THE RELATION OF EXERCISE TO BUBBLE FORMATION IN ANIMALS DECOMPRESSED TO SEA LEVEL FROM HIGH BAROMETRIC PRESSURES*

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Compressed-air illness, often referred to as the "bends" or the "chokes," is a well known clinical entity among divers and others working at high barometric pressures. This condition does not develop during the period of sojourn at increased pressure, but appears on return to atmospheric pressure, if the rate of decompression is excessive. Bert (1878) early showed that animals rapidly decompressed from high pressures to sea level contained bubbles rich in nitrogen in the blood stream, and attributed the symptoms of compressed-air illness to the effects of multiple gaseous emboli. More recently, this interpretation has formed the basis for important research of a practical nature, with emphasis on prevention and treatment of symptoms. Contributions in this direction are especially identified with the efforts of Haldane and coworkers (summarized in Haldane and Priestley, 1935), as well as with those of Behnke and associates (summary, Behnke, 1942). These researches have greatly reduced the danger involved in diving and similar operations and have extended the range of pressures within which work may be carried on.

Recently the problem of decompression sickness has received increased attention in connection with aviation medicine. Symptoms similar to those of compressed-air illness may develop in flyers at high altitudes, and in all probability are associated with bubble formation in blood and tissues (Armstrong, 1939). On the basis of these conclusions we have carried out a series of animal experiments on bubble formation at simulated altitudes. The results of this work are reported separately (Whitaker *et al.* (1945); Harris *et al.* (1945)) and show that muscular activity during decompression is an important causal factor in bubble formation. Additional evidence suggests that this action of exercise at simulated altitudes is mediated through the combined effects of

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mechanical factors[1] and accumulated carbon dioxide, which may attain high local concentrations during muscular activity.

As a result of these findings in animals at simulated altitudes, we were led to investigate the importance of muscular exercise for bubble formation in animals decompressed from high barometric pressures to sea level. On theoretical grounds this is of particular interest since a number of factors affecting bubble formation differ in the two cases. More specifically, one important difference arises from the fact that in animals allowed to equilibrate at high barometric pressures, the total tension of dissolved gases in blood and tissues is much greater than in animals at sea level. Consequently during decompression, the same proportional reduction in pressures (*e.g.* by one-half) will create a higher supersaturation, expressed in gas tensions or numbers of molecules, in the case of the compressed animals. Secondly, the CO_2 tension of blood and tissues is relatively independent of external changes in barometric pressure. At high barometric pressures, therefore, the CO_2 tension is small relative to the external pressure; at low barometric pressures (simulated altitudes) the CO_2 level is much nearer the total external pressure. The significance of this point lies in the fact that at simulated altitudes, an increase of CO_2 during muscular activity may enable the CO_2 tension to rise locally and exceed the external barometric pressure. Under these circumstances the local CO_2 tension would also exceed the internal pressure of bubbles forming in such loci. This is evident since environmental pressure changes are transmitted throughout the body, except in bone. Thus the internal pressure of bubbles in the blood stream is substantially identical with, and follows changes in the external barometric pressure. It is clear that if the total pressure within a bubble is less than the tension of dissolved CO_2 in its immediate neighborhood, growth could occur from this source alone. Actually, however, since the internal pressure of the bubble represents the sum of the partial pressures of contained gases, and since nitrogen and oxygen would also be present within the bubble, the partial pressure of CO_2 inside the bubble would be appreciably less than the total internal pressure of the bubble. Hence the tendency for CO_2 to enter the bubble would be even greater. It is understood, of course, that nitrogen would further augment bubble growth at simulated altitudes, for similar reasons. Local supersaturations of CO_2 built up in the way described above, are conceived to be responsible for the facilitating action of CO_2, not only on growth but also in the initiation of bubbles in animals at simulated altitudes (Harris, Berg, Whitaker, Twitty, and Blinks (1945)).

On the other hand, the importance of CO_2 in the development of bubbles

[1] Harvey (unpublished), Blinks (unpublished), and Dean (1944) have developed the rôle of physical factors in bubble formation. They have shown that in fluids supersaturated with dissolved gases, bubbles do not usually arise *de novo* except in the presence of mechanical agitation.

would appear to be much less in animals decompressed from high barometric pressures to sea level. In this case, the external pressure is much higher, even at the end of decompression (760 mm. at atmospheric pressure) and it is unlikely that CO_2 tensions in the body would ever exceed or reach this level. Considering also that CO_2 would constitute a relatively small fraction of the total dissolved gases in the blood of compressed animals, it appears improbable that CO_2 exerts any great influence on bubble formation in decompression to sea level from increased pressure.

