flask is sealed, it is stored until needed.

Just before administering the fluid, the steel stopper is removed from the flask with a rocking and unscrewing motion. The flask end of the two-way vent tube is now inserted through the hole in the rubber bushing to the point where one of the two dilations on the glass tube just slips into the flask beyond the rubber bushing. The flask is inverted, placed in the ring bracket, and the infusion started.

Calculation of Costs: This method requires an initial equipment which may be divided into two parts, an irreducible item which consists of the reservoir and the filter, and an item which can be expanded according to need, namely, the flasks and their accessories. The rubber tubings and drip and needles do not enter into consideration here as they will have to be used in any case. The irreducible equipment costs about \$1000. If the cost of the flasks for initial use is set at \$500 and to this is added part-time labor of \$50 a month, which can be expanded into full-time labor as the need arises.

the total may be set at \$2200. Any hospital which expends this amount of money a year for infusion fluids will not incur a loss, even in the first year, by adopting this system. Beyond the first year the savings are substantial; the larger the amount of fluids used, the more substantial the savings, since the only items of expense will be the cost of replacements, material and labor.

But the savings are not the entire consideration. The safety from pyrogenic reactions in intravenous medication, especially in blood and plasma banks, should be the major consideration, particularly during war times. Instances are known of tens of liters of blood and plasma which have had to be discarded because the sodium citrate used was pyrogenic. This type of saving has not entered into the above calculation but may be the major item.

SUMMARY AND CONCLUSIONS

1. Methods of intravenous fluid manufacture which depend only upon the purity of distilled water used have no provision for dealing with pyrogen should this substance be present in the final mixture and, therefore, do not entirely eliminate pyrogenic reactions.

2. Chemicals have been shown to be occasionally contaminated with pyrogen.

3. The adsorptive filtration method of preparing assuredly nonpyrogenic fluids, described five years ago, has been developed into a practicable and convenient method for hospital use.

4. The compressed asbestos filter pads, around which this method is built, have been calibrated for speed of filtration and pyrogen retentiveness. The factor of safety is shown to be more than ample for the most exacting use in the manufacture of physiologic salt, dextrose, and sodium citrate solutions.

5. The uniformly nonpyrogenic results of 42 months' use of fluids manufactured according to this method are reported.

6. The mechanical aspects of the method are described.

7. The method not only eliminates pyrogenic reactions but is also more economical than the use of commercial fluids.

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#46
"THE PREPARATION OF NONPYROGENIC INFUSION AND
OTHER INTRAVENOUS FLUIDS BY ADSORPTIVE FILTRATION"

REPORT OF 12 MONTHS' TRIAL

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PART I—BIOLOGIC ASPECTS

IN 1937, there was reported from this laboratory a method of preparing intravenous fluids based on a new principle.^{1,2} As pointed out in that report, it was Siebert's^{3,4} extension of Hort and Penfold's⁵ work which laid the foundation for the various methods of preparing intravenous fluids at present practiced in commercial houses and hospitals. Even the method described by Rademaker^{6,7} and the more recent and mechanically ingenious elaboration by Walter,^{8,9,10} stem from Siebert's original principle. The principle, however, suffers from a critical weakness in the assumption that if the water used in the manufacture of the fluids is pyrogen-free, then the resulting mixture must also be pyrogen-free. But should the water become contaminated, say as a result of failure to clean the still, or should pyrogen be present in the chemicals used in making the solutions, the lack of provision for its removal is a notable disadvantage of the method. It is, in fact, the lack of security behind the old working principle which impels manufacturers to test their intravenous fluids on the rabbit. Unfortunately, not even this precaution has entirely eliminated the occurrence of pyrogenic reactions. As pointed out in a previous communication,¹¹ this failure is perhaps due to the inadequacy of the test doses in use.

That chemicals occasionally are contaminated with pyrogen has not heretofore entered into consideration. In previous papers¹² we have reported on the pyrogenicity of samples of inulin as they have come fresh from the manufacturer.

