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REANIMATION OF ADULT RATS FROM BODY TEMPERATURES BETWEEN 0 AND +2° C

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A wide variety of isolated mammalian cells and tissues survive cooling to and storage at 0 to +5° C (Smith, 1954; Parkes, 1954). By contrast, intact adult non-hibernating mammals are sensitive to moderate reduction in body temperature. The lowest temperature ever recorded from a human being who subsequently recovered is 18° C (Laufman, 1951). The lethal body temperatures for mammals of several species have been determined, and 15° C is the generally accepted lethal level for adult rats (Adolph, 1948). Heart beats have been recorded, however, from rats cooled to 8–12° C (Kayser & Hiebel, 1952). Rats with body temperatures between 15 and 20° C are in a state of cold narcosis in which hypophysectomy and other major operations can be performed (Giaja & Andjus, 1949). In dogs and men, body temperatures below 25° C are usually regarded as unsafe for surgical anaesthesia because of the risk of ventricular fibrillation and cardiac arrest (Swan, Zeavin, Holmes & Montgomery, 1953; Bigelow, Callaghan & Hopps, 1950; Lynn, Melrose, Churchill-Davidson & McMillan, 1954). In recent years research has been concentrated on studying the effects of cooling mammals to body temperatures between 15 and 30° C. Comparatively little attention has been devoted to reanimating animals with temperatures below 15° C or after cessation of breathing and heart beat. This problem is of theoretical interest; when solved it might have practical applications to resuscitating accidentally cooled subjects and should facilitate the use of hypothermia for anaesthesia in cardiac surgery and in other operations which are difficult to perform at higher temperatures (Juvenelle, Norberg, Lind, Bergstrand & Wegelius, 1953). There is already a good deal of experimental evidence which indicates that revival of adult animals with body temperatures approaching 0° C is not impossible even after comparatively long periods without respiration or heart beat. As long ago as 1917 Winterstein cooled guinea-pigs and rabbits until heart beat and breathing had stopped,

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and when their body temperatures reached 6–11° C he resuscitated them by centripetal intra-arterial injection of adrenaline in hot Ringer's solution, followed by artificial respiration. In every instance, however, the animals thus revived succumbed shortly afterwards. Similar treatment of a guinea-pig cooled to 0° C resulted in transitory restoration of heart beat only. More recently, Lutz (1950) found that a small proportion of guinea-pigs with body temperatures close to 0° C could be resuscitated by immersion in hot baths (about 45° C) to rewarm the whole body. Only 9% of the animals so treated survived for more than a few days.

During the last few years remarkable results have been obtained from dogs cooled by circulating their blood through a refrigerating system outside the body. Delorme (1952), who was one of the pioneers with this technique, cooled dogs to body temperatures between 22 and 26° C. Juvenelle and his colleagues succeeded in lowering the body temperature of dogs to 9° C by pumping the blood through a circuit of this kind and back into the cardio-vascular system. When the blood was rewarmed the body temperature rose and normal cardiac rhythm was restored in spite of ventricular fibrillation which, in some experiments, lasted several hours (Juvenelle, Lind & Wegelius, 1954). Gollan (1954) also used extra-corporeal cooling, oxygenation and rewarming of the blood, and revived dogs from body temperatures as low as 0° C and after cardiac arrest of 1 hr. It is important to note that, in these experiments on dogs, oxygenated blood was pumped continuously into the vascular system during the period of cardiac arrest or during ventricular fibrillation.

So far as we know, hypothermia has never been induced in rodents by cooling the blood stream in an external circuit, but only by exposing the intact body to cold air or cold water. Similarly, rewarming has usually been performed by transferring the cold animal to a warm environment. The possibility of re-establishing spontaneous heart beats and natural circulation in ice-cold rats without operative intervention, by heating the chest wall overlying the heart before rewarming the whole body, has already been investigated by one of us (Andjus, 1951*a*, *b*). Rats cooled to 1° C were kept with heart and breathing at a standstill and without natural or artificial circulation for periods up to 1½ hr. The heart beat was re-initiated by heating the praecordium locally, and artificial respiration was given. Spontaneous breathing was resumed in the majority of animals when the colonic temperature reached approximately 15° C and after the neck had been heated. Only then were the reanimated rats transferred to a warm bath. A high proportion of animals succumbed, either during the process of warming the whole body or within a few hours or days of regaining normal body temperature and reflex and voluntary activity; few survived for longer periods (Andjus, 1951*a*, *b*). Jaulmes and Richard have followed the technique of Andjus, but although they only cooled their rats to

4° C, all died within 48 hr of reanimation (see Laborit, 1953). Nevertheless, these results were encouraging because they showed that heart beat and breathing could be re-established and full recovery obtained, if only temporarily, without either circulating warm aerated blood artificially or raising the temperature of the whole body rapidly from 0° C. Rapid rewarming which was at one time thought to be essential (Weltz, Wendt & Ruppin, 1942) would, in practice, be difficult to achieve with larger animals or humans (Edholm, 1952).

The failures in reanimating rats and the high mortality after initial success might have resulted from irreversible damage during cooling. On the other hand, they might have been due to imperfections in the various procedures hitherto used for reanimation. We therefore set out to investigate alternative methods for applying heat locally to the cardiac area of hypothermic rats with body temperatures between 0 and 2° C and with arrested circulation and respiration. Recently we demonstrated a new technique (Andjus & Smith, 1954), by means of which the percentage of rats revived was much higher and the incidence of delayed deaths much lower than could have been obtained by previous methods.

The object of this paper is to describe in detail the modifications of Andjus's original method used by us and also the new technique and apparatus. The results of 112 experiments on different rats will be described, together with observations on fifty-eight survivors of these experiments which have been kept and observed for periods varying from 66 to 630 days.

METHODS

Animals

The rats were young adults of the Medical Research Council's hooded strain bred and reared at Mill Hill. They were fed on the stock diet, no. 41 (Bruce & Parkes, 1949). The animals were weighed immediately before the experiment and ranged between 90 and 250 g, the majority being between 120 and 200 g. Those which survived hypothermia were weighed daily for at least 2 weeks, then weekly for 4 weeks, and monthly thereafter. The oestrous cycles of females were studied by examining vaginal smears daily for 10 days before cooling, and, in survivors, for 4 successive weeks thereafter. They were then paired for 3 weeks with normal males. Four to six weeks after the experiment each resuscitated male was mated and left with three normal females for 3 weeks. The experimental animals were then separated and 4 weeks later those without progeny were remated to normal animals. Some of the fertile experimental males and females were mated together in pairs.

The food and water intakes of ten rats were measured daily for 2 weeks after revival from hypothermia. These animals had drinking bottles both of 1% NaCl and of water.

Apparatus

Closed vessels. Two types of glass vessel were used in the first stage of cooling. The 2½ l. capacity jars had metal screw-on lids and were not completely airtight. The 1½ l. Kilner jars were closed with a rubber washer and either a glass lid or the bakelite sampling tops previously described (Smith, Emmens & Parkes, 1947); the lid was secured by a metal screw-band and was effectively airtight. The closed vessels were put in a domestic refrigerator running at about +5° C.

Gas sampling and analysis. Samples of residual air were taken into Brodie sampling bottles and their O_2 and CO_2 content estimated by the method and apparatus of Scholander (Scholander,

1947). Oxygen consumption was measured by a slight modification of the method and apparatus described by Giaja (1953).

Receptacles for ice. In the second stage of cooling, rats were placed singly in melting ice in glass dishes (9 × 4 × 2 in.) surrounded by crushed ice in an enamel tray (12 × 18 × 5 in.).

Temperature recording. Body temperature was measured either by a mercury thermometer or by a thermocouple inserted into the colon so that its tip was 5 cm from the anus. The thermometers were graduated from 0 to +60° C, with the zero reading 8 cm from the tip. In some experiments a thermocouple was passed approximately 5 cm into the oesophagus so that its tip lay adjacent to the heart. In corpses intrathoracic temperature was recorded from a thermocouple inserted so that its tip lay anterior to the heart.

The thermocouples were of constantan-nichrome twin laid wire (36 s.w.g.) and were protected by narrow polythene tubing. They were connected in series to a reference thermocouple kept at a constant temperature of +38° C by a Sunvic cold junction thermostat (Type CJI). The e.m.f. developed was measured with a potentiometer using a spot galvanometer as a null-point indicator.

