Virchows Archiv A Path. Anat. and Histol. 366, 137—147 (1975)

Light and Electron Microscopic Examinations of Experimentally Produced Heart Muscle Necroses Following Normobaric Hyperoxia * **

Carl Michael Büsing, Ute Kreinsen, Fritz Bühler and Uwe Bleyl

Pathologisches Institut, Universität Heidelberg
(Direktor: Prof. Dr. W. Doerr)

Received November 27, 1974

Summary. Heart muscle necroses following normobaric hyperoxia as examined under a light microscope occur after 40 hours exposure in the rabbit and increase in intensity with prolongation of the exposition time. Simultaneous arterial blood gas analysis excludes a hypoxemia as the cause. In addition electron microscopic examination of the necroses demonstrate the primary changes in the myofibrils but not in the mitochondria. The ultramorphological picture is, however, quite similar to the myocardial necroses after epinephrine application in the rabbit. A pathogenetic relationship between heart muscle necroses after normobaric hyperoxia and after epinephrine should be considered.

Key words: Normobaric Hyperoxia — Heart Muscle Necroses — Oxygene Toxicity — Myocytolysis — Epinephrine Myocarditis.

Zusammenfassung. Herzmuskelnekrosen nach normobarer Hyperoxie sind beim Kaninchen nach ca. 40 Stunden lichtmikroskopisch nachweisbar und nehmen mit der Länge der Expositionszeit an Stärke zu. Simultane arterielle Blutgasanalysen schließen eine generalisierte Hypoxämie als Ursache aus. Elektronenoptisch zeigen die Nekrosen die frühesten Veränderungen an den Myofibrillen im Sinne einer Myozytolyse, weniger an den Mitochondrien. Das ultramorphologische Bild gleicht hierin weitgehend dem der Myokardnekrosen nach Adrenalin-Medikation beim Kaninchen (sog. Epinephrine-Myocarditis). Eine pathogenetische Beziehung zwischen Herzmuskelnekrosen nach normobarer Hyperoxie und durch Adrenalin läßt sich vermuten.

Organic changes due to normobaric hyperoxia (100 per cent oxygen at 1 ata pressure) are primarily seen in the lungs (Lorrain-Smith-effect). However, other organs also demonstrate tissue damage (Liebegott, 1941; Kaunitz, 1942; Kühn, 1944; Pichotka und Kühn, 1947; Berfenstam *et al.*, 1958; Caulfield *et al.*, 1962; Nolte, 1968; Johnson *et al.*, 1972). The pathogenesis of these oxygen-induced changes is still debatable but animal experiments awake the possibility of that endocrine processes, especially adrenergic substances, are involved (Bean, 1965; Clark and Lambertsen, 1971).

Heart muscle necrosis in rabbits following either normobaric hyperoxia or careful administration of epinephrine (socalled epinephrine myocarditis) have been repeatedly described and are easily reproduced (Fleisher, 1909; Szakacs, 1958; Ferrans, 1969). Our experiments are concerned with the time-dependency of

* In thankful appreciation to and in honor of Professor W. Doerr on the occasion of his 60th birthday.

** Sections of this report have been presented at the 57th congress of Deutsche Gesellschaft für Pathologie at Karlsruhe, Germany, 1973.

the myocardial necroses on the oxygen exposition as well as the comparison of the ultramorphological findings with the distribution pattern of the myocardial necroses after normobaric hyperoxia and after administration of l-norepinephrine.

Material and Methods

Thirty bred rabbits with body weights between 2000 and 4000 g were held for varying periods of time in a 40 l cage which were coupled to an overflow system with 100 per cent oxygen. Ten animals were held in an identical manner with compressed air instead of oxygen. The humidity ranged between 60 and 100 per cent, the temperature between 20 and 25 degree Celsius at a flow rate of 5–10 liters per minute. Food and water were always available. After an exposition time of 30–90 hours the animals were anesthetized with pentobarbital and wasted with a blow to the neck. Eight animals died spontaneously. The organs were fixed in a 10 per cent formalin solution, bedded in paraffin and 4 μ sections with hematoxylin-eosin stained, exposed to the PAS-reaction and, in some cases, stained with van Gieson.

Preparation for the electronmicroscopic examination was carried out by retrograde perfusion in pentobarbital anesthesia via the aorta with buffered 3 per cent glutaraldehyde solution while the heart was still beating (method according to Forssmann, 1967). The tissue samples were bedded in Araldit, sectioned with the Ultramicrotome (Fa. Reichert, OMU 2) and after contrasting with uramylacetate and lead citrate examined under the electron microscope (Carl Zeiss EM 9).

Prior to the start of the experiments, an arterial catheter for blood samples was placed in the right femoral artery and fixed to the subcutaneous tissue. Arterial blood gas analysis was performed according to the method of Andersen and Astrup (1960).

The light microscopic examination was performed on horizontal and longitudinal sections through the heart chosen in a manner so that specimens from the septum, right and left ventricle, both atria and portions of the papillary muscles were obtained. A topographical analysis of the severity of necroses was performed by histological examination of the following regions: internal layer, external layer and papillary muscle of the left ventricle, left and right septum, right ventricle wall. Uncommon locations of necroses were specially noted. In this way an integrated total analysis of the destruction of the heart muscle was achieved. The grading of the necroses was performed by two of the authors independently and a definite degree of agreement was attained: grade 1=disseminated necroses, individual fiber necroses, grade 2=small focal closely arranged necroses, grade 3=large bordering necrotic fields.

