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Care and Observation of a Germ-free Neonate

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Within the spectrum of childhood diseases that present with recurrent and persistent infections, the 'combined immunity deficiency syndrome' (Soothill, 1967) or 'Swiss' type of a-γ-globulinaemia (Hitzig, Barandun, and Cottier, 1968) has recently been well defined. This familial disease is considered to result from a recessive genetic inheritance (Hitzig and Willi, 1961). Treatment has been invariably unsuccessful and every recorded case, like the previous child in this family (Thompson 1967), has died in early life. When this mother became pregnant again it was decided to deliver and maintain the infant in the germ-free state until it was established whether it was affected. In the event of the 'combined immunity deficiency syndrome' being present, it was proposed to treat the infant in the germ-free unit with a fetal tissue graft. In this environment it was postulated that the infant's immunological state would be relatively immature in the absence of foreign antigenic stimulation from the normal bacterial flora.

The infant was delivered by elective caesarean section into a flexible plastic sterile surgical isolator, and passed directly into a connected transfer isolator for resuscitation. This sterile transfer isolator was then moved into the vicinity of the larger sterile maintenance isolator, assembled in a conventional hospital ward. The infant was introduced into this isolator aseptically (Barnes *et al.*, 1968b), and the medical and nursing care of the infant during the period of investigation is described here.

Apparatus and Methods

Maintenance isolator. The Cornercroft type 'A' human germ-free isolator (Fig. 1) has been described elsewhere (Barnes, Tuffrey, and Cook, 1968d), with the method for its assembly and sterilization (Barnes, Holliday, and Cook, 1968c). The method of sealing the plastic tent to the consoles was modified slightly to incorporate high pressure pneumatic/hydraulic seals. The isolator was fitted with certain of the fail-safe devices described and used with our prototype, the germ-free animal isolator (Cook, Tuffrey, and Barnes, 1968). The microbiological efficiency of the Cornercroft type 'A' human isolator and of the method of sterilization was initially confirmed by maintaining mice germ-free for 7 days (Barnes *et al.*, 1968c).

For the care of the infant, it was necessary to include a 'nylon'-coated metal cot and a polyvinyl chloride (PVC) sealed mattress. A PVC sealed electrically heated incubator, sterilized by ethylene oxide gas, was also installed before sterilization of the assembled isolator. An electricity supply for the incubator (6 V) and for the thermostatically controlled bottle warmer (230 V) was supplied with the filtered oxygen and suction lines through the wall of the tent. A stethoscope was also provided through the tent wall. Gloved sleeves necessary for nursing and medical care formed an integral part of the tent.

Transfer isolator. The transfer isolator was designed to receive the infant after aseptic caesarean section delivery into the joined but independent surgical isolator. The mobile transfer isolator incorporated all features necessary for resuscitation and was specially designed to allow the subsequent aseptic introduction of the infant into the larger maintenance isolator. The surgical procedure, the apparatus required for the germ-free delivery, and the method for introduction of the infant into the maintenance isolator have already been described (Barnes *et al.*, 1968b).

Preparation of isolation system. The preparation and sterilization of the three different isolation units have already been described (Barnes *et al.*, 1968c, d). The nursing and medical requisites for the proposed care of the infant in the isolator were sterilized in advance. The method of sterilization selected depended primarily on the nature of the object and its ability to withstand the proposed method of sterilization. Articles likely to be required were first sealed in two layers of nylon or PVC film before sterilization.

To facilitate nursing care and to reduce the number of times that objects had to be introduced into the maintenance isolator, certain items were sealed together, e.g. feeding, clothing, and bedding packs. After removal of the protective film, articles were introduced through the entry lock of the maintenance isolator.