Table I shows the effect on test animals of samples of sodium chloride, of dextrose, and of sodium citrate, all "commercially" and "technically pure," which had been made into solutions with nonpyrogenic water and tested immediately after the containers had been opened. The sodium chloride sample was the more reactive of two found pyrogenic in a test of 38 samples, an incidence of slightly more than 7 per cent. The dextrose was the most reactive of four positive samples of 32 tested, an incidence of 14 per cent, and the sodium citrate was the most reactive of four of 18 tested samples, an incidence of over 22 per cent. It may also be mentioned that sodium citrate

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is usually more heavily contaminated than dextrose which, in turn, is usually more heavily contaminated than sodium chloride. Because of the fact that test animals injected with such large doses of sodium citrate are liable to die in cardiac arrest, perhaps as a result of the blood calcium being thrown out of ionization, it is necessary to add an equivalent dose of calcium chloride to the sodium citrate test solution before it is injected.

TABLE I EFFECT OF VARIOUS COMMERCIAL SOLUTIONS						
Expt. No	Chemicals	Test Dose Gm.	Animal Used	Weight Kg	Temp. Rise Deg. F	Leukocyte Count Change x 1000
1	Sodium chloride	Vol 4 50	Rabbit	3	1.2	
2	Sodium chloride	12 1500	Dog	15	2.0	-5.7
3	Dextrose	7.5 150	Rabbit	3	2.0	
4	Dextrose	75 1500	Dog	15	2.4	-9.4
5	Sodium citrate*	0.28 11.5	Rabbit	3	2.2	
6	Sodium citrate*	2.8 115	Dog	15	2.8	-8.2

*These dosages were calculated on the basis that the maximum dose of sodium citrate given to a 60-Kg man is 5.6 Gm. (3 liters of blood), and that the rabbit is approximately three times and the dog six times less sensitive to pyrogen than man. An equivalent amount of nonpyrogenic calcium chloride was added to each sodium citrate solution.

The chemicals in these cases were perhaps contaminated by pyrogen in the process of manufacture. Nonpyrogenic chemicals even in the "dry" state can, however, become pyrogenic on standing under unsterile conditions, as has been found by Smith¹³ to be the case with inulin, and by us with dextrose, sodium citrate and heparin. Apparently the better medium the chemical is for bacterial growth, the more easily it becomes pyrogenic. Humidity and a warm environmental temperature seem to accelerate pyrogen production, probably by acceleration of bacterial growth. Among aerial contaminants, Co Tui and Schrift^{14,9} have found *B. proteus vulgaris* and *B. subtilis* fairly prolific pyrogen producers. In the light of these findings, therefore, the assumption that to guard against the occurrence of pyrogenic reactions it is necessary only to keep the water pyrogen-free can be costly as well as unsafe. The problem will perhaps become increasingly acute with the increasing use of blood and plasma transfusions.

The adsorptive filtration method of preparing intravenous fluids differs from the older method in that it is based on the principle of filtering the resulting mixture of chemicals and distilled water through specially prepared compressed asbestos pads proved to remove pyrogen. This method is efficacious not only for infusion fluids but for all crystalloidal intravenous medications. With colloidal substances such as sera and gum acacia, however, it is ineffective. The method has been in experimental use on the New York University Surgical Division of the Bellevue Hospital continuously since July, 1937 except for an intermission of 16 months. It is the purpose of the present communication to summarize the results of the experiment as well as to describe the practical method of manufacture which has been evolved.

The practical difficulties encountered in the course of this work may be briefly discussed since they are difficulties which many hospitals and similar institutions which undertake the manufacture of their own infusion fluids will have to face.

1. The first problem was the establishment of the criterion of pyrogenicity. With fluids which are frankly pyrogenic, this is an easy matter since the symptom-complex of the pyrogenic reaction is a familiar one. The difficulty was with fluids containing a borderline dose of pyrogen, which would elicit only a slight temperature rise easily missed in the four-hour routine ward temperature readings but might, when administered in doses two or three times larger than the usual one, give rise to a fairly severe reaction.

It was, therefore, necessary during the first six months of this experimental period to subject the fluids made to animal tests, using the rabbit and the dog, according to the technique described elsewhere.¹¹ If the fluids were found nonpyrogenic according to the tests, they were then released for ward use. The temperature readings of all patients receiving the fluid were taken hourly for four hours, both during and after the administration of the fluid. In this way transient temperature rises could be detected.