In 8 additional animals with an average body weight of 2350 g we could produce severe myocardial necroses by injecting subcutaneously 1.2 mg l-norepinephrine (Arterenol from Hoechst, Frankfurt) per kg body weight in a 10 ml physiologic sodium chloride solution. The animals were wasted 24 hours later by a blow to the neck in light pentobarbital anesthesia and the hearts examined according to the method described above. Two animals in this group were exposed to perfusion fixation.

Results

1. Severity of the Necroses Dependent upon the Exposure Time

Employment of an unlimited oxygen exposure time results in the spontaneous death of all animals in approximately 100 hours (see Büchner, 1933; Deen, 1969). In these cases extended myocardial necroses could be found. A shortening of the exposition time under 70 hours results in a reduction in the severity of necroses which are graded into 3 categories. Less than 50 hours exposure causes inconsistent findings, i.e. heart muscle necroses are either discrete or not at all present in some of the animals. In contrast exposition times in excess of 60 hours result in heart muscle necroses in all animals. The control animals were with the exception of one negative. On a coordinate system, the heart muscle necroses graded according to the staging mentioned above on the ordinate and the oxygen exposition time or

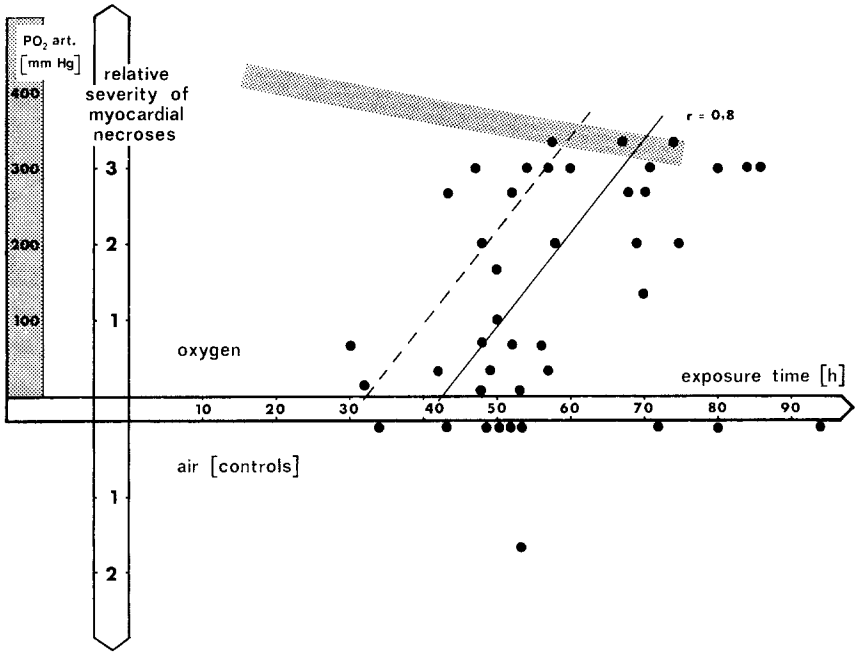

Fig. 1. Total degree of the myocardial necroses divided into 3 degrees (ordinate) as dependent upon exposition time (abcissa). The following point pattern resulted, each point represents one animal. The regression (solid line) has a coefficient of correlation of $r=0.8$. The broken parallel is advanced 10 hours and cuts the abcissa at 30 hours. The shaded area represents the measured arterial oxygen partial pressure

survival time on the abcissa, each animal is represented by a point (Fig. 1). From the pattern a regression can be calculated with a coefficient of correlation=0.8.

Intermittent arterial blood gas analyses were performed on 6 animals whose oxygen pressure is graphically presented in Fig. 1. The arterial oxygen tension value (PO_2 art.) were still around 300 mmHg even after an exposition time of 70 hours. The oxygen saturation was always 100 per cent. The CO_2 partial pressure was not increased. In both groups (6 oxygenated animals, 2 controls) arterial pH values between 7.45 and 7.55 were measured at a CO_2 partial pressure of 20–30 mmHg and a standard bicarbonate of 22–25 mMol/l. Higher blood pH values and decreased CO_2 partial pressures were found apparently in the oxygenated animals as compared to the controls.

2. Light Microscopic Findings and Location of the Heart Muscle Necroses

Under the light microscope a leucocytic, mainly histiocytic and phagocytic reaction which varied in its intensity could be found in the periphery of partially homogenised, pale and streaky-stained muscle fibers which themselves were reduced in caliber or split. Throughout scattered empty sarcolemma tubes or necrotic zones with early fibroblastic proliferation could be observed. Occasionally an interstitial edema surrounding small vessels was visible (Fig. 2).