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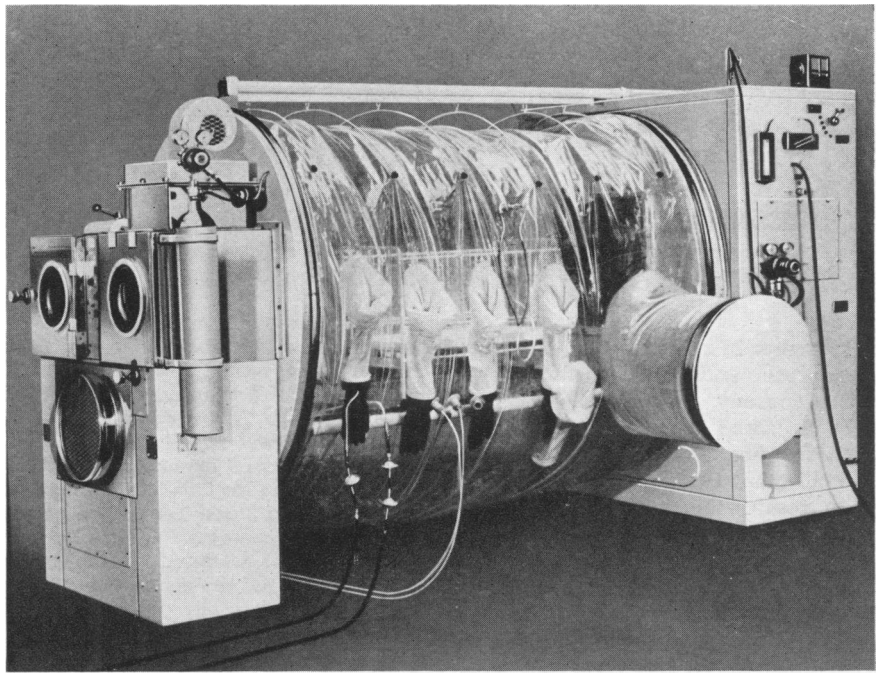

FIG. 1.—*Cornercroft type 'A' human isolator.*

γ-irradiation was most frequently employed to sterilize objects for use inside the maintenance isolator. Other means of sterilization were only used if the substance of the article was known to react unfavourably to such treatment or where there was any doubt about the reaction, e.g. drugs. A relatively high dose of 4·0 M rads was administered from a ^{60}Co source, and radiation was confirmed by observation of the colour change in the radiation indicators. Care was taken not to use objects such as syringes if they had been previously γ-irradiated by the manufacturer. Here it was anticipated that a cumulative dose of 6·5 M rad might seriously alter the physical properties of the plastic components and render the objects brittle.

Ethylene oxide gas was selected for the sterilization of the incubator. When articles were sterilized by gas, cotton wool wicks were incorporated into the sealing coats of plastic film to facilitate entry and exit of the sterilizing gas. γ-irradiation is known to alter the colour of glass, and therefore gas sterilization was the method of choice for such instruments as a laryngoscope and ophthalmoscope. Gas sterilization was confirmed by the colour change in indicator tape attached to the inner aspect of the article to be sterilized. Ethylene oxide was the agent of choice for the sterilization of the air filters, for the maintenance isolator, and for the outer surface of the cans of evaporated milk. The evaporated milk itself was presumed sterile upon data supplied by the manufacturer, and this was further confirmed by the successful care of pigs germ-free with this particular product (P. C. Trexler, 1967, personal communication). Unlabelled and undamaged cans of milk were carefully sealed in PVC film for ethylene oxide sterilization. A list of drugs likely to be needed in the isolator in the event of an emergency was prepared. By using the parental and presumed sterile form, it was only necessary to sterilize the outer surface of the ampoule, and this was done with a 10% formalin in 1% Tego solution. After a minimum exposure of 5 hours the ampoule was ready for entry into the isolator. Sterilization of the ampoules was performed in advance, and it only remained then to remove the ampoule from the antiseptic solution for its immediate introduction into the isolator through the entry lock. Where ampoules were supplied with paper labels these were first sealed with 'Nobecutane' before being immersed in the germicidal fluids.

Medical and nursing requisites, apart from those introduced before the sterilization of the isolator, had to be introduced through the entry port. This port consisted of a chamber and two electrically-linked doors. This chamber was provided with four ultraviolet lights for sterilizing the interior, which were extinguished automatically on opening either door. When the outer door was opened for the introduction of articles, a high velocity stream of sterile air was directed down and out through the open door of the port (Fig. 2). In practice, with the outer door of the port open upwards, contamination by droplet was prevented by the mechanical barrier of the door, and, in addition, chance contamination was minimized by the sterile air washing that occurred at this time. In