Artificial respiration. During resuscitation, air was insufflated into the lungs by means of single-hand bellows, through a rubber tube 1 cm in diameter, applied to the nostrils.

Apparatus for local rewarming

The metal spatula consisted of a steel plate (2·8 × 2·4 cm and 0·8 mm thick) brazed on to a steel handle.

The source of the beam of light was a 115 V/300 W tubular projector lamp (Ediswan Type A1/37, single centre contact). Its peak emission was at 9000 Å. The voltage supply and intensity of light and heat emitted were adjusted by means of a variable transformer. The lamp was supported vertically so that its cap was 3 in. and its painted tip 7 in. above the operating platform. The tip projected through the central aperture of a parabolic lamp reflector, 5 in. diameter across the rim. This reflector brought the beam of light to a focus 1 in. above the platform (Fig. 1).

The operating platform consisted of a perforated zinc top (12 × 6 in.), mounted on a frame (Fig. 1).

A pear-shaped duralite shield (10 in. long, 4 in. max. width, 2 in. min. width) was supported 1½ in. above the platform. The shield was perforated by a circular aperture 2½ in. from the broad end. The aperture was so arranged that the beam of light passed through it (Fig. 1).

Apparatus for general rewarming

An electrically heated thermostatically controlled water-bath (18 × 7 × 8 in.) was fitted with a perforated zinc shelf on which the rat was placed supine with its abdomen and thorax submerged and its nose and mouth protruding above the water-level. A double-doored Hearson incubator was thermostatically controlled so that, with the glass door open a crack and the wooden door ajar, it maintained an internal temperature of 28–32° C.

Warm cupboard for convalescent animals

Two shelves of an animal house rack were encased in insulating boards (½ in. thick) leaving a 2 in. gap for air entry along the entire length of the base. Six doors were fitted in front and a pair of 2 ft. tubular heaters (40 W each) and a Sunvic air thermostat were installed. A domestic fan geared to run at low speed was placed at one end separated from the heaters by an aluminium baffle. The well-ventilated cupboard thus formed held twelve cages for five rats at an air temperature of 29° C (±3°).

Procedures

Observations on heart beat and respiration

The cardiac impulse transmitted to the praecordium, the jugular pulse and the respiratory movements were observed visually. Results of electrocardiography will be reported elsewhere.

Arterial blood pressure

Arterial blood pressure was recorded in heparinized rats by means of a syringe needle or polythene cannula inserted into the carotid artery and connected to a mercury manometer.

Blood samples

Blood samples for chemical analysis were withdrawn from the jugular vein of animals in the second stage of surgical anaesthesia with body temperatures at or below 18° C. Blood sugar was estimated by the method of Hagedorn and Jensen, and inorganic phosphorus by the procedure described by King (1946). Blood counts were made on blood withdrawn from the tail.

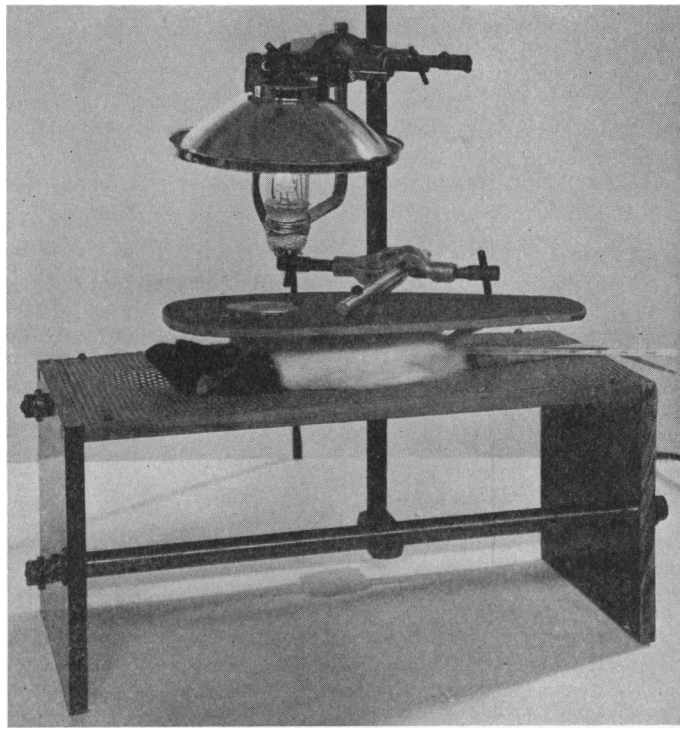

Fig. 1. The projection lamp and set-up for local rewarming.

Procedure for cooling and reanimation

First stage of cooling. The rats were enclosed singly in vessels and placed in the refrigerator (+5° C approx.) for 1½–2½ hr. They were examined at intervals without opening the jars. When they appeared flaccid and lethargic and approaching a state of anaesthesia samples of the residual air were taken for analysis. The animals were then removed from the vessels and examined. The rate and rhythm of heart beat and breathing were noted and reflexes were examined. The presence of a corneal reflex at this stage was regarded as essential in animals which were to be cooled further. A thermometer or thermocouple was inserted into the colon and the deep body temperature recorded. The rats were thoroughly wetted with ice-cold water to expel the insulating layer of air enmeshed in the fur. They were then immersed in dishes of melting ice and buried under crushed ice so that only the nostrils, the tail and the graduated part of the thermometer protruded. Unless otherwise stated, they were left under ice for exactly 1 hr from the time the colonic temperature reached 15° C and for approximately 40 min after it had reached 6° C.

Reanimation. Stage I: local rewarming of the cardiac area of the chest. The animals were taken out

of the ice with body temperatures ranging between 0 and 1·8° C. The praecordium was then rewarmed by one of the following methods:

(*a*) *Application of the hot metal spatula.* The spatula was heated in a Bunsen flame for about 30 sec, or until sufficiently hot to make steam rise from a moist surface. It was applied to the left side of the chest wall anterior to the heart. The fur on the heated area was kept wet. At first the spatula was applied as often as 20 times per min. As soon as a heart beat was seen air was insufflated into the lungs at the approximate rate of five puffs every 30 sec. Artificial respiration was given in such a way that the chest wall rose and fell slightly more than in normal respiration and so that a small volume of air entered the stomach. Local rewarming was continued when not insufflating. When a regular cardiac rhythm had been re-established the hot spatula was applied less frequently. Heating was discontinued when the heart rate was increasing spontaneously.

TABLE 1A. Procedure for rewarming with the beam of light

Duration of exposure (min)	Variac setting (V)	Site of heating	Notes
1	85	Praecordium	—
2	55	Praecordium	—
1	75	Praecordium	—
1	50	Praecordium	—
2	30	Praecordium	—
1	50	Praecordium	—
2	30	Praecordium	Remove shield. Continue until colonic temperature reaches 11·5° C
1	50	Front of neck	—
1	50	Back of neck	—
1	50	Front of neck	—
	30	Praecordium	Continue heating the praecordium until colonic temperature reaches 15° C and respiration is regular

TABLE 1B. Modified procedure for rewarming with the beam of light

Duration of exposure (min)	Variac setting (V)	Site of heating	Notes
1	90	Praecordium	—
3	60	Praecordium	—
	50	Praecordium	Continue heating the praecordium until colonic temperature reaches 10° C
1	50	Front of neck	—
1	50	Back of neck	—
1	50	Front of neck	—
	30	Praecordium	Continue heating the praecordium until colonic temperature reaches 15° C and respiration is regular

The frequency of insufflation was then increased to 30 to 60 per min. When the colonic temperature had reached 10–11° C the neck was heated for about 1 min under the hot tap with water at 40–45° C. Artificial respiration was resumed until the animal took a spontaneous breath and was continued until the breathing was regular in rhythm and increasing in rate.

(*b*) *Focusing a beam of light.* The rats were placed supine on the operating platform under the duralite shield and arranged so that the beam of light passing through the aperture was focused on to the praecordium. The intensity of the light and heat was controlled by altering the variac setting (see Table 1A, B). Air was continuously insufflated into the lungs at the rate of 30 to 60 puffs per min with occasional 15 sec intervals to observe and count heart beats and to remoisten the fur. When the colonic temperature reached 10–12° C the position of the animal was shifted and the neck was heated front and back under the light beam. Artificial respiration and local heating of the chest was then resumed until the rats breathed spontaneously. Insufflation was discontinued when the rhythm of natural breathing was regular and the rate steadily increasing.