2. The second problem was the elimination of contamination from external sources, such as the rubber tubings, glass connections and needles used in the dispensing set. Originally, the latter were sterilized in the ward instrument sterilizers in common with contaminated pus basins and surgical instruments. The occurrence of four reactions on patients in the first three weeks of this period led to tests of the water from the sterilizer. Ten and 15 cc. samples of this water gave unmistakable reactions in both rabbits and dogs. This result led to the institution of a method of washing, packing and sterilizing the dispensing sets which will be described in a later section of this paper.

3. A third difficulty was the presence of a yellowish-brown coloration in the solutions after they had been autoclaved. This was first thought to be due to caramelization but has been attributed by Krno¹² to the production of levulinic acid.¹⁰ In our preparation two factors were found responsible for the coloration: (a) The asbestos pads originally used were made for the filtration of fluids which did not have to be autoclaved. They imparted some principle to the filtrate which caused the formation of this coloration on heating. The adoption of a pad specially prepared for infusion fluid use has eliminated this factor; and (b) the second factor was overheating of the solution. This occurred when, after sterilization, the autoclave pressure was permitted to sink so fast that the fluid inside the flasks began to boil over. The film of fluid on the inner wall of the neck of the flasks evaporated and the residue became browned, imparting the coloration to the rest of the solution. The remedy to this was obviously to lower the autoclave pressure so slowly that no boiling over took place.

4. The fourth and most serious difficulty was the presence of shreds and particles in the filtrate. While the injection of solutions containing these particles into numerous experimental animals caused no detectable symptoms,

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their presence was none the less objectionable. It was because of this difficulty that the use of these fluids was suspended for 16 months in 1938 and the first part of 1939, until a manufacturer could be found who would cooperate in devising means of eliminating these particles. Jena filters, Chamberland and Berkefeld filters were interposed at the outlet of the asbestos filter without avail. Finally, the difficulty was overcome by both improving the quality of the filter pad and interposing an aloxite fiber eliminator between the asbestos filter and the collecting bottle.

5. Standardization of filter pads. The compressed asbestos pads, as now prepared for our use by the Ertel Engineering Corp., are calibrated with regard to two properties: (a) The speed of filtration; and (b) degree of pyrogen retentiveness.

Speed of Filtration.—This is controlled by three factors: The filtering pressure applied; the filtering area; and the composition of the pad. The filtering pressure is *safest at 12 lbs.*, and the filtering area is limited by the size of filter practicable in hospitals. Our present filter takes pads of 12 inches, with an actual filtering area of 29.5 cm. in diameter. The pads are made of asbestos and pulp. The more asbestos used, the greater the pyrogen retentiveness, but the slower the filtration rate. The interplay of these two factors, therefore, determines not only the rate of filtration but also the efficiency of the pads to remove pyrogen. The pad now in use has a delivery volume of two liters every 2.5 minutes. A six-hour filtration would, thus, yield 288 liters, which would be more than enough to meet the daily requirements of a fairly large hospital. Hospitals using larger amounts of fluid may enlarge the filtering area of the filter by the method to be described later or install two such filters.

Pyrogen Retentiveness of the Filter Pads.—The ability of these pads to remove pyrogen was shown in a previous work¹⁶ to be due to adsorption and not to sieving. This means that the property can be exhausted. It is, therefore, necessary so to standardize the pads that they are not only uniform but also possess such a margin of safety in pyrogen retentiveness that they remove the maximum amount of pyrogen which can occur in the distilled waters and the chemicals in use.