Another point of difference between compressed animals and those at simulated altitudes lies in the complicating effects of anoxia, usually present in experiments at low pressures. In animals decompressed in air from high barometric pressures to sea level, anoxia does not enter the picture. The importance of these differences for an understanding of bubble formation in compressed animals will be evident in the following discussion.

Materials and Methods.—A small cylindrical steel chamber was used for compression and the animals compressed in air throughout. Pressures were regulated by gauge and a small continuous flow of air was maintained through the chamber during all experiments. Decompression from increased pressures was rapid, the pressure drop to sea level occurring in a few seconds. Muscular activity after decompression was induced by intermittent electrical stimulation (5 to 25 volts, 60 cycle A.C.). In bullfrogs, this was applied by placing the animals on a copper wire grid; with rats, electrodes moistened with saline were attached to the hind limbs. In experiments involving anesthesia, nembutal (for rats) and urethane (for bullfrogs) were used. Anesthetics were administered prior to the compression treatment in all cases where given.

Bubble Formation in Compressed Bullfrogs

Effect of Exercise.—Large bullfrogs (*Rana catesbiana*) were compressed in an initial series of experiments to test the effect of exercise, following decompression, on bubble formation. These frogs were all compressed for 1 hour at various selected pressure levels, varying in different experiments from 3 to 60 pounds per square inch.[2] The animals fall into three groups according to the degree of muscular activity on subsequent decompression to sea level. Frogs in the first of these groups were not anesthetized, and were subjected to violent muscular activity immediately after decompression to atmospheric pressure. The exercise, extending over a period of 30 minutes, was violent and maximal, resulting in a state of exhaustion. The animals were then pithed, dissected, and examined carefully for the presence of bubbles under a dissecting binocular microscope.

[2] All pressures given throughout this paper are gauge pressures, in excess of atmospheric pressure. Thus a pressure of 60 pounds per square inch is equivalent to 5 atmospheres absolute pressure.

Animals in the second group likewise were not anesthetized but were not electrically stimulated on return to sea level from increased pressure. Instead, the frogs were allowed to remain quietly in the compression chamber, with slight spontaneous movements, for 30 minutes after reaching sea level. At the end of this time they were pithed and examined as before for bubbles.

TABLE I

Bubble Formation in Bullfrogs on Decompression to Sea Level from High Barometric Pressures

Pressure	No activity (urethanized)	Slight activity (normal, not stimulated)	Violent activity (electrical stimulation)
lbs. per sq. in.			
60	−− −−	+++ +−	++ ++
45	−− −	+++ −	++ +
30	−− −	−− −−	++ ++
15		−−− −−−	++ ++
11		−−	++ ++
8		−− −−	++++ ++++
5		−−	++ ++
3		−−	++− −−

+, bubbles; −, no bubbles. Each symbol represents an individual animal. All frogs compressed for 1 hour.

The third group involved frogs which had been anesthetized with urethane before compression, so that muscular activity was completely lacking, both during the period at increased pressure and on subsequent decompression. After a further interval of 30 minutes at atmospheric pressure, these animals were also dissected and a search made for bubbles.

The results of these three series of experiments are summarized in Table I. Bubbles were present in all frogs of the exercised group which had previously been compressed to 5 pounds or more. In animals pretreated at relatively low pressure levels (5 to 15 pounds), bubbles occurred only in the renal portal veins and ventral abdominal vein, which drain the hind limbs, region of highest muscular activity during exercise. In frogs that had been at higher pressure levels (30 to 60 pounds) bubbles were present in large numbers in the heart and in all veins and arteries. In the second group of frogs, which were unanesthetized and had not been stimulated, but had slight spontaneous movements, bubbles were present in some cases if the animals had been decompressed from relatively high pressures, but not in frogs treated at 30 pounds or less. In the complete absence of muscular activity, as seen in the group of frogs anesthetized with urethane, bubble formation did not occur in frogs pretreated up to 60 pounds, the highest pressures used in these experiments.

Minimum Pressure Treatment.—An additional point of interest concerns the minimum pressure to which bullfrogs must be subjected if bubble formation is to occur when the animals are exercised subsequently at sea level. Two out of five animals pretreated at 3 pounds showed very tiny bubbles in the renal portal veins after prolonged and violent exercise, thus suggesting that the pressure differential of 3 pounds ($\frac{1}{5}$ atmosphere) may represent the approximate threshold level for bubble formation in compressed bullfrogs. In terms of decompression started at sea level, the reduction of pressure from $1\frac{1}{5}$ atmospheres to 1 atmosphere is equivalent to a simulated ascent of 5,000 feet. This threshold pressure differential necessary for bubble formation (with exercise) is lower than the comparable threshold altitude (*ca.* 15,000 feet) in bullfrogs decompressed from sea level (Whitaker, Blinks, Berg, Twitty, and Harris (1945)). That is, a smaller percentage drop in pressure is necessary to cause bubble formation when the decompression starts from higher barometric pressures (above 760 mm.).