Reanimation. Stage II: Rewarming the whole body. The rats were laid supine in the water-bath at +37° C with the head supported. Artificial respiration was administered if the breathing flagged and the animals were supervised to prevent them from shifting their position and drowning. As soon as they could turn over and maintain normal posture they were dried and transferred to the incubator at +28–32° C. 1–6 hr later they were moved to the warm cupboard in the animal house to convalesce for the next 3–4 days.

RESULTS

Observations during the first stage of cooling

The rats enclosed in vessels at 5° C moved around actively for 30–60 min, then settled quietly. After 90–150 min, depending on the size of the jar, they became flaccid and lethargic with colonic temperatures at or below 20° C.

TABLE 2. White blood cell counts of rats with normal body temperatures and at the end of stage I of cooling

	Total no. leucocytes per mm³ Body temperature	
Rat no.	Normal	15–18° C
1	9,000	1,000
2	9,000	2,200
3	11,000	3,000
4	5,600	800
5	7,000	3,200
6	5,400	2,200
7	9,600	400
8	6,600	200
9	8,400	1,000

TABLE 3. Analysis of residual air in closed vessels at the end of stage I of cooling

Rat no.	Body weight (g)	Colonic temperature (°C)	% O_2	% CO_2
GA 5	136	20·7	2·8	16·4
GA 7	148	20·3	3·4	15·6
GA 8	156	19·5	2·7	16·6
GA 9	150	20·4	2·2	16·0
GA 10	156	19·8	3·0	16·6
GA 11	161	18·5	2·4	16·3

The oxygen consumption of three rats with temperatures between 18 and 20° C was measured. It had fallen below 600 c.c./kg/hr. The blood-sugar concentration of thirty hypothermic rats was determined towards the end of the first stage of cooling. It had risen to between 142 and 356 mg% with a mean value of 255 mg%. Plasma inorganic phosphorus was raised to a mean value of 12·5 mg%. Blood counts on nine animals showed a leucopenia (Table 2). These findings confirmed results previously obtained in Belgrade on albino rats cooled in this way (Stefanović, 1952).

Twelve animals were enclosed in airtight vessels (capacity 1½ l.). Samples of residual air were taken after 90–100 min when the animals had become limp and unconscious, but were breathing deeply. Analysis showed that the oxygen

content had fallen to between 2·2 and 3·4% and that the carbon dioxide content had risen to between 15·6 and 16·6% (see Table 3). Six of these animals were left at room temperature after removal from the closed vessels; they all recovered without treatment of any kind. The other six were transferred from the jar to icy water and cooled to body temperatures between 0·5 and 1·5° C; when subsequently rewarmed by the beam of light, three revived completely (*vide infra*, pp. 465, 466).

When removed from the closed vessels the animals with body temperatures at or below 20·5° C were in the second stage of surgical anaesthesia with deep regular respiration and active corneal reflexes. A stable state of hypothermic anaesthesia was, however, not maintained unless they were cooled to and kept at body temperatures between 15 and 18° C. In this range anoxia and hypercapnia were no longer necessary or desirable and rats, whether they were to be cooled further, or kept at 15° C, or rewarmed at once, were all allowed to breathe atmospheric air. Rats cooled to body temperatures as low as 15° C and then left at an ambient temperature of 20° C rewarmed spontaneously and regained reflexes and consciousness (see Fig. 8B and p. 458). They suffered no apparent after-effects.

The majority of animals were studied during chilling to and revival from lower temperatures.

Observations during the second stage of cooling

The colonic temperature of rats transferred from closed vessels to icy water reached 15° C within approximately 5 min.

Below 15° C exponential curves were obtained by plotting time against colonic temperature (see Fig. 2). In typical experiments the colonic temperature passed from 15 to 6° C in 15–25 min, and from 6° to about 1° C during the subsequent 35 to 45 min.

Regular respiration generally ceased within a few minutes of covering with melting ice. Occasionally isolated gasps were given by animals with body temperatures as low as 10° C (cf. Andjus, 1953).

The arterial blood pressure of five rats was recorded during the second stage of cooling. It showed a definite rise of about 10 mm Hg when the rats were immersed in icy water and a fall within the next 2–5 min from approximately 80 mm to below 10 mm Hg. The pulse pressure was relatively high and the last irregular heart beats were well seen in the tracings (Fig. 3).

The heart rate slowed suddenly at body temperatures between 10 and 13° C and the last beat was usually recorded at or above 8° C. Rarely, isolated or coupled beats separated by intervals of 1–2 min were observed or recorded at colonic temperatures as low as 5–6° C, 15–30 min after immersion in ice. Loss of sinus rhythm, extrasystoles, heart block and other abnormalities have been recorded and are reported elsewhere (Donzelot, Milovanovich & Andjus, 1953).

The majority of the rats cooled by the method described were without respiration for 60 min and without apparent heart beats for 40–50 min.

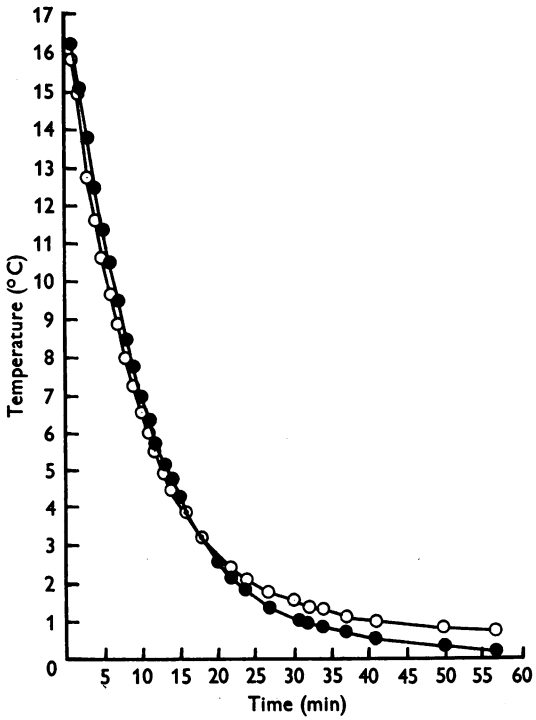

Fig. 2. The fall in thoracic and abdominal temperature of a rat in stage II of cooling, recorded from thermocouples. ●, Colonic temperature; ○, oesophageal temperature.

Observations during local heating of the cardiac area

The rate of rise of intrathoracic and colonic temperatures was first studied in four dead rats which had been cooled to 0° in ice. When the praecordium was heated intermittently with the spatula the temperature recorded from the thermocouple on the anterior surface of the heart reached 6° C in 1 min, 13° C in 2 min, and 25° C in 4 min. The maximum temperature reached in 10 min was 28° C (Fig. 5).

When the praecordium of ice-cold corpses was heated by the beam of light, following the procedure shown in Table 1 A, the temperature on the surface of the heart rose even more steeply than in the previous experiment so that 13·5° C was reached in 1 min, 24° C in 2 min, and 39° C in 5 min (Fig. 5).

There was little difference in the rate of rise of colonic temperatures in corpses heated by the two methods (Fig. 5).

The intrathoracic temperature of six experimental animals was recorded from a thermocouple in the oesophagus during local heating with the beam of

light. A typical record is shown in Fig. 6. The temperature rose gradually at first, reaching 3° C after 3 min heating. It then rose steeply, reaching 11° C during the next 3 min. Thereafter the oesophageal temperature rose gradually,

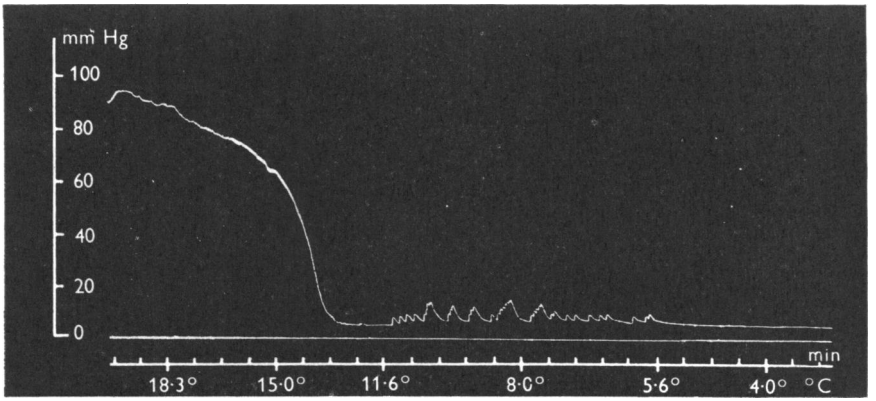

Fig. 3. Kymograph tracing of arterial blood pressure during stage II of cooling. The corresponding colonic temperatures in °C are written below the time scale.