In order to calibrate the pads in this respect it was necessary to secure a sample of pyrogen and use it as a standard. The pyrogen used in this work was isolated from the cell-free Berkefeld filtrate of the supernatant portion of concentrated stock suspension of typhoid vaccine (1 cc. = 88 billion cells). From seven liters of this suspension, four grams of the substance was yielded. The method of isolation as well as the chemical and biologic aspects of this substance will be reported elsewhere. It is necessary, however, to state that the substance is still in the crude stage, containing six per cent nitrogen, which on further purification has been reduced to 1.6 per cent. The biologic (pyrogenic) potency of this crude sample, called "pyrogen-A," has been determined on rabbits, dogs, and men. Table II shows the effect of different doses of this preparation. If the minimum effective dose (MED) is arbi-

trarily defined as that dose, per kilo body-weight, which on intravenous injection will cause a temperature rise of 0.8° to 1.0°F. , the MED in rabbits is 0.1 *gamma*, in dogs, 0.4 *gamma*, and 0.03 *gamma* in men. The reason for using this crude substance as a standard instead of the more purified substance is that the process of purification is so tedious that it may be a long time before enough of the purer substance can be available for this purpose. In any case, the biologic calibration of "pyrogen-A" makes it unnecessary to wait for the purer preparation.

TABLE II
EFFECT OF DOSES OF PYROGEN-"A" IN RABBITS, DOGS, AND MEN

Subject	Weight Kg	Dose per Kg	Temp. Change Deg. F.	Leukocyte Count Change x 1000
Rabbit 1	3.0	0.12	0.8	
Rabbit 2	2.5	0.12	1.0	
Rabbit 3	3.0	0.12	0.9	
Rabbit 4	3.0	2.0	3.5	
Dog 1.	15.0	0.3	1.0	-6.1
Dog 2	16.0	0.3	1.1	-5.5
Dog 3	15.5	0.3	0.8	
Dog 4	16.0	2.0	3.2	-10.2
Man 1	52.0	0.04	1.2	
Man 2	64.0	0.04	0.9	
Man 3	72.0	0.04	1.1	
Man 4.	70.5	2.0	5.0	

Process of Calibrating the Pads.—One milligram of "pyrogen-A" (1000 *gammas*) is weighed out on a sensitive analytic balance and dissolved in 100 cc. of pyrogen-free distilled water. Ten cubic centimeters of this solution contains 10 *gammas* of pyrogen. Amounts of from five cubic centimeters up, in multiples of five, are filtered through test pads of a convenient size mounted in convenient filters. The filtrates are then tested in both rabbits and dogs. Table III shows the protocols of some of these tests.

TABLE III

Experimental Animal	Weight Kg.	CALIBRATION OF FILTER PADS		Temp. Change Deg. F.	Leukocyte Count Change x 1000
		Volume Prefiltrate	Total Dose Pyrogen in Prefiltrate <i>Gamma</i>		
Rabbit 1	2.5	30	300	-2	
Dog 1	16.0	30	300	+1	-2
Rabbit 2	3.0	35	350	+2	
Dog 2	15.0	35	350	25	-3
Rabbit 3.	3.0	40	400	+15	
Dog 3.	15.5	40	400	+1	+1
Rabbit 4	2.5	45	450	+4	
Dog 4	16.5	45	450	+2	-8
Rabbit 5	3.0	50	500	+4	
Dog 5	15.5	50	500	+8	-1.2

It will be seen from Table III that the pad removes nearly 500 *gammas* of "pyrogen-A." Since the pad is 2.5 cm. in diameter, it has a surface of 49 sq. cm., and, therefore, each square centimeter removes over 100 *gammas* of "pyrogen-A." The filtering surface of the four large filter pads used in the hospital filter assembly are each 29.5 cm. in diameter, making a total filtering area of 2734 sq. cm. The amount of pyrogen removable by the entire filter

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assembly is approximately 273,400 *gammas*. Since about 140 *gammas* are required to cause a temperature rise of 5.6° F. in an average man of 70 kilos, this filtering area will remove about 1953 such doses.

When applied to solutions of sodium chloride or dextrose in distilled water this range of safety is more than ample as the amount of pyrogen contained in any combination of these three substances in the usual dose is tenuous and, further, the filter-pad assembly is made to deliver not more than 288 liters with each filtration. The case of sodium citrate requires special consideration. As shown in Table I, gram for gram, the contamination of pyrogen in sodium citrate is heavier. This, however, is more than offset by the fact that the dose of sodium citrate used per 500 cc. of blood is usually in the neighborhood of 1875 mg. in 75 cc. of water. This, by weight, is only about one-fifth of the dose of sodium chloride in one liter of physiologic salt solution and less than one-twenty-seventh of the dose of one liter of 5 per cent dextrose solution. Therefore, in making sodium citrate solutions, it is only necessary to use two filter pads and restrict the manufacture to not more than 300 doses.

