Experimental Studies on Decompression Sickness

—Consideration about Hypercoagulability of Blood in Decompression Sickness—

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Abstract

Experimental data is presented to support the hypothesis that in an acute stage of decompression sickness with intravascular bubble formation, activation of thrombogenesis may occur.

Platelet aggregation in the vicinity of intravascular air bubbles and occurrence of thromboembolism were seen in the sinusoidal system in the bone marrow of femurs of rabbits which had decompression sickness.

Similar platelets aggregation and thromboemboli were seen in the bone marrow of femurs of experimental rats. Moreover, these rats showed a sharp decrease in the numbers of circulatory platelets.

Related evidence linking these observation to intravascular clotting and thrombus formation is reviewed.

Introduction

Depending upon the autopsy findings, the authors previously described that the most important pathogenetic factor of decompression sickness was intravascular air bubbles associated with activation of the thrombogenesis.

The bone marrow tissue of femurs of the divers died from acute decompression sickness possessed much amounts of air bubbles in the sinusoidal lumina and platelet aggregation and thrombus formation were marked in the vicinity of the bubbles.

In their spinal cords, thrombosis was dominated in the intramedullary and extramedullary vein systems.

Disturbance of blood flow due to bubble embolization together with thrombosis seemed to be the most important for the pathogenesis of the decompression sickness.

The present study was performed in an attempt to provide more direct evidence of activation of thrombogenesis in an acute stage of decompression sickness in experimental animals.

Materials and Methods

(1) Production of decompression sickness in experimental animals.

Ten male Wistar rats, 350-450 grams in weight, were compressed for two hours at five
gauge pressure (six ATA) in the experimental hyperbaric chamber (NAKAMURA), then were quickly decompressed to ambient pressure in five minutes. All of them fell in dyspneutic condition about five minutes after returning to ambient pressure. Some animals coincidently revealed paralytic movements in their posterior limbs. They all expired ten to fifteen minutes after the decompression.

Four rabbits (A-group), weighing 3.5-4 kgs were compressed for six hours at five gauge pressure, then they were decompressed in five minutes to ambient pressure. They all expired five to ten minutes after the finish of decompression under the shock state.

Three rabbits (B-group), weighing 3.5-4 kgs which had been compressed for one hour at the same pressure, were decompressed in five minutes to ambient pressure. They showed some dyspneutic conditions and paralysis in the posterior limbs and expired fifteen to sixty minutes after the decompression.

(2) Hematological examination

From all the rats, blood was aspirated for two times, before the compression-decompression procedures and immediately after the death respectively. Then the platelets were counted.

One animal of A-group, blood was aspirated for two times as same as rats. Platelet counting and thrombelastographical examination were performed on each blood.

Same procedures as mentioned above were also performed on one rabbit of B-group.

(3) Histopathological studies

All the experimental animals were necropsied for the subsequent histopathological studies on various organs. From several rabbits of B-group, the lungs and the femoral bone marrow tissue removed and quickly frozen at -70°C and then paraffin sections were prepared. They were stained by FITC-conjugated anti-rabbit-fibrinogen sheep-IgG.

Results

1. Hematological examinations
   a. Before compression-decompression procedures, the numbers of circulating platelets of the rats were ranging from 515,000/mm³ to 634,000/mm³ and the average was 569,700/mm³.

   Most the animals showed more or less decrease in the numbers of platelets after the procedures and they were ranging from 408,000/mm³ to 566,000/mm³ (average 491,300/mm³). The difference of the numbers of the circulating platelets was highly significant (P<0.01) (Table 1).

   b. The thrombelastograph of one rabbit belonging to A-group showed shortening of k and r and widening of ma. after the compression-decompression procedure, in comparison with the thrombelastograph taken before the procedure (Figs. 1 & 2).

   c. The thrombelastographs of one rabbit of B-group showed no particular changes between the bloods taken before and after the procedure.

   But the number of platelets decreased after the procedure in some value (before: 355,000/mm³ and after: 255,000/mm³).

