

## Review article

# Apnea testing for the diagnosis of brain death

Lang CJG, Heckmann JG. Apnea testing for the diagnosis of brain death. Acta Neurol Scand 2005; 112: 358–369. © Blackwell Munksgaard 2005.

**Objectives** – A review is given on various methods, preconditions and pitfalls of apnea testing for the diagnosis of brain death. **Materials and methods** – An extensive medical data base search was implemented by information gathered from books and our own experience with more than 2000 apnea tests. **Results** – While testing for apnea (AT) is considered indispensable worldwide, recommendations and handling differ. Rather than relying on elapsed time, a specific target value for the partial arterial pressure of carbon dioxide (PaCO<sub>2</sub>) should be aimed at being the maximum physiological stimulus for respiration.

Methodological points are elaborated upon in detail for apneic oxygenation and hypoventilation. **Conclusion** – AT is an indispensable element of diagnosing brain death. Although with proper handling and adequate precautions AT is safe, it should be performed as a last resort. An international agreement on target values for the PaCO<sub>2</sub> is desirable.

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Apnea testing (AT) is a *conditio sine qua non* in determining brain death or brain stem death (1) worldwide (2, 3), although formal AT at a specific target PaCO<sub>2</sub> is required in only 59% of the countries (3). It is an important sign of loss of brain stem function and signifies that breath as an essential element of life has vanished from man. In the Bible, Genesis 2:7 reads as follows: 'The Lord God formed the man from the dust of the ground and breathed into his nostrils the breath of life, and the man became a living being.' AT is, however, the most time-consuming, difficult and potentially harmful of all clinical assessments. Hypotension of various degrees is a frequent and rather harmless concomitant (4, 5), but cardiac arrhythmia (6) and even asystole may become threatening (4, 7, 8). Disagreement prevails as to which parameters have to be applied and how to proceed for best performance. This paper reviews the preconditions and procedures of AT and addresses some special problems and pitfalls that have to be overcome in clinical practice to obtain valid results.

### Materials and methods

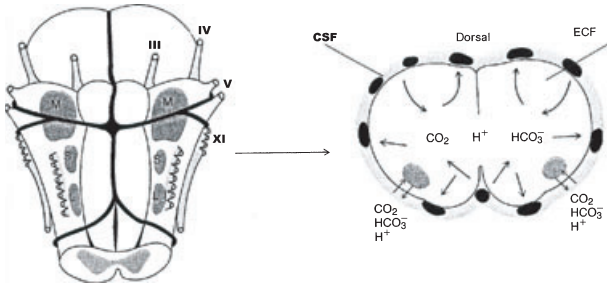
A review was made concerning pertinent articles worldwide on AT for diagnosing brain death using

computer-based scientific bibliographical data bases (MEDLINE, PubMed) and monographs. The key words brain, cerebral, death, apnea, apnoea, and test\* were used. After collecting data the papers were scrutinized for special problems such as hypotension, excessive hypercarbia, hypoxia, acidosis, cardiac arrhythmias or cardiac arrest, and pulmonary disorder and the respective bibliographies searched for any relevant additional publications. While not intending to be encyclopedic this paper tries to address the most important points with AT for brain death. The problems, proposals and results were evaluated against the background of our own experience of more than 2000 AT to date.

### Results

Physiology of respiration

Neurons for modulation and rhythmogenesis of automatic respiration are located in the pons and medulla oblongata. Chemoreceptors are seated in the ventral respiratory group (VRG), a longitudinal tissue column along the nucleus ambiguus. Neurons of the dorsal respiratory group (DRG) sitting in the ventral portions of the nucleus tractus

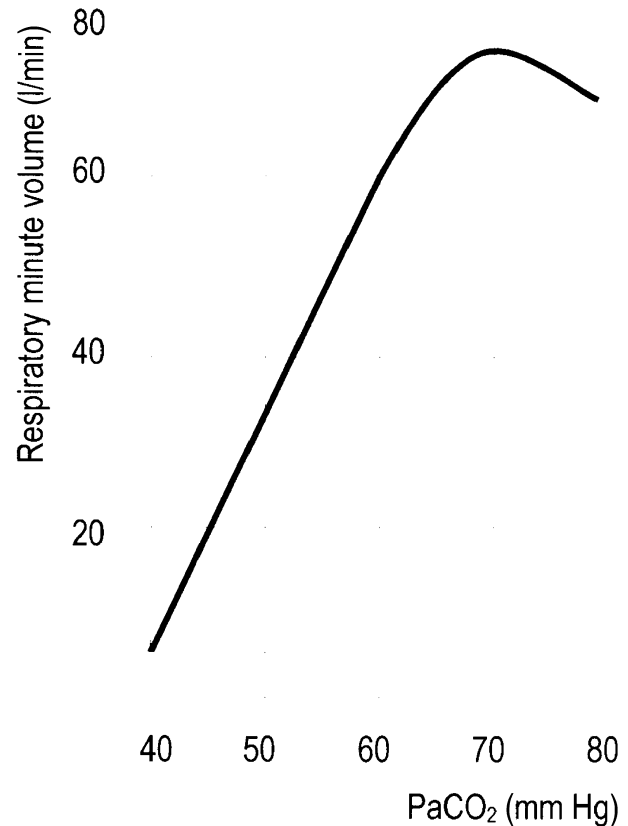


**Figure 1.** The brain stem respiratory centers and chemosensitive areas. On the left, a sagittal ventral view is given; on the right, a cross-section at the level of the arrow is shown. M is the rostral area (Mitchell), S the intermediate area (Schl afke), and L the caudal area (Loeschke). Cranial nerves are numbered in roman numerals. CSF, cerebrospinal fluid; ECF, extracellular fluid. The black dots on the right-hand side of the figure denote arteries and veins, the gray dots are chemosensitive areas, situated within the medulla oblongata about 100–400  $\mu\text{m}$  below its surface (10).

solitariae do not participate in rhythmogenesis (9) (Fig. 1) and are mainly inspiratory.