These calculations are admittedly based on biologic effects, and assume that pyrogen from sources other than the typhoid bacilli have the same potency per unit-weight as typhoid pyrogen. This assumption has no direct experimental basis at present. The only justification for applying the figures derived from "pyrogen-A" to the pyrogenic substances found in these fluids, whose exact source is unknown, is that they have so far held true in the samples whose pyrogenic content was roughly determined by the biologic test, then filtered through asbestos pads of the required area and then retested and found negative for pyrogen. In the absence of more exact knowledge, this approximate correspondence for the present can be used as a rough working principle. This method of pretesting before filtration may be used for any intravenous material whose maximum pyrogenic content has not been as thoroughly studied as have those of dextrose, sodium chloride, and sodium citrate.

RESULTS OF 42 MONTHS' TRIAL

During this experimental period, 3867 liters of intravenous fluids prepared by the nonpyrogenic method have been administered. They may be tabulated as follows:

No. of Administrations	Amount Given Liters	Total Given Liters
1815	1	1815
652	2	1304
123	3	492
64	4	256
Totals 2654		3867 liters

In none of these was there a reaction. One patient who complained of a chilly sensation after receiving 1000 cc. did not have a temperature rise.

is clear that if any pyrogen was present in the solutions, it was in such minute amounts as to be below the MFD even in three to four liters.

PART II—MECHANICAL ASPECTS

The parts which comprise the entire filter assembly may now be enumerated and the more important items described. The filtering portion of the assembly is diagrammatically represented in Figure 1. It consists of a compressed nitrogen or air tank, A, which furnishes filtration pressure; the reservoir, B, which holds the prefiltered mixture, the filter itself, C; the aloxite fiber eliminator, D; and the collecting flask, E.

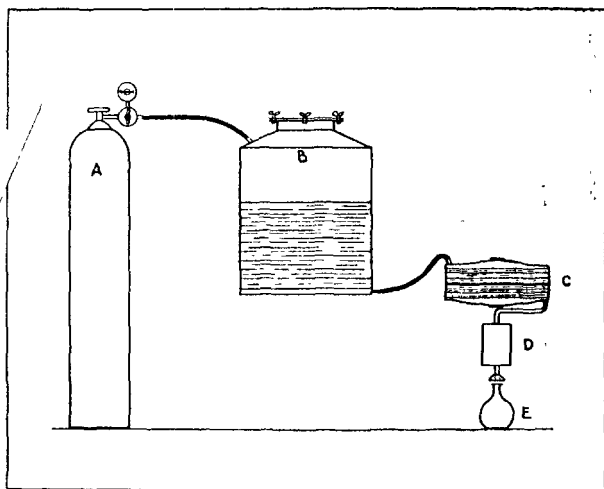


FIG 1.

The reservoir, usually of 250 liter capacity, is made of stainless steel, and may be equipped with an electric stirrer.

Figure 2 illustrates the essential parts of the filter as well as the path taken by the solution passing through it. It may be seen that each filter pad is held between two perforated Monel metal plates or frames, the three forming a pad-frame unit. Two such units form a compartment. The fluid flows from the solution inlet, A, into a conduit system, B, which splits it into two streams. One stream filters through one pad downward and the other upward to meet in the collecting chamber, C, situated between the two pad-frames. From C the solution is conducted into the second compartment to have the process repeated, after which the conjoint stream from the second collecting chamber, D, passes through the fiber eliminator into the collecting flask. Both the latter are to be described in a subsequent section.

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Since provision is made in the present filter to accommodate two additional compartments, the filter has a still wider range of usefulness than has been calculated. Any one of the three factors—capacity, speed of filtration, or retentiveness—can be increased two-fold by this doubling of the filtration area. The stream in this case would be split in four instead of two as in the filter illustrated in Figure 2.