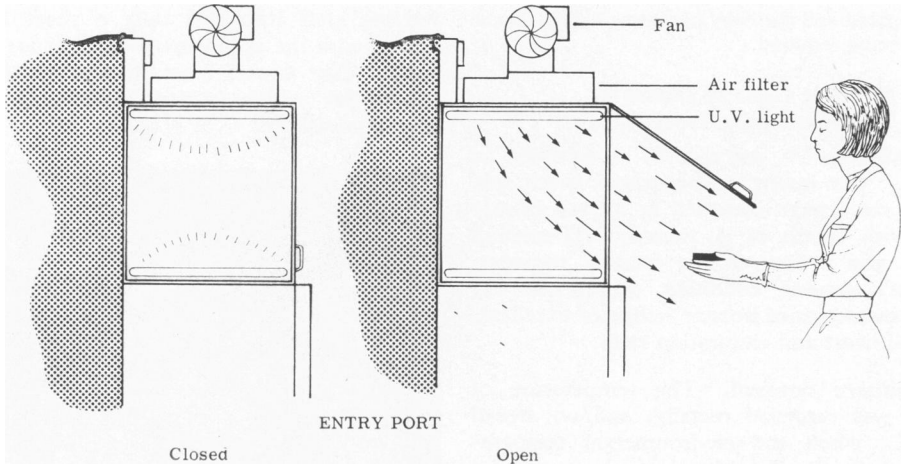

FIG. 2.—*The entry port.*

this protected area, the layers of plastic film were aseptically removed and the objects were introduced into the chamber. Closure of the outer door resulted in the cessation of the air washing and switching on of ultraviolet lights. An ultraviolet sterilization cycle of 6 minutes started, and during this period both doors of the port were locked electrically. This cycle was designed to eliminate any chance bacterial contaminant, and at the end of this period, the inner door could be opened to allow the article to be introduced into the isolator.

Removal of objects from the isolator was more simply performed through the exit port. This chamber, with an inner and outer door, was also provided with an ultraviolet light, and like the entry port, this was extinguished on opening either door. In this chamber both doors were again interlinked to prevent the chance of them being opened together. The linkage here, however, was solely mechanical and independent of any electrical supply. This was so designed that in an emergency and independent of an electricity supply, objects could be introduced into the isolator quickly by using the exit port in reverse.

Delivery and transfer of infant to maintenance isolator. The apparatus and technique for the aseptic delivery of the infant and the subsequent transfer have been described in detail elsewhere (Barnes *et al.*, 1968b). Here it is only necessary to note that the infant was introduced into the maintenance isolator through a temporary tunnel formed from the union of adhesive membranes constructed into both the maintenance and transfer isolators (Fig. 3). Before the arrival of the infant in the maintenance isolator the

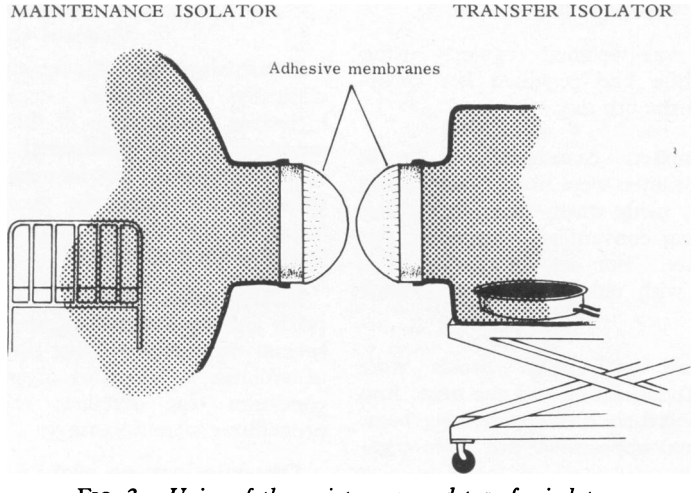

FIG. 3.—*Union of the maintenance and transfer isolator.*

bed was prepared and the baby incubator heated, in the event of its being required.

Nursing Care of Infant in Maintenance Isolator

Observations. A nurse was constantly in attendance. The infant's temperature, heart, and respiration rates were recorded at quarter-hourly intervals, with details of all nursing and medical procedures and a note of the infant's response. The infant's general condition throughout her stay in the maintenance isolator remained excellent, with normal heart and respiration rates.

Temperature control. The temperature of the infant was recorded rectally, and on arrival was 35° C. when the environmental temperature was 25·6° C. Additional heating provided in the ward caused the isolator environmental temperature to rise to 29·4° C., restoring the infant's temperature to normal. After this the isolator environmental temperature was easily maintained at 26·7 to 27·8° C. by controlling the temperature in the surrounding room. This proved to be completely satisfactory, and the electrically heated incubator was never required.