2. Histopathologic findings

   Numerous, large or small, up to 2000 microns in diameter air bubbles were existent in
the sinusoids of the bone marrows from all the animals. Platelets aggregation was noted in the vicinity of some bubbles (Fig. 4). The thrombus formation was marked around the bubbles in other areas (Fig. 5). Accumulation of fibrinogen substance was demonstrated by application of FITC-conjugated anti-rabbit-fibrinogen sheep-IgG on femoral bone marrow around the bubbles in the sinusoids (Fig. 6). Numerous air bubbles were seen in the blood of the vena cava and the right atrium and ventricle of the heart (Fig. 3) and

<table>
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<th>Rat No.</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
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<td>63.0</td>
<td>52.5</td>
<td>58.8</td>
<td>63.4</td>
<td>51.8</td>
<td>55.8</td>
<td>56.3</td>
<td>60.3</td>
<td>51.5</td>
<td>56.3</td>
<td>56.97</td>
</tr>
<tr>
<td>After 'Compression-Decompression'</td>
<td>51.8</td>
<td>43.1</td>
<td>51.9</td>
<td>54.2</td>
<td>43.7</td>
<td>45.0</td>
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<td>48.8</td>
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<td>51.6</td>
<td>49.13</td>
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F = 1.06 < p < 0.01
Fig. 5: Dilatated sinusoids of the femoral bone marrow (rabbit) including air bubbles (b). Thrombus formation is developed around the bubbles. H, E, × 30.

Fig. 6: Reaction for fibrinogen. Fibrinogen is massively accumulated around the bubbles in a sinusoid of a rabbit femur. FITC-conjugated anti-rabbit-fibrinogen sheep-IgG, × 75.

Fig. 7: Reaction for fibrinogen. Intensely positive reaction is observed in the fine vasculature of the lung (rabbit). FITC-conjugated anti-rabbit-fibrinogen sheep-IgG, × 75.

Discussion

The pathogenesis of the ill effects following decompression can not be ascribed solely to the space occupying and surface tension effects of the air bubbles altering the normal blood flow through the vasculature.

When the blood comes into contact with the foreign surface, such as the interface of blood and air bubbles (blood-bubble interface), the changes in the state of circulating platelets and conversion of fibrinogen to fibrin are appeared.

Hartveit et al. (1968) studied the pathologic changes induced by venous air embolism in the experimental animals. They found that the cause of death was the fibrin plugs in the branches of the pulmonary arteries and that the prior heparinization increased to the survival rate. So they concluded that the death of venous air embolism was due to intravascular clotting.

Hallenbeck et al. (1973) studied on the acceleration of coagulability of the blood by bubbling, and considered that the air bubbles have a capacity to accelerate some hematological reactions through their actions on the cellular elements of blood by the activation of a
plasma factor, presumably Hageman (XI).

Philp et al. (1969) showed that there was a decrease in the numbers of circulating platelets in experimental decompression sickness in rats.

Observations by light and electron microscopy of vessels in experimental decompressed rats have confirmed the presence of a layer of platelets and leukocytes to intravascular air bubbles (Philp et al. 1971, Philp et al. 1972).

Warren (1973) described in detail that some of the ultrastructural morphology of air embolism were seen in decompressed rats. In particular, this included the attachment of platelets and eventual aggregation around the protein at the interface.

Platelet aggregation in the vicinity of the intravascular bubbles and the occurrence of thrombus formation around the bubbles were seen in the sinusoidal system in the bone marrow and in the branches of the pulmonary arteries of the animals used in this study. These findings suggested that the intravascular bubbles act as foreign surfaces to cause denaturation of plasma proteins, platelet adhesion and aggregation. The observation offers an explanation for the post-decompression thrombocytopenia which was observed in the experimental rats.

So, there is now conclusive evidence that a blood-bubble interface contributes to the formation of thrombi and the significance of this observation to the pathogenesis of decompression sickness should be recognized because it presents that the circulatory disturbance would not be caused simply by bubble embolization.

Acknowledgement

The authors are grateful to Emeritus Prof. T. Amako, Director of Kyushu Rosai Hospital for his helpful guidances.

References

質問 九州大学整形外科 杉岡 洋一
人間の減圧症での骨壊死には好発部位が知られてい
るが実験で全身の骨の部位による気泡発生に違いがあ
ったかどうか、御教示ください。

回答 九州労災病院 北野 元生
骨頭では小さい気泡が多数、骨幹部では大きい気泡
が少数みられる傾向がある。 (大腿骨)。
実験動物について FDP の検索は行なっていません。

質問 長崎大学整形外科 齋田 健
FDP の data はどうだったでしょうか。

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