EVALUATION OF STANDARD DECOMPRESSION SCHEDULE BY AGAROSE GEL METHOD

BY

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ABSTRACT

The Standard Decompression Schedule was evaluated by the method of bubble formation in agarose gel, the result of which can be summarized as follows:

- l) The number of bubbles formed in agarose gel corresponded well with the exposed pressure.
- 2) The technique of this method was simple and the number of bubbles was accurately counted.
 - 3) Eventually, this method was useful for examining the decompression schedules.
- 4) It is not always safe to follow the Standard Decmopression Schedule in some pressure conditions.
- 5) As to the period of time that a person is able to tolerate a high pressure condition, the prescription of the Standard Decompression Schedule is not necessarily correct.
- 6) The number of bubbles was small by the proper decompression schedule, for example, in the cases of exposure above the 60-meter depth of water.
- 7) This method can be applied for the prevention of decompression sickness when the agarose gel samples are attached to the workers during the compressed air work.
- 8) The number of bubbles was inconsistent with the coefficient of body pressure (I. N_2 in the body), therefore it is not necessarily safe to rely only on the coefficient of body pressure.
- 9) To prevent osteonecrosis, the Standard Decompression Schedule is not proper, a deeper first stop and slower ascent being recommended.

Introduction

Decompression sickness is mainly classified into two categories, namely, diver's sickness and caisson disease. The former is common in the divers who ascend rapidly to the surface of the sea and the latter is related to the compressed air workers who are engaged in underground shield work or so-called caisson work, taking inefficient decompression procedures. The cause of this sickness is generally considered to be due to the bubble effects and the bubbles originate from the supersaturated gas dissolved

in the blood and other tissue (Mano^{1,2)}; Strauss³⁾).

Robert Boyle suggested a hypothesis concerning decompression sickness in 1670 and Bert⁴⁾ proved it experimentally in 1880 and thereafter these experiments have been succeeded by many researchers to clarify the pathogenesis of this sickness. And the most prominent achievement in this field appeared in 1907 when Haldane et al.⁵⁾ proposed the stage decompression schedule on the basis of the decompression ratio of the gas perfusion theory.

This schedule was modified several times

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to decrease the incidence of decompression sickness. The Standard Decompression Schedule based on the "Haldane ratio principle" was issued in 1961 by the Ministry of Labor of Japan.6) Since then, most of the compressed air workers and divers have been instructed to observe it as prescribed. A decompression ratio of 2:1 was empirically thought to be safe for the divers and compressed air workers in the "Haldane ratio principle", which was originally based on the long experience by the divers who could stay at a water depth of 10 meters as long as they liked. Various models were used to determine the perfusion ratio of the N2 gas into the tissues under different pressure conditions and their half-saturation time ranged from 5 to 120 (or 180) minutes.

But it has been actually impossible to prevent the sickness completely. The main cause of the sickness was regarded to be the result of inadequate application of the schedule (Nashimoto and Mano⁷⁾). Recently, aseptic osteonecrosis, in particular in the articulation, has been reported, its main cause being considered to be due to the excessive decompression (Ohta et al.8); Wade et al.9); Kawashima et al10). The Standard Decompression Schedule was originally designed to prevent "bends", so it contributes insufficiently to the prevention of osteonecrosis. In order to decrease the incidence of this disease, re-examination of the Standard Decompression Schedule is one of the most urgent problems in occupational medicine.

The N_2 gas perfused into the tissues is mostly expirated from the lung by respiration during decompression, while it is partly supersaturated and frothed in the tissues. A stage decompression method must be developed to prevent such bubble formation.

The coefficient of body pressure was de-

fined as the quantity of the N_2 gas remaining in the body at the end of decompression and, therefore, the coefficient of body pressure of 2.0 indicates that the residual N_2 gas is twice as much as that under the normal pressure condition (1 ATA). According to the "Haldane ratio principle", this quantity of N_2 gas is thought to be critical to the incidence of decompression sickness. This also means that the smaller the coefficient of body pressure is, the less is the incidence of decompression sickness.