The pontine respiratory group modulates breathing frequency. Automatic breathing can be overridden by voluntary control. The most efficient respiratory stimulus is the partial arterial pressure of carbon dioxide ( $\text{PaCO}_2$ ) (Fig. 2), hence it is used for maximal stimulation in AT. The slope of the response curve reflects the sensitivity of respiratory regulation and amounts to approximately 2–3 l/min/mmHg.

Chemosensitive cells just below the ventral surface of the medulla oblongata are stimulated by a rise in  $\text{PaCO}_2$  more than by a drop in pH, as the blood–brain barrier is poorly permeable to ions such as  $\text{H}^+$  but not to  $\text{CO}_2$  (10). With preserved circulation, any change in respiration will promptly result in feedback to the lower brain stem. Respiratory changes of  $\text{PaCO}_2$  and pH are highly and inversely correlated. Under otherwise normal conditions the  $\text{CO}_2$  response curve rises up to a level of about 70 mmHg. When  $\text{PaCO}_2$  considerably exceeds this level a narcotic effect ensues and ventilation decreases. In wakeful persons a rise in  $\text{PaCO}_2$  causes dyspnea (air hunger) and an increase in respiratory frequency rather than in single respiratory volume. Different brain disorders cause different alterations of breathing: brain stem disorders may lead to rapid superficial breathing; hypoxia, hypercapnia or intoxications may yield deep breathing (Kussmaul type); with disturbed pontomedullary perfusion apneusis (abnormally prolonged inspiration) may be seen; with increased intracerebral pressure respiration may become irregular and aperiodic (Biot type); hypoxia or opiate intoxication may induce periodically increasing and decreasing respiratory movements



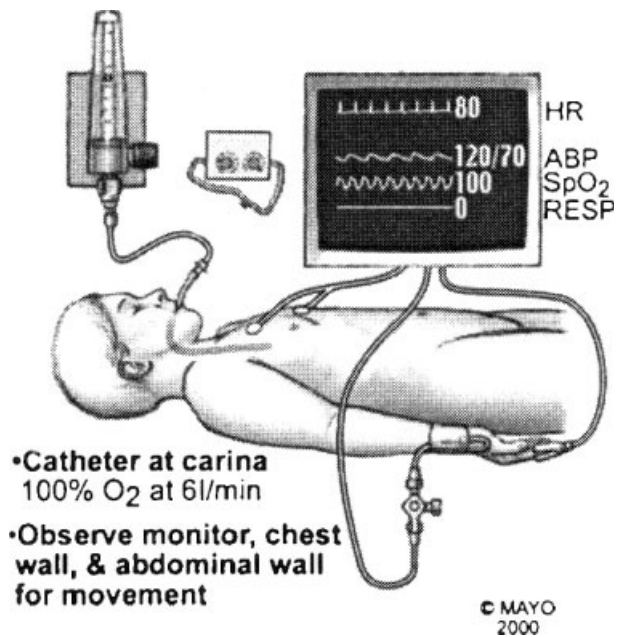
**Figure 2.** Correlation between arterial partial pressure of carbon dioxide ( $\text{PaCO}_2$ ) and respiratory minute volume. It increases up to levels of about 70 mmHg. It can be seen from the graph that during the linear phase an increase in  $\text{PaCO}_2$  of 10 mmHg corresponds to an increase in respiratory minute volume of about 30 l/min (11).

(Cheyne-Stokes type); and finally gasping followed by periods of apnea is seen with severe damage to the brain stem respiratory centers during agony before terminal apnea sets in (11).

#### Techniques of apnea testing

There are mainly two techniques for ascertaining sufficient oxygenation during AT: Apneic oxygenation and hypoventilation. In the first case the patient is disconnected from the respirator and receives pure oxygen at a rate between 4 and 10 l/min via a catheter (e.g. a 16 French suction catheter or cannula) that is inserted into the endotracheal tube down to the level of the carina (Fig. 3).

Gas convection ensures that the alveoles are ventilated enough to transport oxygen into the bloodstream even if there are no respiratory movements (13). In the second case the patient is not disconnected from the respirator but minute volume is reduced to a very low level (0.5–2 l/min) using for example (synchronized) intermittent



**Figure 3.** Apneic oxygenation. The patient is disconnected from the respirator while receiving pure oxygen via a catheter inserted into the endotracheal tube. HR, heart rate; ABP, arterial blood pressure; SpO<sub>2</sub>, oxygen concentration as measured by pulse oxymetry; RESP, respiration rate (12).

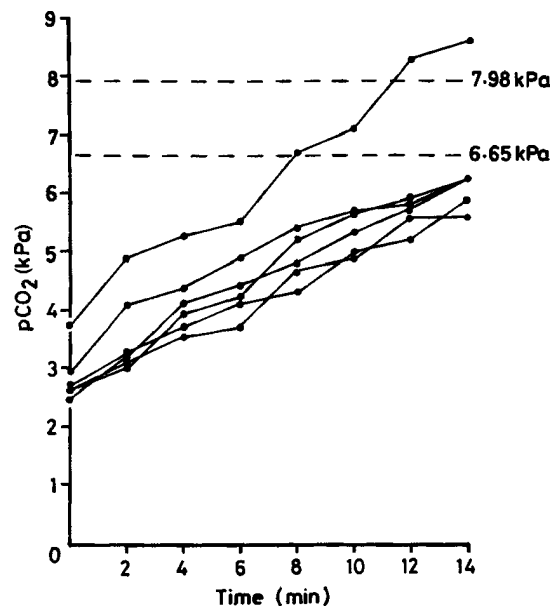
mandatory volume ventilation [(S)IMV] and pure oxygen for inspiration. The patient is not disconnected until the final phase when the required PaCO<sub>2</sub> is attained. This has the advantage of preventing tracheo-pulmonary complications and maintaining sufficient oxygenation at the same time with a high degree of probability while still allowing the examiner to detect any spontaneous breathing. This is our preferred means. A further modification has been described by Al Jumah et al. (14) using biphasic intermittent positive airway pressure (BIPAP). This method is also called bulk diffusion. Not disconnecting the patient from the respirator allows the maintenance of a continuous flow of oxygen and positive endexpiratory pressure (PEEP) (15, 16) without mechanical ventilation while the patient remains attached to the IMV circuit.