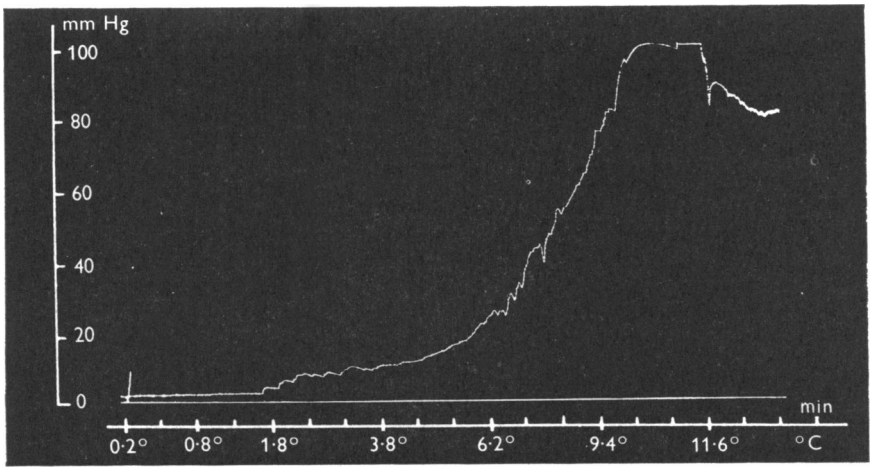

Fig. 4. Kymograph tracing of arterial blood pressure during rewarming with the beam of light. The corresponding colonic temperatures in °C are written below the time scale.

and after 25 min heating had reached 21° C. The temperature in the colon rose less rapidly then in the oesophagus (Fig. 6). The rate of rise increased suddenly after the 11th min presumably because of restoration of a circulation of warm blood.

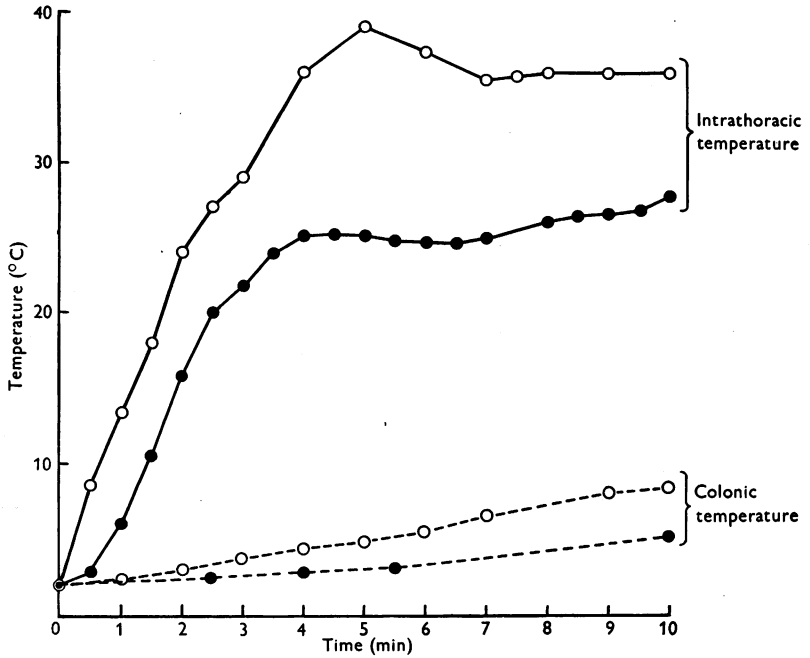

Fig. 5. The rise in intrathoracic and colonic temperature of rat corpses during local heating of the praecordium with the spatula (●) and with the beam of light (○).

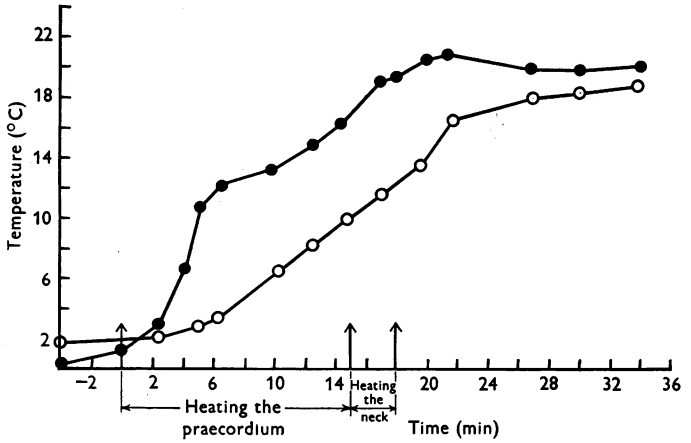

Fig. 6. The rise in intrathoracic and colonic temperature of a living rat during local heating of the praecordium with the beam of light. ●, Intrathoracic; ○, colonic.

The experiments on corpses and on rats during reanimation are not comparable because the recording thermocouples were differently situated in the thorax. The thermocouple inserted through the chest wall of corpses lay anterior to the heart and registered a maximum intrathoracic temperature during the local heating of the praecordium. The thermocouple in the oesophagus of the experimental animals lay behind the heart at a depth too great to be penetrated by the light and heat impinging on the chest wall. It probably recorded the minimum intrathoracic temperature.

The first cardiac impulse transmitted to the chest wall was generally seen between 2 and 10 min after local application of the spatula had started (Fig. 7). Heart beats not sufficiently forceful to move the praecordium may, of course, have occurred sooner. In rats heated by the beam of light the position of the

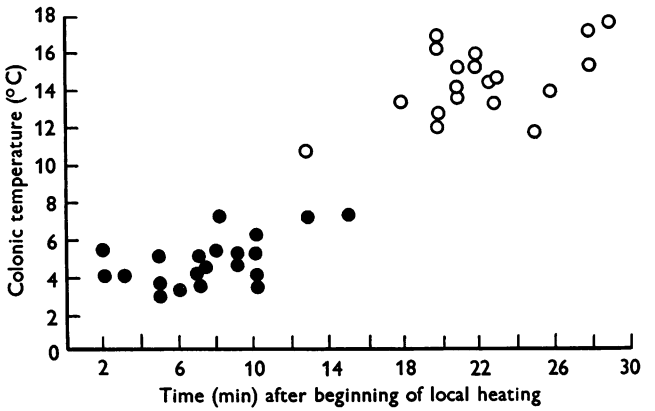

Fig. 7. The time and temperature at which the first visible cardiac impulse and the first spontaneous breath occurred in twenty-one different rats cooled to 0–2° C and reanimated by local warming of the praecordium with the metal spatula. ●, first heart beat seen; ○, first spontaneous breath.

shield prevented a good view of the left side of the chest. By the time the shield was moved aside at the end of 10 min (Table 1) the heart was usually beating steadily 30–70 times per min and the jugular pulse could often be seen.

Spontaneous breathing was imminent when artificial respiration elicited weak reflex responses of the thoracic musculature and when there was slight resistance to insufflation of air and a tendency to maintain the expiratory position.

There was no difficulty in seeing the animal take its first breath, which was generally between 18 and 30 min after the start of resuscitation, and when the body temperature lay between 12 and 17° C (Fig. 7). Rats heated under the lamp tended to take their first breath at a slightly lower temperature and to resume a regular respiratory rhythm and to be independent of artificial respiration sooner than those heated with the spatula.

The rise in arterial blood pressure during rewarming the cardiac area with the beam of light was studied in five animals. A typical tracing is reproduced in Fig. 4. The first isolated heart beats produced visible pulsations on the tracing, but had little influence on the blood-pressure level, which showed no change during the first 4 min of rewarming. Between the 8th and 14th min the heart rate increased and the pressure rose steeply to 100 mm Hg; it fell gradually to 80 mm, and maintained this level from the 18th min onwards.

At the end of the first stage of reanimation by either method rats were still limp and unconscious, with dilated pupils and without corneal or other reflexes. They were breathing spontaneously but not with a regular rhythm.