Yount¹¹⁾ reported that the number of bubbles formed by decompression changed in proportion to the quantity of N_2 gas contained in the bubbles, explaining that the N_2 gas was considered as the total amount of microbubbles formed by decompression. This explanation seems correct but the bubbles were not always produced in proportion to the coefficient of body pressure by the use of the Standard Decompression Schedule in this study.

Most of the decompression schedules are based on the "Haldane ratio principle", such as the U.S. Navy Table, the French Navy Table and so forth. (12,13) Among them, the U.S. Navy Table has been considered to be most extensively usable and most reliable. Recently a new model of decompression table was issued by the R.N.P.L. (Royal Naval Physiological Laboratory) (Hempleman¹⁴); Beckman¹⁵). The "Haldane ratio principle" was based on the "gas perfusion theory" but this new issue was based on the opinion that the gas not only perfused but also diffused into the tissues, therefore the decompression schedules must be discussed from both sides. The important differences of the R.N.P.L. Table from the others are that the first stop is taken at a deeper depth during decompression and the total decompression time is much longer and that the last stop is

taken at the 20-foot depth and is the longest period of all the stops.

The decompression tables are different in each country, especially the French Navy Table being different from that of the Ministry of Labor of the same country, 16) and many discussions on the preference have been made.

In 1976, Mano et al.^{17,18)} closely studied the relationship between the number of bubbles and the incidence of decompression sickness in the salvage works in the Guam Island by the use of the modified decompression table of the U.S. Navy (Model I). They also reported that the Model I would be more reliable than the present U.S. Navy Table or the Japanese Standard Decompression Schedule. The Model I is characterized by the following points: 1) A slower ascent (3 ft/min.) is taken at the lowest 30-foot depth from the bottom and the first stop is taken at a deeper depth; 2) Ten minutes are added to the bottom time of the U.S. Navy Table; and 3) Oxygen should be inhalated for 15 minutes at the 10-foot depth in a diving $bell.^{17,19)}$

The same idea concerning bubble formation was adopted in this study. The bubbles in the tissues originate from the preexisting gas nuclei, and the gas initially diffuse into the gas nuclei in the tissue and then is frothed out by decompression depending on the difference between the inside tension of the gas nuclei and the surrounding tension of the tissue. The larger the ratio of the decompression is, the greater is the increase of the bubbles in number and in size as well. Then the multiple bubbles unite together to form a larger bubble. The size of the bubbles are usually very fine at the time of cavitation, being about 1.35 μ m in the smallest diameter (Yount²⁰⁾). By increasing the size, they unite to make silent bubbles with a diameter of about $100 \,\mu\text{m}$ and are detected by the Doppler method (Evance²¹⁾). They further increase in size sufficiently to cause embolism in the vessel or oppression of the other tisues, which is the sign of "bends".

Considering the process of bubble formation in the tissue, the risk of decompression sickness will be shown by the number of bubbles originating from the gas nuclei, therefore, the danger of "bends" as well as the reliability of decompression schedules can be judged from the number of bubbles in the agarose gel samples attached to the divers and subjected to the actual decompression schedule. Thus, the onset of decompression sickness can be detected at a comparatively early stage of decompression, before the "bends" appear, if the correlation between them can be determined.

MATERIALS AND METHODS

The pressure vessel (Fig. 1) is composed of a plexiglass cylinder with a pressure guage, an adapter to the gas cylinder, a safety valve and an air exit for decompression. The glass counting cell (Fig. 2) is rectanglar and its inside dimension is $15 \times 6 \,\mathrm{mm}$ and provided with a plexiglass holder. There are two horizontal red lines on the front wall of the cells, each line being 4 and 3 mm from the bottom of each