Figure 3 is a photograph of the filter ensemble. A is the spindle with which the pads are loaded and unloaded and the filtering system rendered watertight. B is the filtering system, C is the pressure gauge, D the housing for the aloxite fiber eliminator, and E the outlet. The self-autoclav-

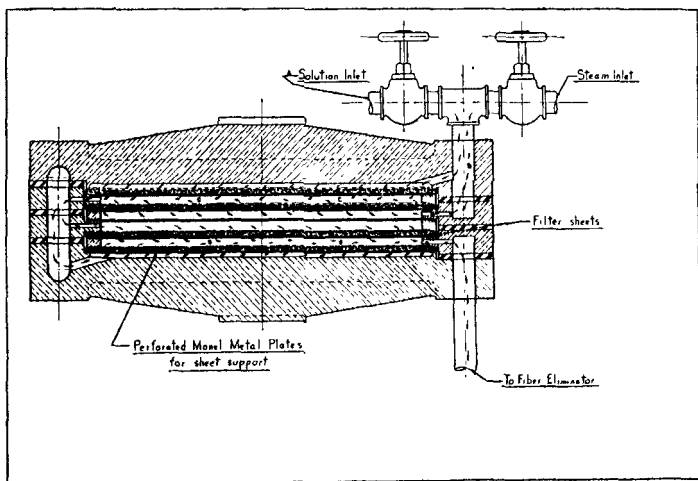


FIG. 2.

ing system, F, consists of a steam generator in which the source of steam used is pyrogen-free distilled water. It may be heated electrically, by gas, by house-steam, or by an oil burner. The steam generated is led into the interior of the filter by the flexible hose, G. The course of the steam through the filter is identical with that of the fluid which follows it and whose course has been described in the foregoing paragraph.

The aloxite fiber eliminator is a thick, finely porous filter made of carborundum. Because of its composition it is sturdy and may be heated to a temperature high enough to destroy any pyrogen which may have developed in the interstices of the stone.

The dispensing part of this system, mainly the collecting and storing flasks and accessories, is essentially the same as that described by Walter.^{8, 9, 10} It is the most satisfactory one available at present, made to withstand hard

usage, and in three years of use has suffered the minimum amount of breakage. It has, however, the slight inconvenience of having too many parts.

Figure 4 illustrates the part of this system which consists of: A, pyrex flasks of specially rugged construction of one to two liters capacity, preferably graduated; B, a one-hole rubber bushing to fit the flask; C, a special stainless steel stopper with a partially grooved stem, D, a two-way vent tube for the administration of the fluids and for the admission of air into the flasks to displace the fluids.

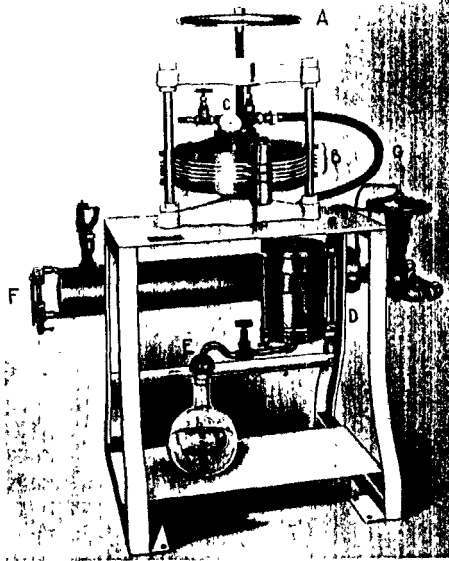


FIG. 3.

The bushing, B, is of special rubber which can stand 500 sterilizations without shrinking. It is provided with a hole in the center for the insertion of the two-way vent tube when fluid is being administered, and for the insertion of the steel stopper when the flask is not in use. The stopper is provided with a partially grooved stem which is fitted partly into the hole of the bushing while sterilization takes place. The groove allows equalization of inside pressure with outside pressure. When sterilization is completed, the stem of the stopper is pushed all the way in, as in E, the umbrella of the stopper serving as an hermetic seal for the rubber bushing.