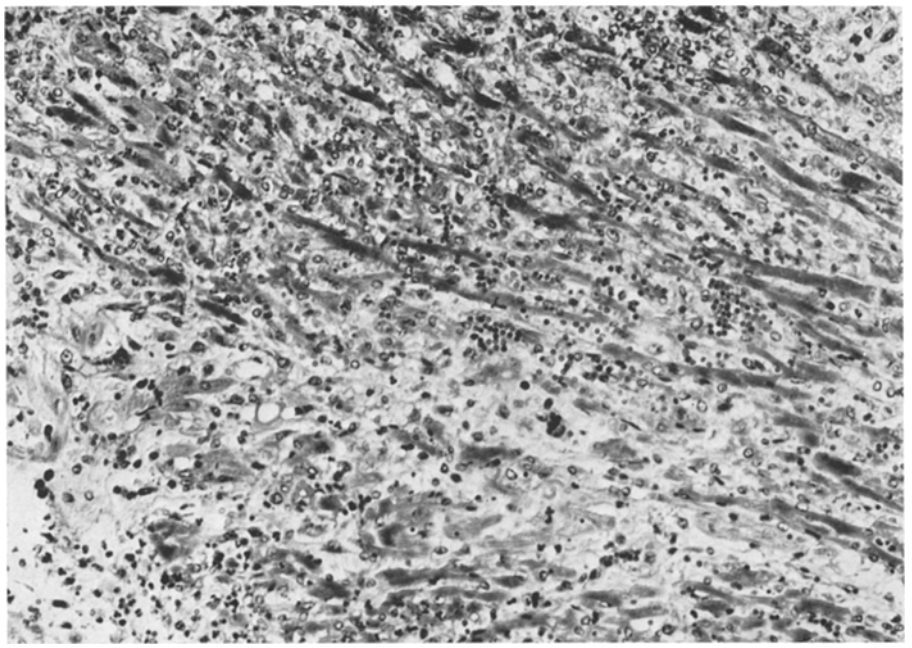

Fig. 2. Severe, aggregated myocardial necroses in the left ventricular wall with round cell reaction and beginning fibroblastic proliferation. Formalin fixation, paraffin, hematoxilin-eosin. Magn. 1:200

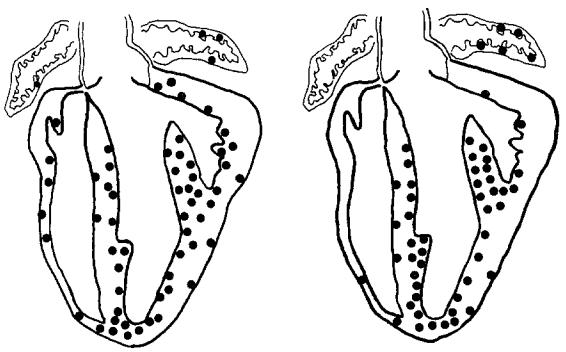

Fig. 3 and 4. Longitudinal cuts through both ventricles showing the distribution of O_2-induced necroses (left) and necroses after application of epinephrine (right). The inner layer of the left ventricle and papillary muscle are primarily affected, the right ventricular wall to a lesser degree

A schematic projection of the heart muscle necroses graphically superimposed upon one another would result in a topographical distribution pattern of the necrotic fields (Fig. 3 and 4). A concentration is found in the internal layers of the left ventricle expecially in the papillary muscle. The frequency increases in the free ventricular portion from the base to the apex. The left ventricular portions of the septum are affected in a similar manner, less often are necroses in the right

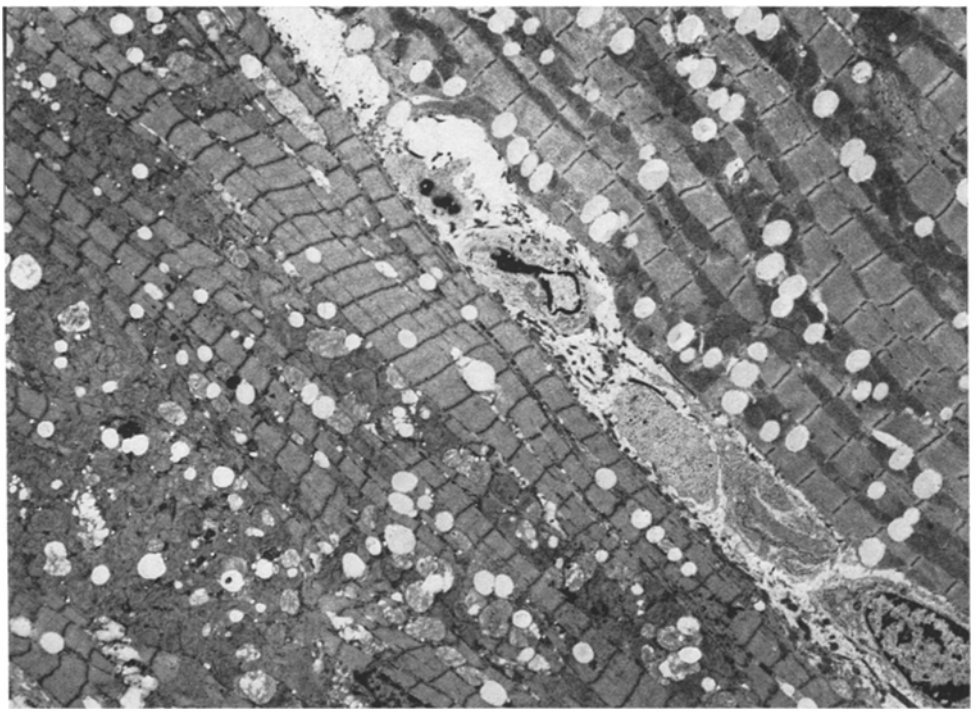

Fig. 5. Severe, diffuse, fine fat droplets in the myocardial fibers following oxygen exposure. Perfusion fixed, ultra-thin section. Magn. 1:1700×3.0. Photo: Derks