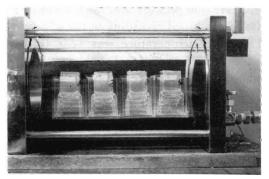


Fig. 1. Apparatus for the Experiment

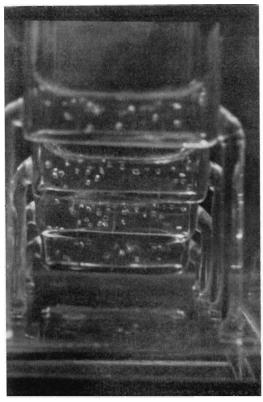


Fig. 2. Counting Cells and Agarose Gel Subjected to the Decompression Schedule

cell. They were filled with agarose solution to a depth of 4 mm (the upper red line). After gelation, these agarose samples were exposed to a predetermined pressure for a predetermined period of time. As the purpose of this experiment was to evaluate the reliability of the Standard Decompression Schedule, the pressure and the bottom time were determined according to the schedule. The room temperature was kept in the range of 19°C to 22°C throughout the experiment. The bubbles formed 3 mm from the bottom (below the lower red line) were counted, and the total volume of the gel amounted to 0.2 7ml.

A stock solution containing 0.5 mM of HEPES (N-2-hydroxyethylpiperazine-N'-2-ethanesulphonic acid, pKa 7.55) buffer and

0.5 mM of MES [2-(N-morpholino)-ethane-sulphonic acid, pKa 6.15] was prepared first. The pH was adjusted to 7.4 in each experiment by adding a small quantity of NaOH. The buffered solution was heated to 80°C, and thereafter a highly purified agarose powder [by Bio-Rad Lot. No. 16320 (moisture content=4.0%, sulfur content <0.1%] was added to the solution (0.5% W/W) and agitated carefully for 10-minutes. The agarose solution was then poured into the counting cells previously made acid-free and subjected to the experiments.

RESULTS

Table 1 shows the number of bubbles in the agarose gel samples during and after decompression which were pressurized and decompressed according to the 23 different pressure schedules chosen from the Standard Decompression Schedule (Table 2), covering the pressure range from the 10-meter to the 90-meter depth of water and the bottom time range from 5 to 360 minutes. The number of samples used in each pressure schedule were 13 to 16, thus totaling 335 cells and 29 cells in the control in this study.

A) Results by 10-meter depth exposure

Two different schedules of decompression were examined under *this pressure*, namely, by the 120-minute bottom time exposure (the longest bottom time for nonstop decompression) and by the 240-minute bottom time exposure. The number of bubbles by the 120-minute bottom time exposure was 5.2 ± 2.6 (S.D.; S.E.=0.7) after nonstop decompression and its coefficient of body pressure was 1.6 and that by the 240-minute bottom time exposure was 2.3 ± 1.4 at the first stop and finally 10.8 ± 4.4 at the end of decompression and its coefficient of body pressure was 1.9, respectively.