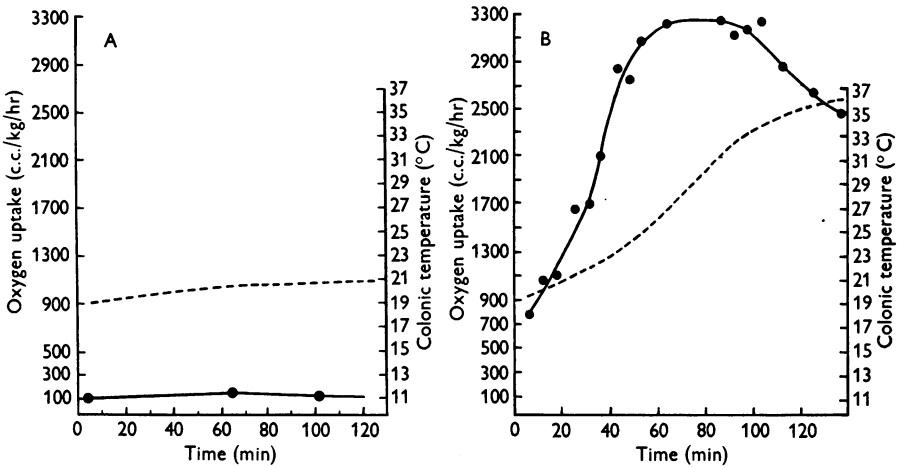

Fig. 8. The oxygen consumption and colonic temperature of two rats left at room temperature (+20° C). (A) Previously cooled to +2° C and rewarmed to 15° C; (B) previously cooled to a colonic temperature of 15° C. ●, oxygen consumption; – – – –, colonic temperature.

The heart rate was between 60 and 100 beats per min, and the colonic temperature was 15–20° C. In this state, they were still incapable of spontaneous recovery and, if left unattended on the bench, their body temperature failed to rise above the ambient temperature, and their rate of oxygen consumption remained at the low level of 100–200 c.c./kg/hr (see Fig. 8A). For this reason further active treatment was necessary and was carried out by rewarming the whole body by immersion in a water-bath at 37° C.

By contrast, rats previously cooled to colonic temperatures not lower than 15° C recovered without treatment when left at room temperature. Their oxygen consumption and body temperature rose spontaneously. Characteristic curves are given in Fig. 8B, and show that the oxygen consumption reached a maximum at a body temperature slightly above 30° C and then decreased to the normal level.

Observations during the second stage of rewarming

Respiration usually became regular soon after immersion in the warm bath when the body temperature had risen to about 25° C. Thereafter the rate of respiration increased considerably, and above 30° C was sometimes faster than normal.

The pupils constricted when the animals had rewarmed to 20° C or above. The corneal reflex could be elicited again at body temperatures ranging from 28 to 35° C.

The forelimbs recovered sensitivity, muscle tone and response to direct and indirect stimuli sooner than the hindlimbs. The first sign of recovery in the back legs was when pinching the feet caused reflex changes in respiration and reflex movements of head, jaws and front legs. The hindlimbs remained flaccid and motionless for many minutes thereafter, although there were spontaneous movements of the jaws, eyelids and head.

Righting reflexes were not regained until the body temperature was over 30° C. The rats were then removed from the bath, dried and transferred to the incubator at 28–32° C. At first they had difficulty in maintaining normal posture and crawled around, staggering, tumbling and righting themselves. Later they sat motionless in a normal attitude. Many animals at this stage showed signs of thirst and drank when water or saline was presented to them, but they seldom moved in search of a container. It was rare for them to take food during the first 2–3 hr after transfer to the incubator. During the same period the body temperature continued to rise until the normal level was reached. The animals were not at first able to maintain a normal body temperature at ambient temperatures below 28° C. They were therefore transferred to the warm cupboard in the animal house to convalesce until normal thermo-regulation was regained.

Revival of hypothermic rats by the spatula technique

Table 4 summarizes the fate of twenty-five rats which had been cooled as described to body temperatures between 0·8 and 1·8° C and were treated by heating the praecordium with the hot metal spatula and by giving artificial respiration.

The heart resumed beating in all the animals, but in four a regular rhythm was not established and the heart stopped beating within 10–30 min. In four other animals a regular cardiac rhythm was resumed for a short period but they died during the first stage of rewarming without taking a breath.

Seventeen of the twenty-five rats started to breathe spontaneously. Three of these failed to regain a regular respiratory rhythm. Six animals which had been breathing regularly died during the second stage of rewarming in the water-bath. The corneal reflex had been elicited from four of these, and skin

sensitivity and muscular activity was demonstrated in three before they succumbed.

Eleven rats regained righting reflexes, but one died within an hour of transfer to the incubator. Five others survived more than 4 hr but less than 18 hr, and were found dead in the warm cupboard next morning. Only five of the twenty-five rats in this series survived more than 24 hr. All these animals were observed for 66 days or longer after reanimation. After the first 2 days of convalescence they appeared to be in good condition apart from second degree burns on the praecordium. Their growth and breeding performance are described below.

TABLE 4. Revival of rats with colonic temperatures 0·8–1·8° C by the spatula technique

Series RWJ*	
Total number of rats	25
Extent of reanimation:	
No signs of life	0
Irregular heart beats for <30 min	4
Regular heart beats. No spontaneous breathing. Dead within 1 hr	4
Spontaneous breathing. No reflexes. Died in the 37° C bath within 2 hr	6
Reflexes recovered. Died within 1 hr in the incubator	1
Apparently complete recovery. Dead within 24 hr	5
Survived more than 66 days	5

* The rats in this series ranged in weight from 130 to 190 g on the day of the experiment.

These results were obtained by an inexperienced operator (A. U. S.) after collaborating in twelve experiments. The results were similar to those of an experienced worker (R. A.), using the same technique to reanimate rats in Belgrade and at Mill Hill.

Revival of hypothermic rats by the beam of light

The beam of light from a projection lamp was next tested as a means of heating the praecordium. In preliminary trials, twelve rats with body temperatures between 0 and 2° C were used. Success limited to restoration of heart beat resulted from discontinuous periods of intense illumination or from gentle continuous heating. The thirteenth rat was successfully reanimated by the scheme shown in Table 1A. A series of twenty-five rats which had been kept, as before, with body temperatures below 15° C for 1 hr and until 0·6–1·2° C was reached were then rewarmed in this way. The results are given in Table 5. In two rats a cardiac impulse was not seen, but may have been obscured by the shield. In seven others the cardiac rhythm never became regular and ceased within 30 min. In a tenth animal the rhythm became regular for 10 min before flagging and failing. The other fifteen animals all regained spontaneous regular respiration, muscle tone, reflexes and consciousness. Of these, four survived at least 5 hr but succumbed overnight, whereas eleven survived and have been studied for 66 days or longer after reanimation.

All these animals had local burns on the chest, which healed in about 3 weeks. The proportion of long-term survivors in this experiment was 44%, as compared with 20% obtained by local heating with the spatula.

The amount of illumination was then increased and a second scheme was followed (see Table 1B). The results are shown in Table 6. In two of the twenty-five animals, heart beats were not seen. Three regained a regular cardiac rhythm but never breathed spontaneously. The remaining twenty

TABLE 5. Revival of rats with body temperatures 0·6–1·2° C by the beam of light (Procedure as in Table 1A)

Series REL*	
Total number of rats	25
Extent of reanimation:	
No signs of life	2
Irregular heart beats for <30 min	7
Regular heart beats. No spontaneous breathing. Dead within 1 hr	1
Spontaneous breathing. No reflexes. Died in the 37° C bath	0
Apparently complete recovery. Died within 24 hr	4
Survived more than 66 days	11

* The rats in this experiment ranged in weight from 127 to 153 g on the day of the experiment.

TABLE 6. Revival of rats with body temperatures 0·6 to 2° C by the beam of light (Procedure as in Table 1B)

Series RA*	
Total number of rats	25
Extent of reanimation:	
No signs of life	2
Irregular heart beats for <30 min	3
Regular heart beats. No spontaneous breathing. Dead within 1 hr	0
Spontaneous breathing. No reflexes. Died in the 37° C bath	1
Apparently complete recovery. Dead within 24 hr	0
Died between the 4th and 10th day	2
Survived more than 66 days	17

* The rats in this experiment ranged in weight from 180 to 150 g on the day of the experiment.

resumed regular breathing, and nineteen regained reflex and spontaneous muscular activity. After 24 hr there were nineteen survivors, but on the 4th and 9th days respectively there was a casualty. The seventeen remaining rats in this series have been kept and studied for 100 days or more since reanimation. In this series the modified method of warming the praecordium with the beam of light resulted in full revival of 76% of the rats and long-term survival of 68%. In subsequent experiments a survival rate of 75% has been maintained. Delayed deaths were rare in spite of burns on the chest.