subendocardium and in the right ventricular wall. Necroses in the right and left atria were only sporadically seen. A prevalence within the right ventricle was not recognized. The above-mentioned distribution pattern coincides with the findings in several cross-sectional preparations. The necroses of the clinically suspicious control (infection of the respiratory tract and minimal pareses) are distributed mainly in left ventricular papillary muscles. The experimentally produced heart muscle with l-norepinephrine was schematically represented in a similar manner (Fig. 4). A comparable distribution pattern is attained. Eventually the free portions of the right ventricle appear less affected compared with oxygen-exposed animals, albeit the difference is minimal.

3. Electron Microscopic Findings

Under low-power magnification the left ventricular myocardium of the oxygen-exposed animals demonstrates definite, fine fat deposits in the heart muscle fibers (Fig. 5). The fat vacuoles are diffusely distributed between the cell structures, often without any apparent order, sometimes accumulated near the mitochondria. The osmiophilia is minimal which can be considered a sign of higher saturation of the fatty substances (Fawcett, 1969). The sarcoplasmatic reticulum is segmentally dilated. In addition to the severe necrotic zones with monocytic-histiocytic phagocytic reaction and near-total destruction of muscle

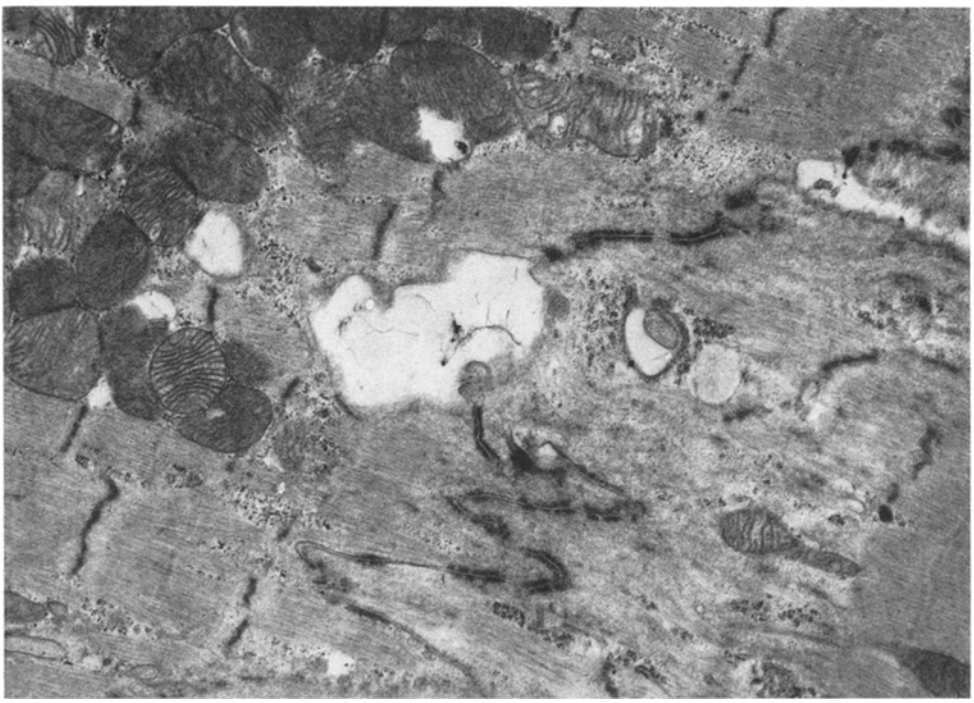

Fig. 6. Early changes of the myocardial fibers after normobaric hyperoxia for 74 hours in the rabbit. Loss of myofibril structure including the Z-bands expecially near the disci intercalares. Relatively well-preserved mitochondria. Perfusion fixed, ultra-thin section. Magn. 1:6200×3.1. Photo: Derks

fibers, the zones with minimal or primary damage is of particular interest to us (Figs. 6 and 7). They show a splintering of the Z-bands with fuzzy contours and irregular structure. The M-band is scarcely visable in the A-band region. An advanced disintegration of the myofilaments is partially present. The mitochondria are intact, well-defined, compact and the mitochondrial cristae are mainly intact, too. Only with advanced destruction the mitochondria are also affected. The capillary lumina are not remarkable, the endothelium is not swollen and a pericapillary edema is only occasionally present. Rarely hyaline concentration bands (cross banding) appear. In contrast to the lungs (Büsing and Bleyl, 1974) disseminated microthrombi were never found in the myocardial vessels.