#### Requirements and preconditions

*Partial tension of oxygen* – There are no explicit recommendations for the partial arterial tension of oxygen (PaO<sub>2</sub>) except preoxygenation with 100% O<sub>2</sub> for some time, mostly for 10 min, and avoidance of hypoxia. Patients given room air during a 10-min apnea test (i.e. disconnected from the respirator) became hypoxic (8). The Quality Standards Subcommittee of the American Academy of

Neurology (17) recommends a normal PO<sub>2</sub> or preoxygenation to obtain an arterial PO<sub>2</sub> ≥200 mmHg. If the patient is disconnected without supplying oxygen to the trachea, the level may drop to dangerously low levels within minutes even after preoxygenation (18, 19). We have, however, seen one temporary PaO<sub>2</sub> level as low as 14 mmHg without serious harm to the cardiovascular system. If continuous or intermittent oxygen supply is preceded by denitrogenation of blood gases, high PaO<sub>2</sub> levels can be sustained for very long periods of time (13).

*Partial tension of carbon dioxide* – A normal arterial PCO<sub>2</sub> or PaCO<sub>2</sub> of ≥40 mmHg is recommended by the Quality Standards Subcommittee of the American Academy of Neurology (17) before AT. Belsh and Schiffman (20) recommend a starting PaCO<sub>2</sub> of 36 mmHg or higher. Telleria-Diaz (21) thought it possible to consider 20 mmHg above the starting PaCO<sub>2</sub> level. The requirements for the final PaCO<sub>2</sub> differ according to national guidelines. In many national guidelines a distinct PaCO<sub>2</sub> is prescribed for maximal stimulation of the respiratory centers. This is 50 mmHg = 6.7 kPa [e.g. in the UK (1, 22), Switzerland (23) and Portugal (24)] or 60 mmHg = 7.98 kPa [e.g. in the USA (25), Canada (26) and Germany (27)]. Both of these target levels are illustrated in Fig. 4. The claim of Wawersik (28) with reference to Belsh et al. (29) and Kaufmann



**Figure 4.** Rise of PaCO<sub>2</sub> during apnea testing. The dotted lines represent the levels of PaCO<sub>2</sub> that are required in the UK (lower line, 6.65 kPa = 50 mmHg) and the USA (upper line, 7.98 kPa = 60 mmHg). This figure illustrates that 14 min of apneic oxygenation may not suffice to reach the target value (31).

and Lynn (30) that recommended target values range from 44 to 90 mmHg is mistaken, because both sources adhere to the President's Commission suggesting a PaCO<sub>2</sub> of 60 mmHg.

During apnea the normal relationship of venous PCO<sub>2</sub> being greater than arterial PCO<sub>2</sub> is reversed (32). Based on empirical findings derived from physiological observations, a PaCO<sub>2</sub> of about 60–70 mmHg would seem ideal as there is a rather linear increase in respiratory minute volume up to this level and a decrease thereafter (33). While Ropper et al. (34) found that spontaneous breathing always began at CO<sub>2</sub> pressures lower than 40 mmHg, onset of respiration has been described with values of 47 and 54 mmHg (35) in adults and even higher values in children (see below). We ourselves have seen a 64-year-old man who started breathing at a PaCO<sub>2</sub> of 53.6 mmHg and a 6-year-old girl who started breathing at a PaCO<sub>2</sub> of 54.8 mmHg. Therefore, a target of 60 mmHg appears to be more appropriate than 50 mmHg.

*Hydrogen ion concentration* – There are no specific recommendations for the pH. Its drop is highly correlated with the rise of PaCO<sub>2</sub> and quickly restored with normoventilation or mild hyperventilation. We have seen uneventful acidosis as low as 6.808. If respiratory acidosis is corrected during AT – e.g. by bicarbonate solutions – alkalosis will ensue after normoventilation. Therefore, pH should not be corrected by buffer or alkaline solutions during AT; instead, a normal pH or a value in the low basic range should be ascertained at the onset of AT.

*Temperature* – Testing at body temperatures below 32°C (32.2°C, 36) is discouraged by most authors (19). In these cases the body must be warmed. Some authors recommend warming up to at least 36 or 36.5°C in every case (12, 17, 37, 38) which may be a very time-consuming measure but can shorten AT thereafter. Maekawa et al. (38) have given a formula describing the dependency of AT duration on body temperature ( $y = 0.54x - 15.2$ , where  $x$  is body temperature in °C and  $y$  is  $\Delta$ PaCO<sub>2</sub>). At any rate, correction of blood gas values for actual body temperature is deemed necessary (39).

*Duration of apnea testing* – Some authors have recommended 10 min, even if blood gas levels cannot be determined (34), others have considered 15 min as sufficient (40, 41). A duration of  $\leq 3$  min used by clinicians in the USA, as reported by Earnest et al. (42), was clearly insufficient.