The original Fenwal-tube is short, so that the flask end is submerged in the fluid when the flask is inverted for administration. There are two objec-

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tions to this feature: First, when the infusion is being given, the air which enters the flask to displace the fluid has to be bubbled through the fluid. This, in effect, means the washing of 1000 to 2000 cc. of unsterile air in the infusion fluid. Bacteriologic cultures of ten samples of fluid subjected to this kind of air-washing gave three positive cultures. By the simple device of

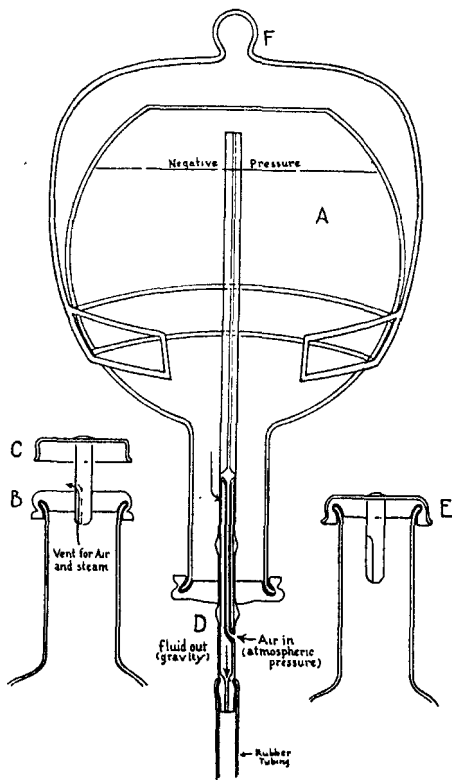


FIG. 4

lengthening the tube so that the flask end extends above the surface of the fluid when the flask is inverted, this washing effect is avoided.

No positive cultures were found in ten fluids subjected to this system of air displacement. If it is desired to admit absolutely sterile air into the flask, the air vent may be modified in such a way as to accommodate a small sterile Berkefeld candle. This was originally used by us but later discontinued because of inconvenience.

The second objection to the short original Fenwal-vent-tube is that unless special care is taken, the fluid is as liable to flow through the air vent as through the fluid vent.

The split ring bracket, F, is simply a wire basket for suspending the flask in an inverted position when the flask is in use.

Some reference must be made to the still which delivers distilled water for making the solutions. Since the still is the heart of all other processes, much emphasis has been laid on it. While it is our opinion that the undue concentration on the still has lulled to rest suspicion of other possible sources of pyrogen, we believe that the still must be cleaned periodically so that it adds no pyrogen to the distilled water to lower the factor of safety of the filter. Moreover, it must be large enough to deliver the necessary amount of distilled water to be used for cleansing as well as to make the solutions themselves. A still that delivers 10 gallons of water per hour (40 liters), will be sufficient to meet the needs of the average hospital. The periodic cleaning also does away with incrustations which lower the output of the still.

CARE OF THE PARTS

The Reservoir. The reservoir when new must be rinsed first with soap and water and then copiously with distilled water. Subsequent care after every use consists in rinsing it out thoroughly with distilled water and then drying it.

The Filter. When new, the conduit system of the filter is rinsed thoroughly with distilled water. Thereafter, the same rinsing process is repeated after each use.

The Aloxite Fiber Eliminator. After every use, the surface of the stone fiber eliminator is simply flushed thoroughly in order to wash away the particles and shreds which may be caught on its surface. After washing, it is placed in an electric oven at a temperature of 120° C. for one hour to dry. This temperature sterilizes the stone so that pyrogen production within the interstices of the stone is halted.

Fenwal-Flasks, etc. The initial cleaning and subsequent care of the Fenwal-flasks, rubber bushings, steel cages, and the modified vent tube have been described by Walter and need no extended treatment here. Suffice it to say, that the flasks are washed and brushed with hot soapsuds and rinsed first in tap and then in distilled water until the walls run homogeneously clear. The same treatment should be applied to the other glassware, namely, the vent tube and the drip as well as to the steel stopper. An occasional treatment of the glassware with a cleaning mixture of 1.5 per cent solution of sodium dichromate in concentrated sulfuric acid may have to be resorted to in order to get rid of spots in the glassware difficult to eliminate otherwise. The rubber bushings when new are first scrubbed with soap and water until no further powdery substance is given off, then rinsed with

distilled water. The rubber tubings when new are likewise rinsed well with hot soapsuds and then with distilled water. Immediately after each washing, the rubber tubings, the vent tubes, the drip and the needles are wrapped and packed in a convenient container. We have used for this purpose a specially constructed copper cylinder, one foot long and $2\frac{1}{2}$ inches in diameter, provided with a detachable cap as well as holes in the wall to allow penetration of the steam in the autoclave. After sterilization, the holes are closed by a special sliding device and the set stored for future use.