Bubble Formation in Compressed Rats

Effect of Exercise.—The results obtained with bullfrogs, as described above, were extended in a further group of experiments with rats. Experimental animals were compressed $1\frac{1}{2}$ hours at various pressure levels, and following a rapid decompression to sea level were subjected to strong muscular activity for 3 minutes. In a few cases, this was accomplished without direct stimulation merely by inducing the animals to engage in active spontaneous movements. In the majority of instances, however, electrodes were attached to the hind limbs and electrical stimulation applied, resulting in activity of a violent type. The animals were then killed, opened, and the vascular system carefully examined under a dissecting binocular microscope. A parallel control series of rats received identical pressure treatment but were not exercised after decompression. Most of these had been anestheized with nembutal before compression, to completely inhibit muscular activity; a few animals were unanesthetized but allowed to remain at rest, or with slight voluntary activity, after decompression. A few of the anesthetized rats were left at atmospheric pressure for 30 to 45 minutes after decompression, but there was no indication that a longer wait before autopsy facilitated the appearance of bubbles in these control animals.

As seen in Table II, bubbles were found in all animals of the exercised group. In rats pretreated at 60 pounds, bubbles were usually present in heart, arteries, and veins; pretreatment at 30 to 45 pounds resulted in bubbles in the veins and right side only of the heart. No extensive study was carried out to determine accurately the lower threshold of pressure necessary for bubble formation in the rat, but each of four rats compressed at 15 pounds after

TABLE II

Bubble Formation in Rats on Decompression to Sea Level from High Barometric Pressures

Pressure	Exercised	Not exercised
lbs. per sq. in.		
60	++	+++
	$+_1+_1$	−−
45	+++	−−−−
	$+_1+_1$	−−−
30	+++++	−−−−
	$++++_1+_1$	⊖⊖⊖⊖
15	++	−−
	++	−−

+, bubbles; −, no bubbles; $_1$, exercised without electrical stimulation. Exercised rats not anesthetized; all controls (non-exercised group) anesthetized except those circled.

exercise at sea level showed a few small bubbles in the right auricle or post-caval vein. In these rats treated at 15 pounds, therefore, a reduction by half in the external pressure (15 pounds to sea level) was sufficient, with exercise, to cause bubble formation. In terms of decompression started from sea level, this pressure drop is equivalent to a simulated altitude of only 18,000 feet. Previous work by our group has shown that living rats decompressed from sea level do not undergo bubble formation at simulated altitudes of less than 45,000 feet (Whitaker, Blinks, Berg, Twitty, and Harris (1945)). It is evident therefore, that in compressed rats, as in compressed bullfrogs, bubbles will form (with exercise) after a lower percentage drop in pressure than is necessary when these animals are decompressed from sea level to simulated altitudes.

The fact that compressed bullfrogs, under conditions of exercise, will bubble more readily than rats (*i.e.* with a lesser pressure treatment) may possibly be related to the more rapid respiratory turnover and elimination of dissolved gases after decompression in the rat. A similar difference between the two forms is seen in decompression from sea level to simulated altitudes.

In the control group of non-exercised rats, as shown in Table II, bubbles were not found in any animals pretreated at 15, 30, or 45 pounds. All of these animals were anesthetized except for a group of four rats treated at 30 pounds as indicated by circles in Table II. These unanesthetized but relatively quiet animals likewise failed to show bubbles, indicating that anesthesia is not in itself the explanation for the negative results in control animals. It is clear, therefore, that in rats pretreated within the 15 to 45 pound pressure range, exercise is demonstrably related to the appearance of bubbles, while in the absence of strong muscular activity no bubbles may be seen at autopsy.

Bubble Formation without Muscular Activity.—By contrast with the foregoing statement, Table II also shows that in rats pretreated at 60 pounds, bubbles were present in both exercised and non-exercised groups. The occurrence of bubbles, however, was more uniform in the exercised rats given this pressure treatment. In the exercised animals which had been at 60 pounds, bubbles were present in all arteries and veins; frequently the large vessels were completely filled with gas. Three out of five anesthetized rats pretreated at 60 pounds contained large numbers of bubbles at autopsy, although muscular activity was completely lacking throughout the experiments. This finding suggests that bubbles may develop in the complete absence of voluntary muscular activity, if the level of supersaturated gases in blood and tissues is sufficiently high.