However, fixed durations are not yielding reliable results (31), as the target value of 6.65 kPa (50 mmHg) or 7.98 kPa (60 mmHg) may not be reached and the necessary duration cannot be firmly predicted assuming a predetermined linear rise of the pCO<sub>2</sub>, e.g. 0.33 kPa (2.5 mmHg) or 0.42 kPa (3.2 mmHg) per minute (19, 31, 35). The rise in PaCO<sub>2</sub> predicted from normal physiology is 3–4 mmHg (43), but may vary considerably under conditions of brain death (cf. Fig. 4).

The rise during the first 4 min seems to be about twice or thrice as steep as thereafter (18, 35). Increases as steep as 12 mmHg have been observed during the first minute (43, 44). Paret and Barzilay (45) have proposed an algorithm for estimating PaCO<sub>2</sub> using the formula  $\ln \text{PaCO}_2 = 0.69 + 0.072 + 0.86 \ln \text{PaCO}_2 \text{O}$  where  $\ln$  is the natural logarithm and PaCO<sub>2</sub>O the PaCO<sub>2</sub> at the beginning of AT. We discourage a time-locked procedure and strongly recommend arterial blood gas determinations which are prescribed by many guidelines. The actual German protocol for the determination of brain death (27) requires the final PaCO<sub>2</sub> to be recorded. To our knowledge the duration of the apnea test may be extremely variable and last between 1 min and more than 1 h. The longest time we have met was 71 min. The use of (S)IMV ventilation instead of apneic oxygenation will not substantially prolong the procedure.

*Duration of observation of apnea* – The patient should be observed during the whole procedure for any respiratory movements and a sufficient time, about half a minute (36), after the recommended partial gas tension levels have been reached. To our knowledge 30–60 s appear to be sufficient indeed. Longer periods as mentioned by Kunesch et al. (46) are unnecessary and may result in profound hypoxia or acidosis. As soon as there is spontaneous breathing during AT, it must be terminated immediately.

*Proposed increase over baseline* – As a rapid increase in PaCO<sub>2</sub> to 20 mmHg above normal baseline is considered a strong stimulus for the respiratory centers, such an increase is recommended when the baseline PaCO<sub>2</sub> is at or above 36–40 mmHg (17, 29, 47, 48). This means that a PaCO<sub>2</sub> of about 60 mmHg will be reached. We strongly recommend determining final blood gases in each case and not to rely on an anticipated increase in PaCO<sub>2</sub>. As stated above, the rate of increase may vary considerably but is usually biphasic, being steeper during the first few minutes than thereafter (49).

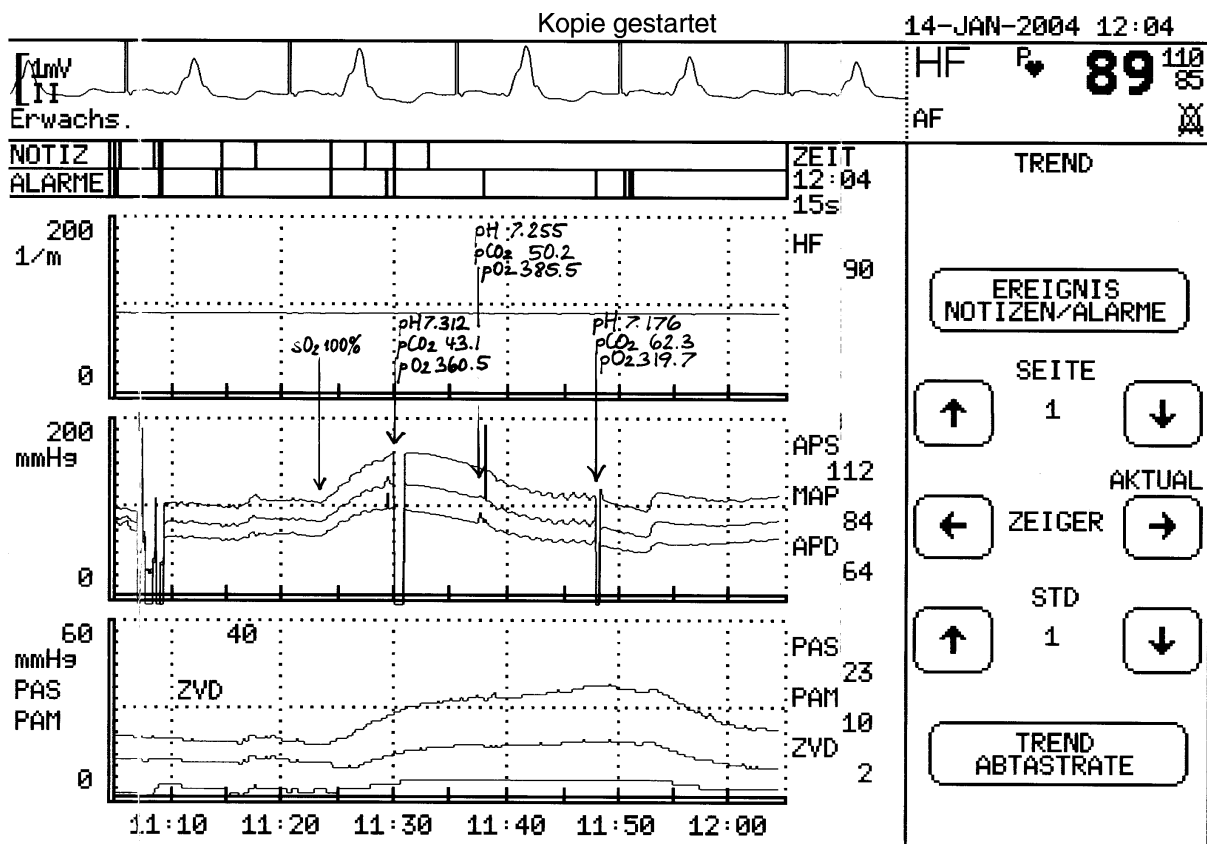
**Blood pressure** – A pretest systolic blood pressure (BP) of  $\geq 90$  mmHg is recommended by the Quality Standards Subcommittee of the American Academy of Neurology (17). The German protocol even recommends 100 mmHg preonset. If during AT there is a fall in BP it is recommended to not let it drop below 80 mmHg, but we have seen values as low as 50 mmHg that promptly recovered afterwards. Usually the BP response is biphasic, being mildly hypertensive with hyperoxygenation and mildly hypotensive with hypercarbia (50) (Fig. 5). We have never met serious problems with hypotension which could always be controlled by fluid balance/plasma expansion or catecholamines or both.