Fig. 7. Early changes in the myocardial fibers after normobaric hyperoxia for 74 hours in the rabbit. In the middle fibers with loss of fibril structure but preserved mitochondria. In the right upper corner the capillary endothelium. Perfusion fixed, ultra-thin section. Magn. 1:3400 ×3.1. Inset: Magn. 1:6200×3.0. Photo: Derks

Fig. 8. Heart muscle changes after l-norepinephrine in the rabbit. Very close correlation with the findings after normobaric hyperoxia: myocytolysis with structure loss of the myofibrils and the Z-bands with relatively well-preserved mitochondria. Perfusion fixed, ultra-thin section. Magn. 1:3400×3.1. Inset: Magn. 1:3400×5.4. Photo: Derks

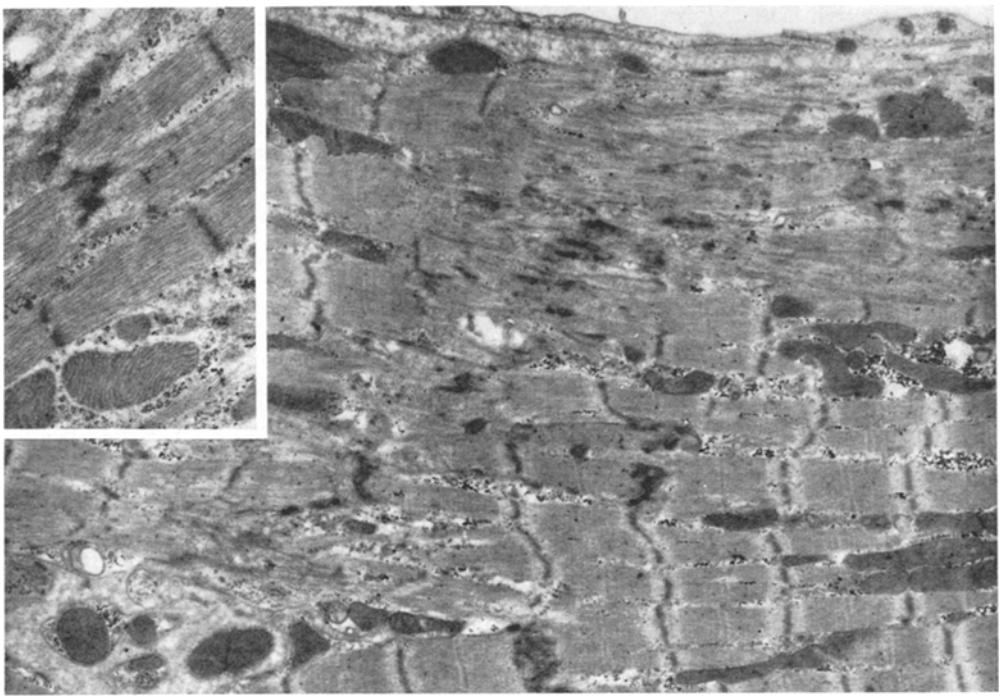

Fig. 7

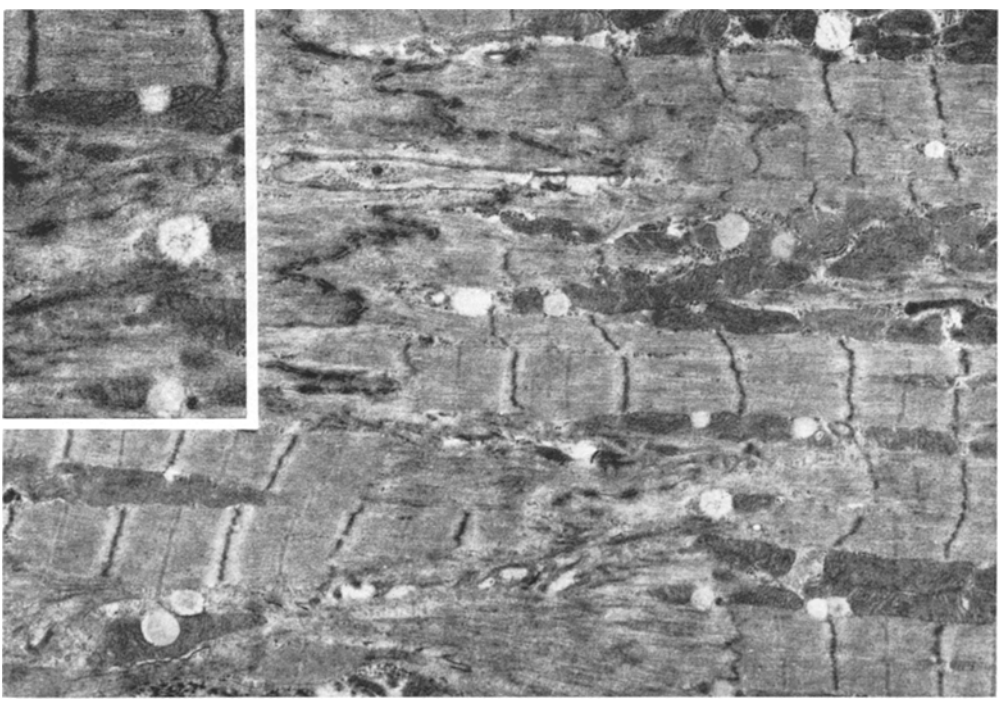

Fig. 8

Quite similar, comparable changes can be seen in the electron-optic pictures of the heart following epinephrine-application (Fig. 8), especially in the fat deposits and the early destruction of the myofibrils. A hyaline cross-banding of the muscle fibers as seen in the examinations on rat hearts (Bühler *et al.*, 1974) is seldom present in the rabbit myocardium.