Feeding. After the initial glucose feed the infant was fed with diluted sweetened evaporated milk. Cans of evaporated milk and water with pre-weighed packets of sugar were used to make up the feed in a disposable feeding bottle inside the isolator in the standard manner. The feed was then heated in the thermostatically controlled electrically heated bottle-warmer. For feeding, the infant was held in the normal position in the nurse's gloved arms without difficulty (Fig. 4).

Weighing. She was weighed regularly using a spring-balance. She had regained her birth-weight of 3·7 kg. on the 6th day.

Clothing and toilet. Standard but sterile infant clothes and cot linen were used. The infant was washed regularly using warm water and cotton wool, the water being conveniently heated in the feeding bottle-warmer. For care of the cord the standard treatment with chlorhexidine and spirit was employed.

Stool and urine collection. Stools were collected on to polythene sheets and the urine into standard adhesive polythene urine collecting bags. Stools were of normal appearance but were completely odourless.

The child was nursed by the routine ward nursing staff who had only a short instruction period with the apparatus before the arrival of the baby. The nurses soon became used to these procedures which only initially occupied more time than usual.

FIG. 4.—*Handling of the infant.*

Medical Care

Examination. The female infant was examined clinically on several occasions. Examination, including examination of the fundi and tympanic membranes, was performed without undue difficulty. However, the femoral pulses could not be felt and the fontanelle tension was difficult to assess through the rubber gloves. Rectal examination was not performed.

Procedures. Blood was collected by heel-prick and by external jugular venous puncture on several occasions without difficulty. An injection of vitamin K was also given. Since the child's condition was excellent throughout, no other procedures were necessary.

Observations on child's behaviour. Nurses regularly observed and noted the infant's activities

and her responses to various nursing and medical procedures. In addition, certain random behavioural assessments were made to amplify these observations.

A further aim of these observations was to confirm that the normal communication between infant and nurse, which may be essential to normal development, could be established despite the physical barrier imposed by the isolation technique. When the pressure inside the isolator was increased for the introduction of requisites, it was noted that the child initially reacted to this change by waking and crying momentarily. Later reactions were less marked and she was undisturbed by changes in pressure. The child startled normally to loud noises, but she was not apparently disturbed by the noise of the apparatus.

During feeding the normal orientating reflex towards the feeder was observed, with eye-to-eye contact sought and maintained. Further interactions were noted during detailed observations.

> *Observations: December 2, 1967, 12 noon.* 'Crying forcibly, appeared hungry, fed very quickly and forcibly after seeking for teat with mouth. Picked up and settled while bed made, cried when put back to bed, wrapped, and soon settled to sleep.
>
> *10 hours later.* 'Eyes wide open, waving arms and legs and yelling vigorously while temperature being taken, stops when rocked or cuddled.'

The baby's warmth could easily be felt, and presumably the infant experienced the nurse's closeness in the same way, judging by response to gentling. She responded to movements around the isolator.

Normal feeding, sleeping, and wakefulness cycles were quickly established, and during the week's observation period there appeared no evidence of deprivation of any of the infant's needs—whether primary, such as food and warmth, or secondary, such as interest, stimulation and arousal.

Subsequent care. The infant was removed from the isolation unit on the 7th day, immunological tests having shown no evidence of the 'combined immunity deficiency syndrome'.

In addition, bacterial tests proved that the infant was 'germ-free', cultures from the infant and the isolator failing to grow bacteria (Barnes *et al.*, 1968a). After much discussion, it was decided that initial bacteriological colonization should occur in the vicinity of children born by caesarean section three days earlier. The child was therefore transferred in a small portable isolator with strict aseptic precautions to the obstetric unit nursery. Here she remained for 24 hours before being handed over to her mother.

Discussion

Germ-free animal techniques have been established for more than 50 years (Luckey, 1963). The apparatus and techniques have been more widely available since flexible plastic film was first used in the construction of germ-free animal isolation units (Trexler and Reynolds, 1957). Various isolation units have been proposed and tested in recent years for patient care. Permanent bricks and mortar units have been described (Bagshawe, 1964; Mathé, Amiel, and Schwarzenberg, 1964; Bowie *et al.*, 1964; James *et al.*, 1967), but suffer microbiologically by the necessary entry of medical and nursing personnel. Haynes and Hench were the first to use a mobile plastic film isolation unit, and showed a reduction in cross-infection in the treatment of burns (Haynes and Hench, 1965). Schwartz and Perry (1966) later demonstrated the suitability of the same plastic isolator for cancer patients undergoing intensive chemotherapy. The human maintenance isolation unit used here (Barnes *et al.*, 1968d) was first tested with germ-free animals. These animals were introduced to the assembled isolator after sterilization, and the fact that these animals were still germ-free after one week confirmed the microbiological integrity of this isolator (Barnes *et al.*, 1968c). The advantages of a plastic film unit compared with a walk-in bricks-and-mortar unit are not only microbiological: such units are cheaper and can be moved, and the isolator can be quickly assembled in a normal hospital ward.