**Fluid balance** – Euvolemia or a positive fluid balance during the previous 6 h is recommended by the Quality Standards Subcommittee of the American Academy of Neurology (17).

**Medication** – Apnea testing must not be performed under the influence of drugs that may paralyze respiratory muscles, i.e. relaxants such as pancuronium, and apnea has been deemed possible by high doses of opioids and narcotics such as barbiturates.

Problems and pitfalls

**Barotrauma** – Some researchers have mentioned the possibility of barotrauma (51) which, however, is considered extremely rare (52). Eight cases have been described in the literature (51–57) and some have never encountered such a complication in over 20 years (52), while others found one instance among 63 within one and a half year (53). Tension pneumothorax and pneumoperitoneum may ensue massive oxygen insufflation into the endotracheal tube with an oxygen supply catheter that obliterates the tube or a valve mechanism which makes the escape of the insufflated gas impossible (53–55).



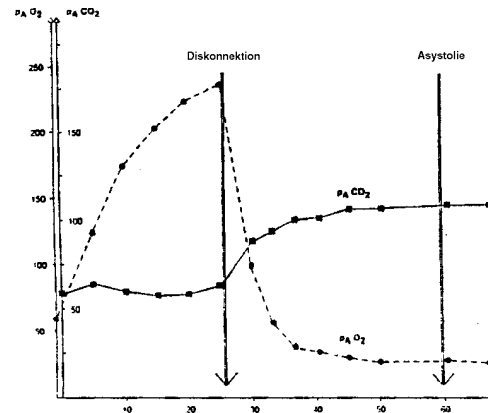
**Figure 5.** Trend monitoring during apnea testing which was performed using synchronized intermittent mandatory volume (SIMV) ventilation. The first downward arrow (from left to right) marks the beginning of hyperoxygenation with pure oxygen (sO<sub>2</sub> 100%), the second the beginning of hypoventilation, the third an intermediate blood gas check and the fourth the last blood gas check necessary for conformation of the required PaCO<sub>2</sub> level (here PaCO<sub>2</sub> = 62.3 mmHg) after which the patient was disconnected and observed for one more minute. Note the initial increase and subsequent decrease in arterial blood pressure. The top row illustrates heart frequency (HF), the middle row intraarterial blood pressure (APS, arterial pressure systolic; MAP, mean arterial pressure; APD, arterial pressure diastolic), and the bottom row pulmonary arterial (PAS, pulmonary arterial systolic; PAM, pulmonary arterial mean) and central venous pressure (ZVD).

Limitation of the insufflation rate, appropriate diameter relation between catheter and endotracheal tube, a not too deep insertion, and close observation of the thorax prevent this unwanted effect. It is best avoided by keeping the patient on the respirator and disconnecting it only during the final observational period after the target PaCO<sub>2</sub> has been reached. The bulk diffusion technique that has been described (14) also helps in avoiding this complication. The hypoventilation method we and others (58) use adhibiting (S)IMV ventilation of about 1 l/min is another safe method. We have never experienced lung trauma.

*Excessive hypercarbia* – Values > 120 mmHg should be clearly avoided because they may result in CO<sub>2</sub> narcosis, although general recommendations do not exist. A reduction in respiratory drive under otherwise normal conditions sets in at about 90 mmHg. In a paper by Rowland et al. (59) the PaCO<sub>2</sub> level after 15 min of AT in nine children was between 50 and 116 mmHg. The highest level we have ever observed was 132.6 mmHg. There are no generally accepted PaCO<sub>2</sub> values for patients whose natural respiration is adapted to a PaCO<sub>2</sub> of more than 45 mmHg. In these cases confirmation of loss of brain stem functions by instrumental investigations is recommended (27).

*Hypoxia* – Values < 60 mmHg should be avoided. As mentioned above we have met much lower uneventful temporary values. As we use controlled hypoventilation with pure oxygen we have not seen severe hypoxia any more. With adequate precautions and use of 100% oxygen such values should not occur and are to be prevented even with pulmonary problems such as lung contusion. If the respiratory center is being driven by hypoxia in patients known to be adapted to high PaCO<sub>2</sub> values, AT may be used that includes cautious lowering of PaO<sub>2</sub>. There are, however, no accepted criteria for this type of test (58). In a study by Ferbert et al. (18) asystole ensued at a PaO<sub>2</sub> of about 35 mmHg (Fig. 6). Artificial CO<sub>2</sub> augmentation without reduction or even with an increase in respiratory minute volume may be advantageous in these cases (7).

If pulse oxymetry is used, values should not drop below 80%. Inadequate preoxygenation is one of the main reasons for complications during AT (5). Neither hyperoxic hypoventilation nor apneic oxygenation should result in hypoxia if handled properly. In a prospective study of 36 patients (60) no relevant hypoxia (PaO<sub>2</sub> < 80 mmHg) was observed with apneic oxygenation.



**Figure 6.** Sharp drop of PaO<sub>2</sub> (dotted line) and slow increase in PaCO<sub>2</sub> (straight line) with disconnection from the respirator (first arrow, 'Diskonnektion'). Asystole (second arrow, 'Asystolie') occurred as a consequence of hypoxia, not hypercapnia or acidosis (18).