Post-mortem studies

Rats were abandoned during attempted reanimation if no heart beat had been seen by the time the body temperature had reached 11·0° C or if at any time the cardiac impulse was not seen for 10 min. Autopsies were then carried out immediately. Rats which succumbed before a regular cardiac rhythm had been established or before spontaneous breathing was resumed often showed

no special features at autopsy. Occasionally the ventricles were still beating weakly though at a slower rate than the auricles. In four of the casualties in the REL series (Table 5) the ventricles appeared to be fibrillating. In other animals, the auricles were beating but not the ventricles, and in these the venae cavae were distended. Other animals including another four from Table 5, had froth and mucus in the nostrils, larynx, trachea and bronchi.

Animals which died during rewarming the whole body in the water-bath did not appear to have heart-block or ventricular fibrillation. In some instances the lungs were congested with small superficial haemorrhagic patches. The stomach was often distended with air. The invariable and salient feature was haemorrhage into the duodenum, jejunum and ileum. Histological sections showed that the mucous membrane was undergoing necrosis.

The rats which died overnight showed no definite macroscopic lesions at post-mortem. Histological studies showed generalized venous and capillary engorgement, particularly in the lungs where there was also oedema, thickening of the alveolar walls and patchy collapse of alveoli. Vascular congestion was also conspicuous in the adrenals and the liver in which the parenchyma showed advanced cloudy swelling. These appearances were consistent with a diagnosis of death from acute heart failure.

Observations on long-term survivors

The thirty-two survivors from the experiments described above (see Tables 4–6) and twenty-five other rats reanimated from body temperatures below 2° C were kept in apparent good health for 66–130 days after the experiment; one rat has been kept for 630 days. There seems no reason to suppose that their normal life span will be curtailed.

Rate of growth after reanimation

All rats reanimated from body temperatures below 2° C lost weight for the first 24 hr after the experiment, and about 50 % continued to do so for 2–5 days. The total weight loss varied from 6 to 23 g, and averaged 12·8 g. The majority of survivors had regained their original weight by the 6th to the 8th day after cooling, and thereafter gained weight at a normal rate. A typical growth curve is shown in Fig. 9. One rat continued to lose weight for 12 days and did not return to its original weight until the 28th day. Its subsequent growth rate was retarded. This exceptional growth curve is also shown in Fig. 9. Rats cooled to body temperatures not lower than 15° C, and allowed to rewarm spontaneously, lost a small amount of weight (2·5 g approx.) during the first 24 hr, and then resumed growth at a normal rate (see Fig. 9).

Food and water intake after reanimation

The food and water intakes were reduced for the first 24 hr after reanimation. They returned to the normal level or above between the 2nd and the 5th days, generally the day before a gain in weight was recorded. During the first week some animals drank very little saline (0·8% NaCl), but thereafter they drank saline in preference to water. Some results are given in Table 7.

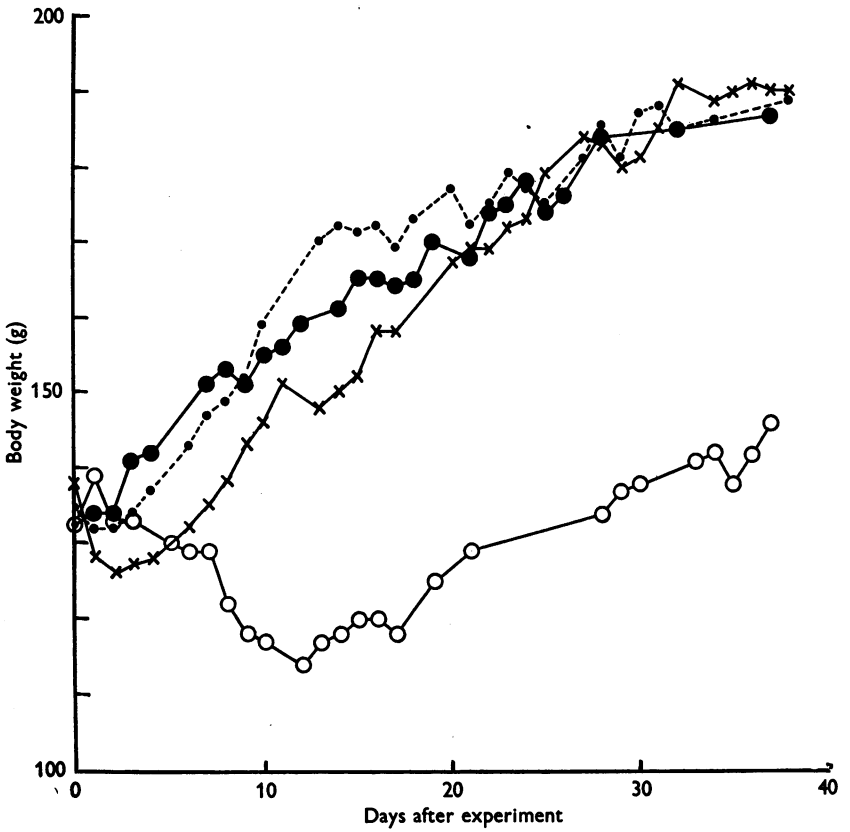

Fig. 9. Growth curves of rats. ●—●, Untreated control; •----•, after cooling to and rewarming from 15° C colonic temperature; ×—×, after cooling to and rewarming from 0° C colonic temperature, a typical curve; ○—○, after cooling to and rewarming from 0° C colonic temperature, an unusual curve.

Oestrous cycles of female rats after reanimation

The vaginal smears of eight females were studied for 1 month following the experiment. In three animals there was no disturbance of the normal 4–5 day oestrous cycle. In four others cyclic changes were suppressed for 13–17 days and a normal rhythm was resumed thereafter. One rat, which suffered from an intercurrent infection, never regained normal oestrous cycles (see Fig. 10).

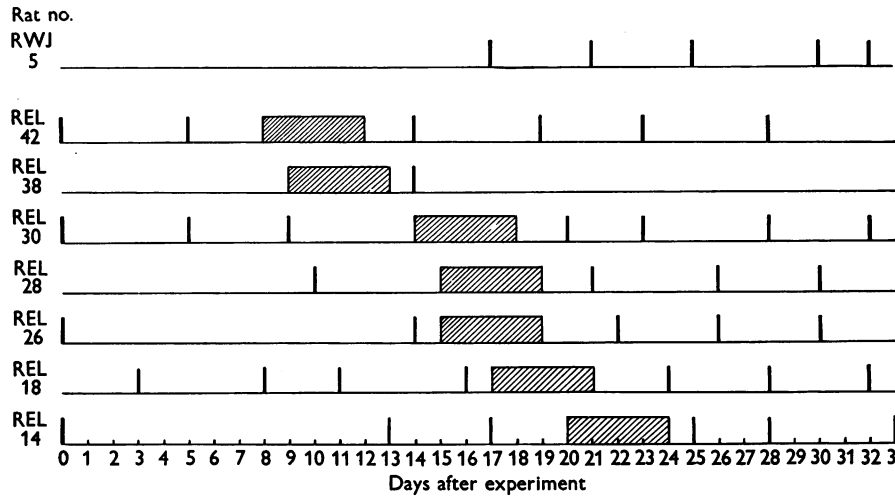

Fig. 10. The oestrous cycles of eight female rats revived after cooling to body temperatures between 0 and 2° C. Results of vaginal smears. I, Represents oestrus (smear composed of cornified cells); ▨, the Christmas holiday when vaginal smears were not taken.

TABLE 7. A specimen chart of food and water intakes of a rat after reanimation from a body temperature of 0·8° C by the spatula technique. Its original weight was 144 g.