E. Newton Harvey and associates (unpublished) have likwise found that in some cases bubbles will occur in compressed cats without muscular exercise (animals given nembutal) if the preliminary pressure treatment exceeds 3 to 3.5 atmospheres absolute (30 to 37.5 pounds gauge pressure). In these anestheized animals, however, electrical stimulation of the hind limbs greatly increased the rate at which bubbles appeared. The fact that a lesser pressure treatment (30 to 37.5 pounds) is required for bubbles to form without exercise in cats, as compared to rats (60 pounds) correlates with the difference in size, and previous findings that cats bubble more readily than rats at simulated altitudes.

That bullfrogs anesthetized with urethane did not bubble without muscular activity when pretreated at 60 pounds may reflect an incomplete equilibration at the higher pressure, due to the inefficient lung and ventilation mechanism of the frog.

Effect of Anoxia.—Rats were also employed to test the possible effect of anoxia on bubble formation in compressed animals. Experimental animals were compressed to 30 pounds for 1½ hours, then decompressed to sea level, and immediately placed in a closed chamber, to which nitrogen was added slowly at atmospheric pressure. Administration of nitrogen was increased gradually until, after about 4 minutes, death occurred from anoxia. A moderate amount of muscular activity occurred during this period, particularly just prior to cessation of breathing. No bubbles were observed at autopsy in any of these experimental animals. As a control series, another group of rats was compressed to 30 pounds for $1\frac{1}{2}$ hours, then decompressed to sea level, and immediately subjected to violent muscular exercise for 3 minutes. Bubbles were found at autopsy in all of these animals (see Table III). Thus, bubbles did not appear in the experimental animals with anoxia and even a moderate amount of exercise, although with a slightly higher level of exercise, bubbles were found in all controls. These results indicate that if anoxia has any facilitating effect on bubble formation in compressed animals, it is too slight to be revealed by the present experiments.

TABLE III

Anoxia and Bubble Formation in Compressed Rats

Treatment after decompression to sea level	Degree of exercise	Autopsy
Killed by anoxia	Moderate (spontaneous)	− − − − − −
No anoxic treatment (controls)	Violent (stimulated)	+++ +++

+, bubbles; −, no bubbles. All animals compressed at 30 pounds per square inch for $1\frac{1}{2}$ hours.

DISCUSSION

From the data given in the preceding sections, it is clear that muscular activity has a definite predisposing effect toward bubble formation in rats and frogs decompressed from high barometric pressures to sea level, just as previously found in decompression of these animals from sea level to simulated altitudes (Whitaker, Blinks, Berg, Twitty, and Harris (1945)). This finding is particularly interesting in view of the statement commonly made in textbooks that exercise during decompression is beneficial in preventing compressed-air illness in divers and caisson workers. For example, Haldane and Priestley (1935, p. 352) state that "During decompression, or immediately after it, it is desirable that as much muscular work as possible should be carried out, so as to increase the circulation, and therefore the rate of desaturation ... (in) ... the body." It may be well to re-examine such conclusions in the light of more extensive information on the effects of exercise. The animal experiments described here suggest that the theoretical benefits of exercise on desaturation are definitely outweighed by a facilitating effect on bubble formation.

The results of exercise as reported here also direct attention to another commonly accepted view, namely that bubbles are not liberated under decompression unless the barometric pressure is reduced to one-half or less of its initial value. For human work under increased pressures, this is expressed by Haldane and Priestly (1935, p. 337) as follows, "...there is a complete immunity from symptoms (in divers) ... if the excess of atmospheric pressure does not exceed $1\frac{1}{4}$ atmospheres....Thus bubbles of nitrogen are not liberated within the body unless the supersaturation corresponds to more than a decompression from a total pressure of $2\frac{1}{4}$ atmospheres." While the empirical value of this generalization has been proved abundantly for man in practical operations, it is obvious from the present experiments that under conditions of exercise, bubbles may develop in animals at lesser supersaturations. Thus bubbles occurred in exercised bullfrogs following decompression from a total pressure of only $1\frac{1}{5}$ atmospheres to 1 atmosphere absolute pressure (3 pounds to sea level). Even in rats, where no systematic attempt was made to determine a minimum pressure treatment, bubbles were found on decompression from 2 to 1 atmosphere (15 pounds to sea level), provided the animals were subjected to strong muscular activity.