*Respiratory acidosis* – Values < pH 7.2 or 7.0 should be avoided according to current recommendations. In the study by Rudolf et al. (60) the mean pH at the end of AT was 7.18 in group 1 (observation time at least 5 min after obtaining a PaCO<sub>2</sub> of 40 mmHg) and 7.13 in group 2 (PaCO<sub>2</sub> at least 60 mmHg). In the study by Rowland et al. (59) the pH of one child came close to 6.9 and in the case report by Brill and Bigos (61) it was 6.94. We have seen values as low as 6.808 without complications and consider values down to 7.0 as acceptable. According to a report by Oikkonen et al. (62) apneic oxygenation could lead to profound respiratory acidosis (pH 6.72) and hypercarbia (PaCO<sub>2</sub> 250 mmHg), but if there was no concurrent profound hypoxia (PaO<sub>2</sub> ≥ 67 mmHg), cardiac sinus rhythm and a low but reasonable arterial pressure (75/55 mmHg) was to be kept up for hours. Work performed by Orliaguet et al. (63) has shown that AT is well tolerated despite severe respiratory acidosis.

*Hypotension* – Systolic values < 70 or 60 mmHg, diastolic values < 40 mmHg and a mean arterial pressure below 50 mmHg should be avoided. The drop in BP is most likely due to a reduction in the effect of catecholamines by acidosis. Usually there is a mild increase in BP with hyperoxygenation and a somewhat more marked decrease with hypercapnia (4, 50). To our knowledge these variations in BP never become threatening or pose serious problems if they are corrected, if necessary, using intravenous fluids, albumin, dopamine, dobutamine or (nor)epinephrine. Close monitoring, however, is strongly recommended (4, 64). The degree of change in mean arterial BP (MAP) can even be estimated before AT using an algorithm proposed

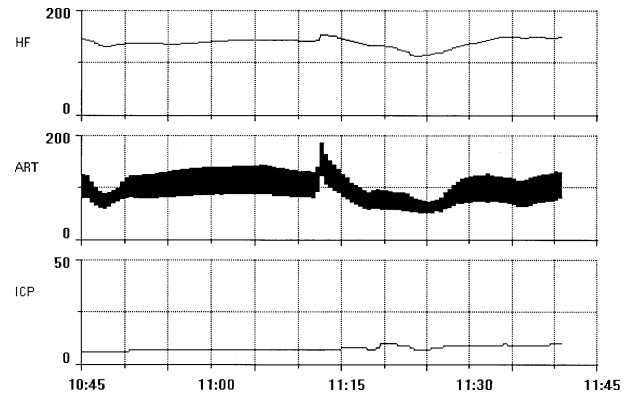
by Paret and Barzilay (45). The formula is  $MAP = 78.22 - 1.7t - 0.33PaCO_2O + 0.038PaO_2O$ , where  $t$  is the time in minutes and  $PaO_2O$  is the  $PaO_2$  at the beginning of AT. We suggest that in a brain dead patient whose parasympathetic cardiac regulation is lost, small changes in catecholamine perfusion rate may result in unusually large effects.

**Hypertension** – An increase in mean pulmonary arterial pressure has been noted by Nicolas et al. (65). There was a raise by 10 mmHg which correlated with an increase in  $PaCO_2$  (and a drop in pH). This effect is reversible and not deleterious for right ventricular function (66). We have most often noted a mild increase in systemic BP during hyperoxygenation and a decrease with hypercarbia (50).

**Cardiac arrhythmia and arrest** – Cardiac arrhythmias induced by AT are rare – <1% according to Goudreau et al. (5) – and cardiac arrest, as occurred in 2 of 63 cases in an Argentine series (53), should be avoided at any rate. The reason is most often excessive acidosis or hypoxia which is frequently heralded by the onset of new cardiac arrhythmias or a marked drop in heart rate (6). We have seen one cardiac arrest with inadvertent hypoxia that could be restored and one permanent cardiac arrest in a multitrauma victim who had suffered severe multiple organ damage including cardiac and lung contusion.

**Increase of intracranial pressure** – Intracranial (IC) pressure is usually not monitored during AT except in some neurosurgical intensive care units. Because theoretically AT may increase IC pressure via local remnant hypercapnic vasodilation and ensuing increase of cerebral blood volume, it should be the final resort of all clinical tests (Fig. 7).

The Japanese guidelines place AT at the end of all tests, after a flat EEG has been demonstrated. Because in primary brain stem lesion a slowing of the electroencephalogram (EEG) has been demonstrated, Schwarz et al. (67) also advocated that AT should be carried out after an isoelectric EEG. We ourselves have documented a case of loss of all brain stem functions without any EEG change during AT. A paper by Coimbra (68) has caused some concern as it suggests the possibility that a global reduction in blood flow may be further reduced and rendered irreversible by AT. This fear is neither warranted nor supported by empirical evidence. The concept of ischemic penumbra is applicable to a two- or three-compartment model where there is viable tissue, ischemic



**Figure 7.** Simultaneous registration of heart rate (HF, beats per minute), blood pressure (ART, systolic and diastolic in mmHg), and intracerebral pressure (ICP, mmHg). Hyperoxygenation started at 11:10 hours, hypoventilation at 11:15 hours. A  $PaCO_2$  of 60.5 mmHg was reached at 11:36 hours and half a minute later normoventilation resumed. There is a mild increase in ICP from 7 to a maximum of 10 mmHg.