Discussion

The following possibilities as pathogenetic mechanisms for the oxygen-induced toxicity of the organs and central circulatory system are discussed:

1. The production of H_2O_2 from oxygen causes damage to the membrane structures (Gerschmann, 1964; Johnson *et al.*, 1972), or blockage of the SH-radical-carrying ferments leads to destruction of the cell metabolism. A hematogenous mediator is discussed (Haugaard, 1968; Weibel, 1973).

2. The cell necroses following hyperoxia are due to a paradoxical tissue hypoxia which is founded in a primary pulmonary disturbance with distribution imbalances and diffusion abnormalities causing the hypoxia (Liebegott, 1941; Kühn, 1944). Peripheral vasoconstriction (Kety *et al.*, 1948; Lambertsen, 1953; Johnson, 1969; Winter *et al.*, 1969) or abnormal gas exchange at the blood-tissue level (Malorny, 1944; v. Lieven *et al.*, 1971) could also be responsible.

In older reports (Liebegott, 1941; Kaunitz, 1942; Kühn, 1944; Pichotka *et al.*, 1947; Berfenstam *et al.*, 1958) the heart muscle necroses in animals which died spontaneously under normobaric hyperoxia were considered due to generalized hypoxemia. The severe morphological pulmonary changes and the clinical picture of severe dyspnea together with the electrocardiographic findings substantiated this theory. Our own examinations show, however, that the heart muscle necroses are already present before any morphological pulmonary changes can be detected. Pulmonary hyaline membranes in rabbits first appear after an exposition time of at least 50 hours (Büsing and Bleyl, 1974). Even Kistler *et al.* (1967) and Kapanci *et al.* (1969) using morphometric techniques perceived the primary changes in the fine structure of the air-blood barrier of the rat after 48 hours. The results of our blood gas analysis under oxygen exposition up to 70 hours show a distribution disturbance but no hypoxemia (Fig. 1). This probably first appears in the final stages (Smith *et al.*, 1963; Caulfield *et al.*, 1972). A tissue hypoxia due to intramyocardial factors can naturally not be ruled out from blood gas analyses. Malorny (1944) and more recently Lambertsen *et al.* (1953) as well as Orzechowski (1968) have discussed an abnormal gas exchange of the erythrocytes by oxygen supersaturation (Haldane effect). A tissue acidosis from the abnormal CO_2 transport is also theoretically feasable (Gesell effect). However, both these factors are not major influences under normobaric hyperoxic conditions (Lambertsen *et al.*, 1955).

Exept for the arterial oxygen partial pressure, the other values attained by arterial blood gas analyses are to be interpreted with caution as we could not derive an acceptable normal value or find one in the literature (for comparison see v. Lieven *et al.*, 1971). The oxygen-exposed animals demonstrated a slight increase in the pH value at low PCO_2 as compared with the controls which we interpret as the result of a central hyperventilation (Deen, 1969; Ardisson *et al.*, 1972).

Prolonged survival time in the O_2 atmosphere is coupled with a corresponding increase in the myocardial necroses. This finding is substantiated through the statistically determined regression. The regression line crosses the time axis at about 40 hours, and at this time the first manifestations of muscle necroses can be expected. Our analysis included only those findings with definite myocytoclasia and reactive phagocytosis, which means an advanced stage of myocardial damage. Therefore, the primary damage to the heart muscle fibers must begin earlier. According to experiments by Büchner (1933) on hypoxic heart muscle injury our findings which demonstrated a strong phagocytic reaction are approximately 8–12 hours old. The irreversible injury to the heart muscle fibers due to the increased oxygen partial pressure must already be present after 30 hours exposure time (Fig. 1).

Under the light microscope our distributional pattern of the oxygen induced myocardial necroses is in correlation to the reports by Liebegott (1941) and Kühn (1944): the socalled internal layer of the left ventricle, especially the papillary muscle is primarily affected, the right ventricle to a much less degree. It presents a similar picture as that pattern of hypoxic heart muscle necroses reported by Büchner (1933). In addition the experiments demonstrate a close topographical correlation between the epinephrine-induced lesions and those after normobaric hyperoxia, too.

But under the electron microscope some differences must be stated. The findings after epinephrine and hyperoxia are quite similar: together with fine fat droplets the early changes with destruction of the Z-bands and structure loss of the myofibrils are found with only minimal alterations in the corresponding mitochondria. Cristolysis of the mitochondria appears only in the advanced stages of destruction of the muscle fibers. Here the hypoxemic heart muscle necrosis differs in that the damage is first seen in the mitochondria (Büchner *et al.*, 1967; compare with Bühler *et al.*, 1974). Korb (1964) observed similar changes primarily in the myofibrils after isoproterenol injection in rats and differentiated this myocytolysis from the hypoxemic heart muscle injury with coagulation necrosis without giving an apparent explanation for this difference.