Table 1. The number of bubbles in 23 schedules

Depth	of Water	24 m	21 m	18 m	15 m	12 m	9 m	6 m	3 m	0 m	
10 m	120 min									5.9 ±2.6	1.6
10 111	240 min	-							2.3 ± 1.4	$10,8 \\ \pm 4.3$	1.9
20 m	50 min									4.5 ± 2.8	1.5
	115 min	-						8.3 ± 1.9	14.5 ± 2.9	15.1 ± 3.1	1.9
	225 min	-						$18.5 \\ \pm 3.6$	21.2	21.8 ± 3.6	2.2
90	25 min									7.2 ± 3.5	1.5
	40 min								11.4 ± 3.3	15.3	1.7
30 m	78 min							13.3 ± 5.6	$16.1 \\ \pm 4.5$	17.1 ± 4.6	1.9
	130 min	-					$28.9 \\ \pm 8.8$	31.1	31.4	30.9 ± 7.8	2.2
40 m	15 min	-								7.3 ± 3.0	1.4
	46 min							$14, 3 \\ \pm 6.6$	15.9 ± 6.2	18.6	1.9
	100 min					10.6 ± 4.7	11.6 ± 4.9	12.2 ± 4.7	12.6	12.9	2.0
	8 min								0.5 $(0-2)$	0.8 ± 0.3	1.3
50 m	33 min							5.4 ± 2.7	5.9 ± 2.8	6.4 ± 2.9	1.8
	80 min				$13.8 \pm 11.$	$ \begin{array}{c} 18.4 \\ 4 \pm 11. \end{array} $		$ \begin{array}{c} 19.5 \\ 9 \pm 10. \end{array} $	$ \begin{array}{c} 20.4 \\ 5 \pm 11. \end{array} $	$ \begin{array}{c} 20.9 \\ 0 \pm 10.8 \end{array} $	2.0
60 m	5 min									0.3 (0-2)	1.3
00 III	70 min		1.1 (0-4)	1.6 (0-5)	2.1 (0-9)	2.4 (0-11)	2.4 (0-10)	2.3 (0-10)	2.5 (0-10)	2.6 (0-10)	1.9
70 m	5 min								0.1 $(0-1)$	0.3 (0-2)	1.4
	60 min	1.6 (0-7)	2.1 $(0-9)$	2.3 (0-9)	2.8 (0-9)	2.9 (0-10)	2.8 (0-10)	2.9 (0-10)	3.0 (0-11)	3.1 (0-10)	1.8
80 m	5 min		. ,	, ,				. ,	0.3 (0-2)	0.4 (0-2)	1.5
00 III	50 min	0.3 (0-2)	0.4 $(0-2)$	0.5 (0-2)	0.5 (0-2)	0.5 (0-2)	0.6 (0-2)	0.6 (0-2)	0.6 (0-2)	0.6 (0-3)	1.8
90 m	5 min	. ,	, ,	. ,	. ,	. ,	- ,	. ,	0	0.1 (0-1)	1.5
90 III	40 min	1.2 (0-4)	1.9 (0-6)	2.7 ± 2.1	2.8 ±2.0	$\begin{array}{c} 3.2 \\ \pm 2.1 \end{array}$	3.3 ± 2.2	4.1 ± 2.5	$\begin{array}{c} 4.1 \\ \pm 2.6 \end{array}$	4.5	1.8

Bottom time

Coeff. of Body Pressure

B) Results by 20-meter depth exposure

Three different schedules of decompression were examined under *this pressure*, namely, by the 50-minute bottom time exposure (the longest bottom time for nonstop decompression), 225-minute bottom time exposure (the maximum bottom time

during which a person can tolerate this pressure exposure by the regulation and 115-minute bottom time exposure (the median time of the former two bottom times). The number of bubbles by the 50-minute bottom time exposure was 4.5 ± 2.8 after nonstop decompression and its coeffi-