penumbra, and infarcted tissue. It is conceived as a focal and temporal process (69, 70). It has never been demonstrated that global cerebral ischemic penumbra is transformed into global brain infarction by AT. However, conclusive evidence would, for example, require prospective serial PET assessments (71). As ischemic penumbra is always a transient phenomenon (72) the long duration of examining for brain death in itself is an argument against its impact in this particular situation. In cranio-caudal cerebral herniation the respiratory centers are the last to lose their function and as long as they are functional they will promptly respond to an increase in  $PaCO_2$  before the effects of raising IC pressure become effective. Any spontaneous respiration will immediately terminate AT. The potential action of  $CO_2$  as a vasodilator (73) and an agent apt to raise IC pressure is counteracted and limited by the rise in  $PaO_2$ . In those cases where we and our colleagues from Munich (H. Angstwurm, personal communication) had the opportunity to register IC pressure during AT no further increase was noted. We have seen single patients in whom respiration was not lost despite one or multiple AT, some of whom underwent a certain degree of recovery thereafter. A number of patients with preserved EEG, despite loss of all brain stem functions, went on producing electric brain activity for hours or days although AT was carried out several times. Moreover, patients with preserved cerebral blood flow may be brain dead due to parenchymal damage without herniation (74). If serious concerns exist, they may be settled by doing an atropine test (see below) or performing

an adequate instrumental investigation beforehand (75).

*False readings* – We have repeatedly noted that very sensitive respirators may be triggered by heartbeat-driven thorax excursions causing minimal air flow. Wijdicks (12) also mentioned this possibility when he remarked that with continuous positive airway pressure (CPAP) settings as low as two, false readings ('spontaneous' respiratory rates of 20–30/min) may occur. Similar may be the case with BIPAP respiration when airway pressure decreases in time with cardiac contraction (76). In these instances disconnecting the patient for observation, raising the trigger flow rate settings or reducing the assisted spontaneous breathing (ASB) level is recommended.

*Spinal reflexes* – Complex spinal cord symmetric upper limb movements resembling decerebrate posture have been described that were triggered not only by each mechanical pulmonary insufflation, but also by superficial pressure and noxious stimuli applied to the arms, thorax, or abdomen. They were abolished by disconnection from the respirator (77). Lazarus' sign may be observed during AT or after removal from the respirator (78).

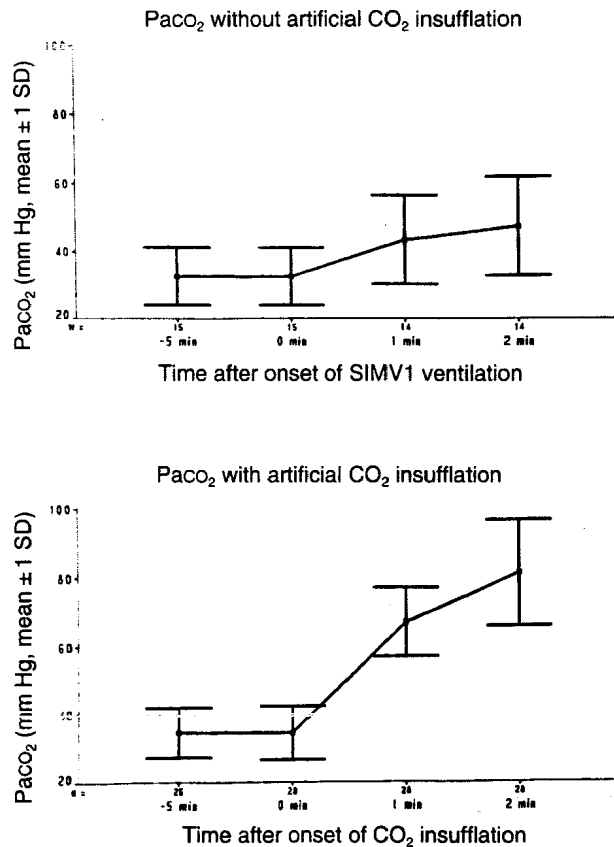
#### Special situations

*Children* – As it is assumed that the threshold for maximum stimulation is similar to that observed in adults, children should be handled accordingly (79, 80). However, as conditions in newborns, infants and even older children may differ from those found in adults due to a still immature brain stem or an open fontanel, new procedures are needed. There is a case report of a 3-year-old child with chronic severe neurological dysfunction who took a single breath after 8 min and 45 s of apneic oxygenation at a PaCO<sub>2</sub> of 112 mmHg determined thereafter but ultimately died 5 days later (61). Another report is that of a 3-month-old infant who after fulfilling the 1987 Task Force Criteria of pediatric brain death developed two or three irregular breaths days later but also finally died (80). In a 4-year-old child a higher PaCO<sub>2</sub> threshold was assumed as the child had minimal respiratory effort at 91 mmHg (81). Levin and Whyte reported an infant who started breathing at 59 mmHg (82). In a study by Ashwal (83) the mean PaCO<sub>2</sub> at the end of AT in children was 73.3 mmHg PaCO<sub>2</sub>. There is a most unusual report from Japan where a 3-month-old girl suffering from hypoglycemia developed low-frequency

irregular spontaneous respirations after fulfillment of current criteria for brain death (84) at PaCO<sub>2</sub> levels between 30.1 and 73.5 mmHg. None of these children recovered, but they all finally succumbed to circulatory failure. This raises the question whether in children of a certain age higher PaCO<sub>2</sub> levels should be requested or whether the demonstration of loss of brain perfusion should be mandatory. Given these observations 5 min of apneic oxygenation – as were actually performed by some clinicians – is definitely inappropriate (42, 79). The Portuguese guidelines (85) suggest different PaCO<sub>2</sub> levels for adults (50 mmHg, 6.65 kPa) and children (60 mmHg, 7.98 kPa). According to the German guidelines (27), in children under the age of 2 years AT has to be performed two times by two examiners 24 or 72 h apart. Although there are reasons to believe that preterm infants do not behave differently from full-term newborns, there are no sufficiently valid recommendations within this age group (86).