It seems unlikely that the facilitating effect of muscular activity in the compressed animals is due in any large measure to CO_2. As pointed out earlier, under these conditions CO_2 forms a relatively small fraction of the total dissolved gases in blood and tissues. The high supersaturation of dissolved nitrogen, combined with the mechanical factors involved in muscular exercise (turbulence and similar effects) would appear to be more important here. This does not detract in any way from the important rôle which CO_2 plays for the development of bubbles in animals decompressed from sea level to simulated altitudes, where supersaturation of this gas may occur in connection with muscular activity.

Mention has been made of the fact that a lesser percentage drop in pressure is required to produce bubbles, with exercise, in animals decompressed to sea level from increased pressures, as compared with those at simulated altitudes. In formulating an explanation, it is significant that the same relative drop in pressure results in a higher supersaturation of dissolved gases in the case of the compressed animals, due to the greater molecular concentration in blood and tissues. That an increased molecular concentration may result in a decrease in the degree of decompression necessary for bubble formation is indicated by the results of treating frogs with CO_2 and decompressing them to simulated altitudes (Harris, Berg, Whitaker, Twitty, and Blinks (1945)). Bubble formation in these animals was greatly facilitated at all altitudes, and in addition occurred at altitudes well below the minimum, or threshold altitude at which bubbles will form in untreated frogs. In the case of compressed animals the relatively high level of dissolved N_2 may have a similar action, as expressed in the comparatively low pressure differential necessary for bubble formation. With pressure treatments of greater magnitude, still higher supersaturations of N_2 may reduce the degree of activity required to initiate bubble formation. Thus bubbles occurred in bullfrogs with only slight spontaneous movements after compression at 45 or 60 pounds, although no bubbles appeared under similar conditions of activity if the pressure treatment had been less. Furthermore, bubbles occurred in anesthetized rats pretreated at 60 pounds in the complete absence of voluntary activity. Here presumably the slight mechanical agitation involved in breathing and other vital activities was sufficient to initiate bubble formation. The action of dissolved N_2 in compressed animals, therefore, would appear to be similar in principle to the predisposing influence of CO_2 at simulated altitudes. The effects of the two possibly have a common basis in the facilitation of bubble formation by high molecular concentration in excess of the saturation level.

SUMMARY

1. Bullfrogs (*Rana catesbiana*) and rats have been subjected to high barometric pressures and studied for bubble formation on subsequent decompression to sea level. Pressures varying from 3 to 60 pounds per square inch, in excess of atmospheric pressure, were used.

2. Muscular activity after decompression is necessary for bubble formation in bullfrogs after pressure treatment throughout the above range. Anesthetized frogs remained bubble-free following decompression. Rats compressed at 15 to 45 pounds per square inch likewise did not contain bubbles unless exercised on return to sea level.

3. Bubbles form without voluntary muscular activity in anesthetized rats previously subjected to pressure of 60 pounds per square inch. Small movements involved in breathing and other vital activities are believed sufficient to initiate bubbles in the presence of very high supersaturations of N_2.

4. Bubbles appear (with exercise) in rats previously compressed at 15 pounds per square inch, and in bullfrogs subjected to pressure at levels as low as 3 pounds per square inch above atmospheric pressure. The percentage drop in pressure necessary for bubble formation is less in compressed animals than in those decompressed from sea level to simulated altitudes.

5. The action of exercise on bubble formation in compressed frogs and rats is attributed to mechanical factors associated with muscular activity, combined with the high supersaturation of N_2. CO_2 probably is not greatly involved, since its concentration does not reach supersatuation, as it does at high altitude.

6. Anoxia following decompression from high barometric pressures has no observable facilitating effect on bubble formation.

REFERENCES

Armstrong, H. G., Principles and practice of aviation medicine, Baltimore, The Williams & Wilkins Co., 1939.

Behnke, A. R., Physiologic studies pertaining to deep sea diving and aviation, especially in relation to the fat content and composition of the body, *Harvey Lectures*, 1942, **37,** 198.

Bert, P., La Presson Barometrique, Paris, G. Masson, 1878.

Dean, R. B., The formation of bubbles, *J. Appl. Physics*, 1944, **15,** 446.

Haldane, J. S., and Priestley, J. G., Respiration, Oxford University Press, 1935.

Harris, M., Berg, W. E., Whitaker, D. M., Twitty, V. C., and Blinks, L. R., Carbon dioxide as a facilitating agent in the initiation and growth of bubbles in animals decompressed to simulated altitudes, *J. Gen. Physiol.*, 1945, **28,** 225.

Whitaker, D. M., Blinks, L. R., Berg, W. E., Twitty, V. C., and Harris, M., Muscular activity and bubble formation in animals decompressed to simulated altitudes, *J. Gen. Physiol.*, 1945, **28,** 213.