From the foregoing findings a generalized oxygen deprivation cannot be held responsible for the heart muscle necroses described. The pathogenetic relationship between normobaric hyperoxia and myocardium necrosis could be a direct myocytotoxic effect of the oxygen on the muscle cell as v. Lieven (1972) has described. This opinion is supported by the in vitro experiments by Haugaard (1968). Less understandable is the location of damage in the subendocardial layers of the left ventricle.

It is known that the perfusion of the inner layer of left ventricle occurs almost totally during diastole, i. e. discontinuously. This is due to the systolic intramural pressure which is greatest in the subendocardium and follows a decreasing gradient to the outside. The left ventricular external layer and the right ventricular myocardium are almost continuously perfused. Research on the cardial microcirculation has not shown any significant difference in the perfusion per unit time under normal circumstances between the inner and external layers of the left ventricle (Buckberg *et al.*, 1972). Caulfield *et al.* (1972) has attributed the injury to the inner layer of the left ventricle to a diffusion of the oxygen out of the blood of the left ventricle. This appears, however, not very probable; particularly since differences between the right and left atrial muscle are not present and necroses are also found in the right ventricular myocardium.

The electron microscopic findings raise the question whether the oxygen affects the heart muscle cell indirectly through an intermediary mechanism thereby leading to myocytoclasia. As intermediator the adrenergic substances deserve first consideration as their participation in oxygen intoxication can certainly be substantiated (Bean, 1965; Wood *et al.*, 1967; Clark *et al.*, 1971). Our ultramorphological findings have at least caused suspicion for a common terminal mechanism in the heart muscle necroses after normobaric hyperoxia and after norepinephrine medication.

We are greatly indebted to Priv. Doz. Dr. W. W. Höpker for his support in statistical problems.

References

Andersen, O. S., Engel, K., Jørgensen, K., Astrup, P.: A micromethod for determination of pH, carbon dioxide tension, base excess and standard bicarbonate in capillary blood. Scand. J. clin. Invest. **12**, 172 (1960)

Ardisson, J.-L., Dolisi, C., Gastaud, M.: Les effects ventilatoires de l'hyperoxie normobare. Path. Biol. **20**, 195 (1972)

Bean, J. W.: Factors influencing clinical oxygen toxicity. Ann. N. Y. Acad. Sci. **117**, 745 (1965)

Berfenstam, R., Edlund, T., Zettergreen, L.: The hyaline membrane disease. Acta paediat. (Uppsala) **47**, 82 (1958)

Buckberg, G. D., Fixler, D. E., Archie, J. P., Hoffman, J.: Experimental subendocardial ischimia in dogs with normal coronary arteries. Circulat. Res. **30**, 67 (1972)

Büchner, F.: Das morphologische Substrat der Angina pectoris im Tierexperiment. Beitr. path. Anat. **92**, 311 (1933)

Büchner, F., Onishi, F., Onishi, S.: Frühstadien der akuten hypoxischen Veränderungen des Herzmuskels im elektronenmikroskopischen Bild und ihre Bedeutung für die akute hypoxische Herzinsuffizienz. Beitr. path. Anat. **135**, 153 (1967)

Bühler, F., Bersch, W., Kreinsen, U.: Zur Pathomorphologie der Epinephrin-Myokarditis nach Gabe von Hypertensin. Virchows Arch. Abt. A **363**, 249 (1974)

Büsing, C. M., Bleyl, U.: Oxygen induced pulmonary hyaline membranes and disseminated intravascular coagulation. Virchows Arch. Abt. A **363**, 113 (1974)

Caulfield, J. B., Schelton, R. W., Burke, J. F.: Cytotoxic effects of oxygen on striated muscle. Arch. Path. **94**, 127 (1972)

Clark, J. M., Lambertsen, C. J.: Pulmonary oxygen toxicity: a review. Pharmacol. Rev. **23**, 37 (1971)

Deen, L.: Hyperbare Oxygenierung. Anaesthesist **18**, 205 (1969)

Fawcett, D. W.: Die Zelle. Ein Atlas der Ultrastruktur. München-Berlin-Wien: Urban & Schwarzenberg, 1969

Ferrans, V. J., Hibbs, R. G., Walsh, J. J., Burch, G. E.: Histochemical and electron microscopical studies on the cardiac necrosis produced by sympathomimetic agents. Ann. N. Y. Acad. Sci. **156**, 309 (1969)

Fleisher, M. S., Loeb, L.: Über experimentelle Myocarditis. Zbl. allg, Path. path. Anat. **20**, 105 (1909)

Forssmann, W. G., Siegrist, G., Orci, L., Girardier, L., Pictet, R., Rouiller, Ch.: Fixation par perfusion pour la microscopie électronique. Essai de généralisation. J. Microscopie **6**, 279 (1967)

Gerschmann, R.: Biological effects of oxygen. In: F. Dickens and E. Neils eds., Oxygen in the animal organism. New York: MacMillan 1964

Haugaard, N.: Cellular mechanism of oxygen toxicity. Physiol. Rev. **48**, 311 (1968)

Johnson, R.: Changes in tissue circulation and oxygenation with alteration in arterial gas tension. Proc. 4th Intern. Congr. Hyperbaric Medicine, Sapporo, 1969, p. 37