*Monitoring* – End-tidal capnometry, pulse oxymetry, transcutaneous blood gas determination (87), and intra-arterial blood gas analysis may all be used (50, 88). However, *in vitro* measurements, i.e. regular blood gas determinations, remain the gold standard and should not be replaced by other means. Some authors recommend determining a basal value and then to do subsequent checks approximately every 5 min. Having obtained two or three values the increase in PaCO<sub>2</sub> may be extrapolated rather precisely. We feel that the number and timing of blood gas checks should be handled individually according to initial values, monitoring, and the expertise of the examiner. Close monitoring is especially useful and necessary with artificial CO<sub>2</sub> augmentation, depending on the anticipated increase in CO<sub>2</sub> (7). Final values should always be corrected for body temperature. The use of a (portable) blood gas check equipment at the bedside is an especially efficient method as it allows performing the blood gas analyses and observing the patient at the same time by one examiner.

*Artificial CO<sub>2</sub> insufflation* – The President's Commission already suggested the use of an oxygen and carbon dioxide mixture (33) in order to raise PaCO<sub>2</sub>. If an especially long duration of observation is expected, if an increase in PaCO<sub>2</sub> is not easily achieved or very lengthy, or if PaO<sub>2</sub> drops excessively with hypoventilation, this method may be used (7). Careful and close monitoring must be ascertained, but if handled properly the test duration may be shortened considerably, rendering the procedure safer in some cases (89, 90) (Fig. 8).



**Figure 8.** Comparison of the rise of PaCO<sub>2</sub> with hypoventilation (top) and artificial CO<sub>2</sub> insufflation (bottom). The increase in PaCO<sub>2</sub> is much steeper with the second condition (7).

*Replacement of apnea testing by other means* – In cases where correct AT is not possible, e.g. because of the impossibility to reach the required PaCO<sub>2</sub> values or when a dangerous drop in PaO<sub>2</sub> is unavoidable such as may be the case in severe thorax trauma or other pulmonary problems, instrumental brain stem testing like arteriography, perfusion SPECT, ultrasound Doppler, or evoked potentials (AEP [auditory evoked potentials], SEP [somatosensory evoked potentials]) may replace this part of the examination according to the German guidelines (27). The same procedure was recommended in patients who were adapted to a PaCO<sub>2</sub> of more than 45 mmHg (39).

#### Recommendations and proposals

Recommendations for AT for the determination of brain death have been given by various committees or persons of different nations (27, 91), including explicit instructions and block diagrams. AT should be the last resort or postponed until an atropine test (26, 92) – usually by injection of 2 mg – is negative (93). If it is undoubtedly positive, AT is not warranted. Care has to be taken not to inject

atropine into a line that was used for catecholamines as the washout of even small quantities may yield erroneous results. Brain stem respiratory centers and vagal neurons are situated in close vicinity such that both functions are highly correlated. In our clinical practice we have seen only one discrepant result (negative atropine test, but still respiration on AT) among more than 100 cases. If atropine testing is negative, AT must be carried out. Hypoventilation after preoxygenation is our preferred means. A body temperature of >32°C should be ascertained. BP, preferentially monitored intraarterially, should be no lower than 80 mmHg initially and the pH preferably in the normal to mildly alkalotic range. If an unwanted drop of BP is seen during testing it can be raised using vasopressor agents or volume substitution or both. After preoxygenation with 100% O<sub>2</sub> the highest possible PaO<sub>2</sub> should be achieved, then ventilatory volume reduced to approximately 5–2 l/min [e.g. by (S)IMV ventilation, alternatively by disconnection from the respirator and insufflation of O<sub>2</sub> into the trachea at the level of the carina at a rate of about six or more liters per minute under close observation until requirements are met (12, 94)], and the PaCO<sub>2</sub> be checked after an appropriate period of time depending on the initial blood gas values corrected for body temperature. Pulse oxymetry, end-tidal capnometry, a BP and heart rate monitor, an IA line and the feasibility of rapid determination of arterial blood gases are very helpful; an optimal setting would be a bedside blood gas check device that enables the examiner to perform blood gas analyses without losing the patient out of sight. As soon as the required PaCO<sub>2</sub> level is reached, the patient should be disconnected from the respirator (if he is not already) and observed for an appropriate period of time (30–60 s), then reconnected and mildly hyperventilated for some minutes until initial values are restored approximately except in those cases where AT is the final test and the respirator may remain switched off thereafter. We use a fine thread for the detection of respiratory activity which is sensitive enough to show displacement by heartbeats, thus proving that the system is capable of monitoring even the slightest breathing. Other methods such as a moist thin piece of soft paper, a cold mirror or blank piece of metal, however, may serve as well.

#### Discussion

Testing for apnea is considered indispensable for the determination of brain death worldwide and may be safely performed under almost any

circumstances, if adequate precautions are met (12, 25, 64), although it remains a formidable task to the examiners. The demonstration of loss of breathing bears high intuitive evidence that life has vanished from man also for laypersons. It is not as difficult or harmful as some authors want to make us believe. To our knowledge AT can be performed *lege artis* in 99.8% of all cases if currently available possibilities are exhausted; the remaining ones may be handled using subsidiary instrumental tests. The complication rate (e.g. cardiac arrest) is very low. Differences in national guidelines exist and there is at least one country (Portugal) recommending different procedures in children and adults. As with criteria for brain death itself, it would be utterly desirable to have identical guidelines worldwide including the target PaCO<sub>2</sub> which, to our opinion, should be at least 60 mmHg in adults. This would abolish the vexing statement raised by Levin and Whyte (82) that clinical criteria – i.e. for AT – make it possible to be brain dead in one country and not in another.

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