The indications for germ-free care of this particular patient have been discussed more fully elsewhere (Barnes *et al.*, 1968a), as were the problems concerned with her germ-free delivery and introduction into the maintenance isolator (Barnes *et al.*, 1968b). Germ-free patient care is now being considered for three groups of cases.

(1) Patients with a primary inability to combat infection, such as the potential case of the combined immunity deficiency syndrome treated here. Perhaps in this group fatal granulomatous disease of childhood might also best be treated in a protective germ-free environment.

(2) Patients with exposed tissue damage, e.g. burns, would obviously benefit if infection could be prevented

(3) The largest group of patients are those presenting with a secondary inability to combat infection, such as occurs in aplastic anaemia,

leukaemia, and when cytotoxic and immunosuppressive therapy is being used

In most patients for whom germ-free care will be considered, the problem of initial bacterial decontamination is of paramount importance, but it did not arise in our patient who was delivered into a sterile environment. However, the future and widespread use of such units will undoubtedly be related to the development programme for bacterial decontamination.

Our experience shows that nursing a neonate in the isolation unit is not difficult, and once the nurses had gained some experience, routine nursing procedures such as feeding took perhaps less time than is the case where a barrier procedure involving hand toilet and donning of masks and gowns has to be employed.

After experience with only one patient it is not possible to say whether, despite the fail-safe devices, it is essential for a nurse to be in continuous attendance. The extra fan designed to start automatically on failure of the first fan, the visual and auditory alarm to signal the failure of both fans, with the reserve bank of air cylinders, were all available but were not required. Even if both fans failed, or alternatively if there was a general failure of the electricity supply and the reserve air were exhausted, the patient would be in no danger. The isolator has been so designed that even under these circumstances the tent could not collapse on the patient, and a rough calculation suggested that the baby would have enough air in such a situation for more than 6 hours.

Problems may be anticipated with the provision of a palatable nutritious, yet sterile, diet for older children, but there was no difficulty in meeting the simple dietetic needs of a neonate. Nor was it difficult to satisfy the neonate's emotional needs. The contact in terms of touch and sound between the attendant nurse and the baby was only slightly less intimate than it is in the barrier nursing situation. During the isolation period the infant's father was encouraged to visit, and on such occasions he established physical contact with the child and assisted in the feeding. In certain respects there may be an advantage in the nursing of the young infant in the germ-free isolator in comparison with the conventional barrier nursing procedures. Doctors and nurses have quick and easy access to the child, and the fact that the infant can see the attendant's face without a covering mask may also be an advantage.

Many problems are anticipated with the isolation of older patients. Needs are more complex during these periods whether for movement, activity, stimulation, or relationships—adult or children. The inevitable restriction of these through the isolation technique may lead to apathy, withdrawal, and anxiety, or alternatively to frustration or even sabotage of the isolator procedure. Isolation units have been used previously for the care of children of various ages, and difficulties have not been insurmountable, patients having been maintained in such units for over one month (S. Perry, 1968, personal communication). The difficulties with the care and occupation of such children in isolation units may be overcome in various ways; the introduction of appropriate play materials, providing adequate communications aids—TV, radio, telephone, a 'play room' extension to the isolator—and play leaders, teachers, and nursing staff to provide relationships through which the stress and frustration of the situation can be 'played out' (Bentovim, 1966).

Special provisions may have to be made for the parents to spend much time with their child to give a sense of security. Occasionally an adult may even have to be introduced into the isolator with the child. Careful selection and preparation of both children and their families are essential.

Considerable nursing and medical endeavour is necessary for the care of acutely ill patients requiring much specialized attention (Levitan *et al.*, 1968; Levitan and Perry, 1967). In our patient only simple medical procedures such as blood sampling were necessary and provided no difficulty. It is likely that with a little practice more complicated procedures such as lumbar puncture or setting up of intravenous infusions could be carried out in the isolator.