Time since reanimation (days)	Body weight (g)	Food intake (g)	Water intake (ml.)	0·8% NaCl intake (ml.)	Total fluid (ml.)
1	132	4	12	2	14
2	126	6	14	0	14
3	—	6	15	2	17
4	126	7	16	2	18
5	124	18	12	1	13
6	125	—	14	1	15
7	127	13	11	1	12
8	131	13	10	14	24
9	—	12	6	12	18
10	—	12	6	12	18
11	138	12	5	12	17
12	139	13	10	7	17
13	145	11	3	19	22
14	145	13	6	23	29

Breeding performance of reanimated rats

Seven males mated at intervals after reanimation, were all fertile, but only three had progeny following the first mating 4–6 weeks after the experiment. Within 20 weeks they had fathered eighteen litters comprising 156 healthy young. In the fifteen litters examined there were 74 males and 74 females (Table 8). Five of the eight female survivors had litters within 7 weeks of the experiment, and of the 48 healthy young 26 males and 22 females were reared (Table 9). Two of the experimental females suffered from intercurrent infections and were destroyed before mating for a second or third time. These

results show that the majority of animals had progeny within 3 months of being cooled to, and reanimated from, body temperatures between 0·5 and 2° C. There is evidence however, of a temporary impairment of fertility.

TABLE 8. The breeding performance of male rats reanimated from body temperatures 0–2° C

Series and no. of rat	Interval between reanimation and mating (days)	No. of females	No. pregnant	No. of litters	Total no. of young	Sex of young		
						Male	Female	
RWJ 25	35	3	—	—	—	—	—	—
	61	3	—	—	—	—	—	—
	132	3	3	3	35	14	20	One died
RWJ 26	35	3	—	—	—	—	—	—
	61	3	1	1	10	Not recorded		—
RWJ 29	32	3	3	3	36	16	20	—
	58	3	2	2	6	3	3	One litter lost
REL 19	43	3	—	—	—	—	—	—
	94	3	3	3	23	12	11	—
REL 31	39	3	1	1	9	6	3	—
REL 39	34	3	1	Abortion	—	—	—	—
	85	3	2	2	22	12	10	—
REL 43	33	3	2	2	12	6	6	—
	91	1 (REL 14)	1	1	9	5	4	—

TABLE 9. The breeding performance of female rats reanimated from body temperatures 0–2° C

Series and no. of rat	Interval between reanimation and mating (days)	Whether pregnant	No. of young	Sex of young		
				Male	Female	
RWJ 5	48	Yes	7	2	5	—
REL 14	42	Yes	12	6	6	—
	103	Yes	9	5	4	—
REL 18	39	No	—	—	—	Killed because of intercurrent infection (otitis media)
REL 26	37	Yes	Abortion	—	—	—
	98	No	—	—	—	—
	140	Yes	Abortion	—	—	—
REL 28	37	Yes	11	8	3	—
REL 30	36	Yes	Litter eaten			
REL 38	31	No	—	—	—	Killed because of intercurrent infection
	92	No	—	—	—	
REL 42	30	Yes	9	5	4	—

Miscellaneous experiments

A number of experiments have been carried out in which the method of cooling was altered. There was no indication that more rapid induction of hypoxia and hypercapnia and more rapid cooling by enclosure in smaller vessels at +5° C altered the subsequent revival rate of rats cooled to body temperatures between 0·5 and 1·5° C. Six of the rats cooled in 1½ l. jars for

collection of samples of residual air (see p. 453) were subsequently cooled in ice and kept in the usual way with body temperatures below 15° C for 1 hr. They were then rewarmed by the beam of light following the technique shown in Table 1A. Three revived completely, and three others succumbed before regular respiratory rhythm and reflexes had been established.

In other experiments the animals were cooled in the second stage by circulating icy water around them so that the colonic temperature fell from 15° C to between 0·7 and 1·4° C in 35 min. They were then rewarmed by heating the cardiac area locally with the spatula. Of eight animals so treated the heart resumed beating in six and spontaneous breathing was recovered in five but,

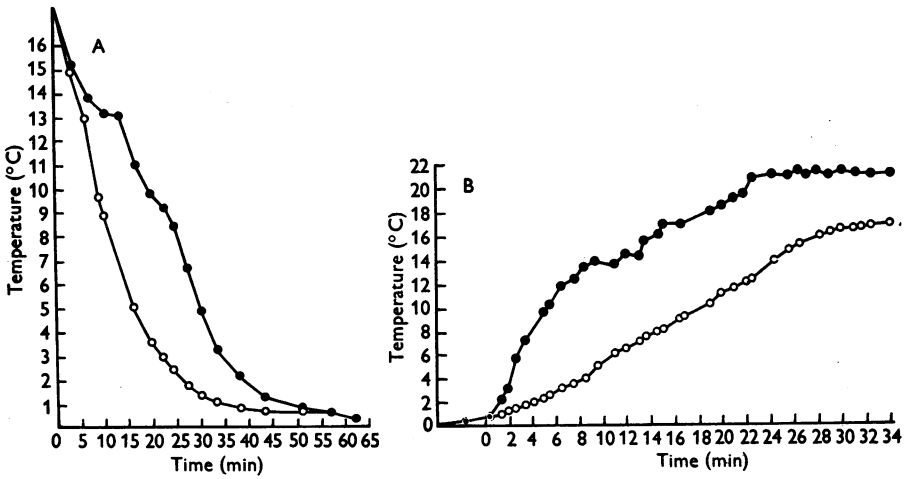

Fig. 11. (A) The fall in thoracic and abdominal temperature of a rat during stage II of cooling when the praecordium was kept warm under the beam of light. ●, Thoracic; ○, abdominal. (B) The rise in thoracic and abdominal temperature of the same rat during rewarming under the beam of light. ●, Thoracic; ○, abdominal.

of these, only four regained reflexes, and one died after apparently complete recovery. The other three revived completely and survived for many weeks, showing that more rapid cooling was not necessarily harmful. More rapid cooling combined with a more effective method of reanimation, has not so far been tested.

In a small series of experiments the praecordium of rats was kept warm by local heating with the beam of light, while the rest of the body was packed in ice during the second stage of cooling. The thoracic and abdominal temperatures were recorded from thermocouples in the oesophagus and colon. The heart was still beating when the thoracic temperature was 7° C and when the colonic temperature was as low as 2° C (see Fig. 11 A). Local heating was then discontinued, the chest was covered with ice and allowed to cool, the heart stopped beating and the animals were left for 40 min, during which time the

thoracic and colonic temperatures fell below 1° C. Nine rats so treated were then rewarmed by the beam of light, following the technique shown in Table 1 B. The rate of rise of temperature of an animal is shown in Fig. 11 B. Five of the rats revived completely and showed no unusual signs or symptoms to suggest that maintenance of a circulation when the hind quarters and limbs were intensely cold had been harmful.

Some preliminary trials were made of cooling under inhalation anaesthesia instead of in closed vessels. Two rats were cooled in icy water under ether anaesthesia until the body temperature reached 22° C. The anaesthetic was then discontinued and the rats breathed air for 10 min or more before respiration ceased at body temperatures approximately 15° C. They were then kept for 1 hr in ice before rewarming by the beam of light. One rat recovered completely but died next day with congested lungs; the other died after the heart had been beating for 10 min. Twelve rats enclosed in 2½ l. jars surrounded by ice were cooled under nitrous oxide anaesthesia. Anaesthesia was induced by passing pure nitrous oxide through the jars; oxygen was introduced after 1½ min, and the ratio of gas to oxygen was maintained at 4:1 for about 40 min. The gas flow was then gradually cut down until pure oxygen was being admitted. The animals remained unconscious, and were then taken out of the jar and packed into ice with colonic temperatures about 20° C. They were left in ice until the body temperature had been below 15° C for 1 hr and had reached 1° C. They were rewarmed under the beam of light following the scheme shown in Table 1 A. In every experiment the heart resumed beating. Eight rats died before breathing spontaneously, and in five of these the ventricles were fibrillating. Two rats resumed spontaneous breathing but died before regaining righting reflexes. Two animals recovered completely, one of which died within 3 hr of reanimation. Only one rat cooled under nitrous oxide anaesthesia survived indefinitely.

The effects of a variety of drugs have also been tested and the results will be reported later.

Other methods for rewarming the heart locally and for rewarming the whole body are still under investigation. The results of reheating the heart with a magnetron microwave generator are particularly promising and in different series 80–100% of the rats have been fully revived. The apparatus, technique and results will be described in full elsewhere (Andjus & Lovelock, 1955).

DISCUSSION

The results given amply confirm the earlier work of Andjus (1951*a*, *b*) and show clearly that cooling to body temperatures as low as 0·5° C is not necessarily lethal to the adult rat. The delayed deaths previously noted by Andjus (1951*a*, *b*) and by Jaulmes and Richard who repeated his work (see Laborit, 1953) seldom occurred when the heart was rewarmed locally by irradiation.