Preparation for Filtration: The flasks without the rubber bushings in place are sterilized in the hospital autoclave. The rubber bushings and the steel stoppers are also sterilized but separately from the flasks.

The filter with pads mounted on the frames is sterilized by its own self-autoclave for 30 minutes at 18 pounds pressure. After that it is allowed to cool to a temperature convenient for handling.

The solution is mixed in the reservoir. The reservoir is then connected, on the one hand, with the nitrogen tank and, on the other hand, with the inlet of the filter. The collecting and storing flask is placed at the outflow tube of the filter and filtration is begun by turning on the gas gradually to 12 pounds. After discarding the first four liters, the flasks are then filled one by one.

After collection, the flasks are partially stoppered with the stainless steel stopper in such a way that the groove in the stem of the stopper allows communication of the inside of the flask with the outside (Fig 4). The flasks are then put into the autoclave and sterilized at 250° F. for 30 minutes. After sterilization, special care is taken to have the pressure in the autoclave gradually diminished so as not to cause boiling. The stem of the steel stopper may then be pushed home, in which case a partial vacuum is created as the solution cools. This partial vacuum, which helps retain the stopper in place and causes a water-hammer effect when the bottom of the flask is struck with the fist, may be used to indicate whether the stopper is leaking after storage.

After the flask is sealed, it is stored until needed.

Just before administering the fluid, the steel stopper is removed from the flask with a rocking and unscrewing motion. The flask end of the two-way vent tube is now inserted through the hole in the rubber bushing to the point where one of the two dilations on the glass tube just slips into the flask beyond the rubber bushing. The flask is inverted, placed in the ring bracket, and the infusion started.

Calculation of Costs: This method requires an initial equipment which may be divided into two parts, an irreducible item which consists of the reservoir and the filter, and an item which can be expanded according to need, namely, the flasks and their accessories. The rubber tubings and drip and needles do not enter into consideration here as they will have to be used in any case. The irreducible equipment costs about \$1000. If the cost of the flasks for initial use is set at \$600 and to this is added part-time labor of \$50 a month, which can be expanded into full-time labor as the need arises,

the total may be set at \$2200. Any hospital which expends this amount of money a year for infusion fluids will not incur a loss, even in the first year, by adopting this system. Beyond the first year the savings are substantial; the larger the amount of fluids used, the more substantial the savings, since the only items of expense will be the cost of replacements, material and labor.

But the savings are not the entire consideration. The safety from pyrogenic reactions in intravenous medication, especially in blood and plasma banks, should be the major consideration, particularly during war times. Instances are known of tens of liters of blood and plasma which have had to be discarded because the sodium citrate used was pyrogenic. This type of saving has not entered into the above calculation but may be the major item.

SUMMARY AND CONCLUSIONS

1. Methods of intravenous fluid manufacture which depend only upon the purity of distilled water used have no provision for dealing with pyrogen should this substance be present in the final mixture and, therefore, do not entirely eliminate pyrogenic reactions.

2. Chemicals have been shown to be occasionally contaminated with pyrogen.

3. The adsorptive filtration method of preparing assuredly nonpyrogenic fluids, described five years ago, has been developed into a practicable and convenient method for hospital use.

4. The compressed asbestos filter pads, around which this method is built, have been calibrated for speed of filtration and pyrogen retentiveness. The factor of safety is shown to be more than ample for the most exacting use in the manufacture of physiologic salt, dextrose, and sodium citrate solutions.

5. The uniformly nonpyrogenic results of 42 months' use of fluids manufactured according to this method are reported.

6. The mechanical aspects of the method are described.

7. The method not only eliminates pyrogenic reactions but is also more economical than the use of commercial fluids.

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