In the preparation for the arrival of this infant much care was given to the selection, packaging, and sterilization of all medical and nursing requirements likely for an arbitrary isolation period of one month. Sterilization, where possible, was by γ-irradiation, a method that we have routinely used for germ-free animal supplies (Cook *et al.*, 1968). Packaging of different articles together in bedding and feeding packs, etc. facilitated nursing care and also reduced the number of times that the entry lock had to be used, perhaps the weakest link in any germ-free isolation system.

The relative ease and success of the use of the human germ-free isolation unit for the care of our patient shows the practical potential of this form of patient care.

Summary

An infant of a mother who had lost a previous child with the combined immunity deficiency

syndrome ('Swiss' type of a-γ-globulinaemia) was delivered by caesarean section into a sterile surgical isolator and then transferred to a 'germ-free' maintenance isolator where she remained for 7 days. The nursing and medical care of the infant in the 'germ-free' state is described.

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References

Bagshawe, K. D. (1964). Ultra-clean ward for cancer chemotherapy. *Brit. med. J.*, **2**, 871.

Barnes, R. D., Fairweather, D. V. I., Holliday, J., Piesowicz, A., Soothill, J. F., and Tuffrey, M. (1968a). A germfree infant. *Lancet*. In the press.

——, ——, Reynolds, E. O. R., Tuffrey, M., and Holliday, J. (1968b). A technique for the delivery of a germfree child. *J. Obstet. Gynaec. Brit. Cwlth*, **75**, 689.

——, Holliday, J., and Cook, R. (1968c). The use of germfree animals in testing human isolation systems. *J. appl.Bact.*, **31**, 349.

——, Tuffrey, M., and Cook, R. (1968d). A 'germfree' human isolator. *Lancet*, **1**, 622.

Bentovim, A. (1966). Psychological Importance of Play. *Proc. Conf. R. Soc. Health*, p. 151.

Bowie, J. H., Tonkin, R. W., Robson, J. S., and Dixon A. A. (1964). The control of hospital infection by design. *Lancet*, **2**, 1383.

Cook, R., Tuffrey, M., and Barnes, R. D. (1968). A germ-free isolator for surgery in mice. *Lab. Anim.*, **2**, 51.

Haynes, B. W., Jr., and Hench, M. E. (1965). Hospital isolation system for preventing cross-contamination by staphylococcal and pseudomonas organisms in burn wounds. *Ann. Surg.*, **162**, 641.

Hitzig, W. H., Barandun, S., and Cottier, H. (1968). Die schweizerische Form der Agammaglobulinämie. In *Ergebn. Inn. Med. Kinderheilk.*, **27**, 79.

——, and Willi, H. (1961). Hereditäre lymphoplasmocytäre Dysyenesie ('alymphocytose mit Agammaglobulinämie'). *Schweiz. med. Wschr.*, **91**, 1625.

James, K. W., Jameson, B., Kay, H. E. M., Lynch, J., and Ngan, H. (1967). Some practical aspects of intensive cytotoxic therapy. *Lancet*, **1**, 1045.

Levitan, A. A., and Perry, S. (1967). Infectious complications of chemotherapy in a protected environment. *New Engl. J. Med.*, **276**, 881.

——, Seidler, F. M., Strong, C. D., and Herman, L. G. (1968). The role of supportive services in operation of an isolator system. *J. Amer. med. Ass.*, **203**, 1009.

Luckey, T. D. (1963). *Germfree Life and Gnotobiology*. Academic Press, New York.

Mathé, G., Amiel, J. L., and Schwarzenberg, L. (1964). Treatment of total-body irradiation injury in man. *Ann. N.Y. Acad. Sci.*, **114**, 368.

Schwartz, S. A., and Perry, S. (1966). Patient protection in cancer chemotherapy. *J. Amer. med. Ass.*, **197**, 623.

Soothill, J. F. (1967). Immunity deficiency syndromes. *J. roy. Coll. Phycns Lond.*, **2**, 67.

Thompson, E. N. (1967). Thymic lymphocytophthisis with terminal aplastic anaemia. *Proc. roy. Soc. Med.*, **60**, 895.

Trexler, P. C., and Reynolds, L. I. (1957). Flexible film apparatus for the rearing and use of germfree animals. *Appl. Microbiol.*, **5**, 406.

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