Johnson, W. P., Jefferson, D., Mengel, C. E.: In vivo hemolysis due to hyperoxia: role of H_2O_2 accumulation. Aerospace Med. **43**, 943 (1972)

Kapanci, M. D., Weibel, E. R., Kaplan, H. P., Robinson, F. R.: Pathogenesis and reversibility of the pulmonary lesions of oxygen toxicity in monkeys; II. Lab. Invest. **20**, 101 (1969)

Kaunitz, J.: Myokardial damage resultin from high oxygen tension. J. Aviat. Med. **13**, 267 (1942)

Kety, S. S., Schmidt, C. F.: Nitrous oxide method for quantitative determination of cerebral blood flow in man; theory, procedure and normal values. J. clin. Invest. **27**, 484 (1948)

Kistler, G. S., Cadwell, P., Weibel, E. R.: Development of fine structural damage to alveolar and capillary lining cells in oxygen poisoned rat lungs. J. Cell. Biol. **32**, 605 (1967)

Korb, G.: Elektronenmikroskopische Befunde am Herzmuskel nach hohen Aludrininjektionen. Verh. dtsch. Ges. Path. **48**, 245 (1964)

Kühn, A.: Elektrokardiographische und histologische Untersuchungen bei der Sauerstoffvergiftung. Arch. Kreisl.-Forsch. **13**, 120 (1944)

Lambertsen, C. J.: Oxygen toxicity, effects in man of oxygen inhalation at 1 and 3,5 atmospheres upon blood gas transport, cerebral circulation and cerebral metabolism. J. appl. Physiol. **5**, 471 (1953)

Lambertsen, C. J., Ewing, J. H., Kough, R. H., Gould, R., Stroud, M. W.: Oxygen toxicity. J. appl. Physiol. 8, 225 (1955)

Lambertsen, C. J., Kough, R. H., Cooper, D. Y., Emmel, G. L., Loeschke, H. H., Schmidt, C. F.: Comparison of relationship of respiratory minute volume to PCO_2 and pH of arterial and internal jugular blood in normal man during hyperventilation produced by low concentrations of CO_2 at one atmosphere and by O_2 at 3 atmospheres. J. appl. Physiol. **15**, 803 (1953)

Liebegott, G.: Über Organveränderungen bei langer Einwirkung von Sauerstoff mit erhöhtem Partialdruck im Tierexperiment. Beitr. path. Anat. **105**, 413 (1941)

Lieven, T. v., Walther, G.: Zum Nachweis einer Myokardschädigung bei der hyperbaren Oxygenation. Med. Welt **23** (N. F.) 200 (1972)

Lieven, T. v., Walther, G., Maillot, K.: Störungen des Säure-Basen-Haushaltes bei intermittierender hyperbarer Oxygenation der Ratte. Anaesthesist **20**, 98 (1971)

Luft, U. C.: Irreversible hypoxämische Organveränderungen bei alten und jungen Tieren im Unterdruck. Beitr. path. Anat. **99**, 351 (1937)

Malorny, G.: Zum Mechanismus der Sauerstoffvergiftung. Habil.-Schrift, Kiel, 1944

Nolte, H.: Die Sauerstoffintoxikation. Z. prakt. Anaesth. **3**, 280 (1968)

Orzechowski, G.: Neuere Erkenntnisse in der Physiopathologie der Sauerstoffvergiftung. 2. Marinemed. wiss. Sympos., Kiel (Germany) 1968

Pichotka, J.: Über die histologischen Veränderungen der Lunge nach Atmung von hochkonzentriertem Sauerstoff im Experiment. Beitr. path. Anat. **105**, 381 (1941)

Pichotka, J., Kühn, H. A.: Experimentelle und morphologische Untersuchungen zur Sauerstoffvergiftung. Naunyn-Schmiedenbergs Arch. exp. Path. Pharmak. **204**, 336 (1947)

Smith, C. W., Lehan, P. H., Monks, J. J.: Cardiopulmonary manifestations with high O_2 tension at atmospheric pressure. J. appl. Physiol. **18**, 849 (1963)

Szakacs, J. W., Cannon, A.: l-Norepinephrine myocarditis. Amer. J. clin. Path. **30**, 425 (1958)

Weibel, E. R.: Toxische Auswirkungen erhöhter Sauerstoffspannung auf die Lungen. In: Lungenveränderungen bei Langzeitbeatmung; K. Wiemers u. K. L. Scholler, eds. Internat. Sympos., Freiburg 1971. Stuttgart: Thieme 1973

Winter, P. M., Williams, B. T., Roding, B., Schenk, W. G.: Coronary arterity blood flow and oxygen transport under hyperbaric oxygenation. Proc. 4th Intern. Congr. Hyperbaric Medicine, Sapporo, 1969, p. 228

Wood. C. D., Seager, L. D., Perkins, G.: Blood pressure changes and pulmonary edema in the rat associated with hyperbaric oxygenation. Aerospace Med. **36**, 479 (1967)

Dr. C. M. Büsing
Pathologisches Institut
der Universität
D-6900 Heidelberg 1
Im Neuenheimer Feld 220/221
Federal Republic of Germany