In three different experimental series using this technique, 44, 68 and 75% of the rats revived fully and survived for long periods in good health. 15° C can, therefore, no longer be accepted as the lethal body temperature for the adult rat.

The longest periods of circulatory arrest which adult mammals have hitherto survived are 15–30 min with body temperatures 20–30° C (Bigelow *et al.* 1950; Boerema, Wildschut, Schmidt & Broekhuysen, 1951; Swan *et al.* 1953; Juvenelle, *et al.* 1954; Jensen & Parkins, 1954). In our experiments, rats have recovered completely after 60–75 min at body temperatures below 15° C, and after 40–60 min without detectable heart beat, circulation, or respiration. The skulls of our animals were packed in ice and could have been little, if at all, warmer than the colon or the oesophagus from which temperatures as low as 0·5° C were recorded. Nevertheless, there was no evidence of the neurological damage described by Jensen & Parkins (1954) in dogs in which the brain temperature had fallen to between 8 and 12° C.

The tibial nerve of the rat no longer conducts impulses at temperatures below 9° C (Chatfield, Battista, Lyman & Garcia, 1948) and electroencephalograms show that electrical activity in the rat brain is suppressed at body temperatures below 18° C (Horsten, 1949; Lemaitre, 1954). In our animals, therefore, cerebral activity was probably arrested for 1½–2 hr. Nevertheless, they recovered reflexes, posture and consciousness within 2 hr, and apparently normal activity and behaviour within 2 days of rewarming from 0 to 1° C. The possibility of cerebral damage is under further investigation; precise psychological tests on animals before and after cooling are being carried out in collaboration with Prof. Russell of University College London, and will be described later.

It is at first sight surprising that previous workers have been unable to revive adult rats cooled below 15° C. There is evidence, however, that 15° C marks a turning point in the physiology of the rat. This is shown by their inability to rewarm or to increase their oxygen consumption spontaneously after cooling below 15° C even if rewarmed to that temperature before leaving them untreated at an ambient temperature of 20° C. It is likely that the method of reanimation is of great importance. When an animal with a deep body temperature of 0–2° C is transferred to a hot bath at +45° C as in the experiments of Lutz (1950) the skin and superficial tissues must rewarm rapidly and experience anoxia for many minutes before the heart is warm enough to beat and provide an adequate circulation. If, on the other hand, the heart is rewarmed first and a circulation established before the temperature of the bulk of the body rises, the degree and duration of tissue anoxia may be greatly reduced. It was remarkable that the revival rate in our experiments was increased from 20 to 75% when local heating of the surface of the chest wall was superseded by heating with a beam of light. The amount of heat

penetrating to the anterior surface of the heart was undoubtedly increased when the chest wall was irradiated, but the oesophageal thermocouple showed that the temperature of the posterior aspect of the heart lagged behind. These results suggested that a more efficient method for rewarming the heart rapidly should make it possible to revive all rats from body temperatures between 0 and 1° C. This suggestion was supported by finding that application of heat to the thorax by means of the magnetron microwave generator permitted revival of 90% of the rats in a preliminary trial, and 80–100% in subsequent series (Andjus & Lovelock, 1955). Direct application of heat to the heart has already been practised on hypothermic animals with body temperatures between 15 and 20° C. Crismon & Elliott (1947), for instance, showed that gentle heating of the sino-auricular node by means of a microheater in the oesophagus resulted in increased heart and respiratory rates and a rise in blood pressure in rats, but was ineffective in dogs. Boerema and his colleagues (1951), however, showed that irrigating the opened thorax with hot water at 40° C resulted in immediate improvement in cardiac action in hypothermic dogs which had previously showed signs of cardiac failure.

Our results indicate that the method of rewarming is of paramount importance. Nevertheless, the technique of cooling may well influence the survival rate even when optimum methods of rewarming are used. For instance, the preliminary trials of cooling rats under ether or nitrous oxide anaesthesia support Kayser & Hiebel's suggestion (1952) that administration of drugs or other agents may reduce the chances of revival from severe hypothermia and raise the lethal temperature level. There is evidence that, at reduced body temperatures, various drugs have increased toxicity (Adolph, 1948; Churchill-Davidson, McMillan, Melrose & Lynn, 1953). On the other hand, conscious animals plunged into cold water as in Adolph's experiments (1948) must be subjected to great psychological and physiological stress and may exhaust their adrenal glands and other protective mechanisms so that they inevitably succumb to cooling below 15° C. Oxygen lack during induction of hypothermia is usually regarded as dangerous because of the decreased dissociation of oxyhaemoglobin at reduced temperatures (Brown & Hill, 1923; von Wertz, 1954) and because myocardial anoxia predisposes to ventricular fibrillation (Lynn *et al.* 1954; Crismon, 1944). As a result, artificial respiration with high concentrations of oxygen is often favoured when cooling animals or humans for surgical anaesthesia. Hypoxia has, however, been used experimentally as an adjunct to cooling ever since Paul Bert showed that hypothermia could be induced in hibernating animals cooled under reduced oxygen tension in closed chambers in which the CO_2 was absorbed (Bert, 1870). Giaja (1940), for instance, has succeeded in inducing cold narcosis in rats, cats, dogs, and other non-hibernating animals by cooling them to body temperatures below 20° C under reduced barometric pressures. Kline (1947) showed that 13% CO_2 in

the inspired air increased the altitude tolerance of cats and thought that the raised CO_2 tension of the blood might increase pulmonary ventilation and facilitate dissociation of oxyhaemoglobin and might promote cerebral vasodilatation. As long ago as 1895 Dubois used a mixture of 45% CO_2, 12% O_2 and 43% air to cool marmots artificially. He called it 'le mélange qui endort'. His findings were confirmed by Benedict & Lee (1938). The closed vessel method of cooling, in which hypoxia and hypercapnia are combined, was introduced by Giaja & Andjus (1949) for producing surgical anaesthesia in rats, and has subsequently been used by them for anaesthetizing guinea-pigs, cats and dogs. It has now been adopted by Courrier & Marois (1953) and by Chauchard & Mazoué (1954) for cooling rats, while Bénitte (1954) follows the same principle and anaesthetizes dogs by cooling them and simultaneously administering gas mixtures high in CO_2 and low in O_2 content. We feel that the use of the closed vessel technique may, in part, be responsible for the resistance of our rats to body temperatures below 15° C. The revival rate of rats cooled to body temperatures of 0–1° C under the influence of drugs instead of narcotized by hypercapnia and hypoxia is still under investigation.

Several other problems are being studied. Hamsters have already been revived from subzero temperatures (Smith, Lovelock & Parkes, 1954), and similar experiments are being performed on rats. The maximum time for which rats can be kept with arrested circulation and respiration is being determined. The effect of repeated cooling and reanimation of individual rats is being examined. Continuous electrocardiographic studies during cooling and reanimation are being made. Finally, attempts are being made to revive larger and higher mammals from body temperatures approaching 0° C, after cessation of heart beat and breathing, and without the use of artificial extracorporeal circulatory systems.

SUMMARY

1. Rats narcotized previously by anoxia, hypercapnia and deep hypothermia were kept in icy water until their colonic temperatures had been below 15° C for 1 hr and had reached 0–2° C. They were without circulation or respiration for 40 min or longer.

2. These rats were resuscitated by heating the cardiac region locally until regular heart beats were resumed, and by insufflating air into the lungs until spontaneous respiration recurred.

3. The whole body was then rewarmed in a bath at +40° C until postural reflexes were regained. They were kept at an ambient temperature of 28° C for 4 days until thermal regulation was recovered and then at normal indoor temperatures.

4. Colonic and thoracic temperatures and arterial blood pressure were recorded and oxygen consumption and blood sugar concentration estimated in a number of animals during cooling and rewarming.

5. Local heating by means of a metal spatula effected reanimation of 20%, whereas irradiation with a beam of light focused on the praecordium resulted in revival of 75% of the refrigerated and inanimate rats.

6. Fifty-eight rats reanimated from 0 to 2° C were studied for periods varying from 66 to 630 days. Normal growth and behaviour were usually resumed 4 to 7 days after revival.

7. The seven males and five of the eight females tested for fertility had progeny within 3 months of reanimation.

We are indebted to Dr J. E. Lovelock who suggested the use of light for local heating, and to Mr C. Payne who arranged the lighting system. Our best thanks are due to Dr A. S. Parkes, F.R.S., for stimulation and encouragement.

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