Table 2. Portion of Japanese Standard Decompression Schedule

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			Decompression time (min)						
Pressure	Bottom time		2.5ATA	2.2ATA	1.9ATA	1.6ATA	1. 3ATA	Coefficient of Body Pressure	
2.8-3.0	120 m	in-150 min	_			9	20	2.	
ATA	150	-180	_	_	_	11	30	2.	
	180	-210	_	_	_	15	35	2.5	
	210	-240		_		20	40	2.	
	240	-270	_		_	25	45	2.	
3.0-3.2		30					1	1.	
ATA	30	- 60	_	_	_	_	11	1.	
	60	- 90				8	15	1.9	
	90	-120	_	_	_	12	20	2.0	
	120	-150		_		15	30	2.	
	150	-180	_	_	_	23	40	2.	
	180	-210		_	_	25	45	2.2	
	210	-240	_	_	_	30	50	2.2	
	240	-270	_			31	60	2.2	
3.2-3.4		30							
3. 2-3. 4 ATA	30	- 60	_			_	1	1.4	
AIA	60	- 90 - 90	_	_	_	9	14	1.7	
	90	- 90 -120	_				20	1.9	
			_			18	30	2.0	
	120	-150			_	26	35	2.	
	150	-180	_		5	30	40	2.2	
	180	-210		_	10	30	50	2.2	
	210	-240			14	30	60	2.2	
3.4-3.6		30	_	-	_	_	1	1.5	
ATA	30	- 60	_	_	_	_	20	1.8	
	60	- 90	_	_	_	13	25	2.0	
	90	-120	_	_		19	35	2.	
	120	-150	_	_	_	33	45	2.	
	150	-180	_	_	6	35	50	2.2	
	180	-210	_	_	15	35	55	2.2	
	210	-240	_		18	35	65	2.2	
3.6-3.8		30	_	_	_	-	2	1.5	
ATA	30	- 60		_	_	10	15	1.3	
	60	- 90	_	_	_	17	30	2.0	
	90	-120	_	_	9	25	35	2.	
	120	-150	_		15	30	45	2.5	
	150	-180	_	_	16	35	55	2.2	
	180	-210			21	40	60	2.2	
3.8-4.0		15	_	_	_	_	2	1.3	
ATA	15	- 30	_	_	_		5	1.	
	30	- 45	_	_	_	3	15	1.	
	45	- 60	_	_	_	13	20	1.9	
	60	- 75	_	_	_	18	30	2.0	
	75	- 90	_	_	4	20	40	2.0	
	90	-105	_		11	25	40	2.	
	105	-120		_	13	30	45	2.	
	120	-135		_	15	35	45	2.5	
	135	-150	_	_	18	35	50	2.2	
	150	-165	_	_	23	35	55	2.2	
	165	-180	_	_	20	40	60	2.2	
	180	-195	_		24	40	65	2.2	
	195	-210			26	40	75	2.5	

cient of body pressure was 1.5, that by the median bottom time exposure was 8.3±1.9 at the first stop, 14.5 ± 3.0 at the second and finally 15.1±3.1 at the end of decompression and its coefficient of body pressure was 1.9, and that by the 225-minute bottom time exposure was 18.5±3.6 at the first stop, 21.2 ± 3.6 at the second and finally 21.8 ± 3.6 at the end of decompression and its coefficient of body pressure was 2.2. The most efficient result of this group was determined by the 225-minute bottom time exposure, i.e. the number of bubbles reached already as much as 18.5 ± 3.4 at the first stop. This means that the pressure ratio of the examining schedule $(P_{max} - P_1/P_{max} - P_0)$ was too large at least in this part (Fig. 3).

C) Results by 30-meter depth exposure

Four different schedules were examined under *this pressure*, their bottom times being 25 minute, 78 minute, 130 minute and

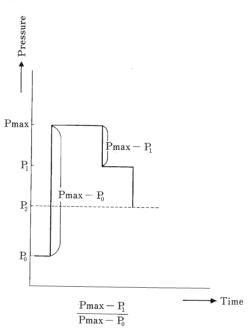


Fig. 3. Decompression ratio

P_{max}; maximum pressure

P1; pressure at the first stop

 P_0 ; pressure at the beginning and at the end of decompression

40 minute, respectively. The last schedule was adopted in Mano's study,17) in which the decompression schedule of the various countries were compared, and it was adopted again in this study to compare the results of this study with that of Mano's, because the nature of the agarose gel used in this study was different from that of Mano's, and it was already known that the number of bubbles formed in the agarose gel would change markedly with the nature of the agarose gel used.17) The number of bubbles by the 40-minute bottom time exposure was 11.4±3.3 at the first stop and finally 15.3±3.9 at the end of decompression and its coefficient of body pressure was 1.7, that by the 25-minute bottom time exposure 7.2±3.5 after nonstop decompression and its coefficient of body pressure 1.5, that by the 78-minute bottom time exposure 13.3 ± 5.6 at the first stop, 16.1 ± 4.5 at the second and finally 17.1±4.6 at the end of decompression and its coefficient of body pressure 1.9, and that by the maximum of 130-minute bottom time exposure 28.9±8.8 at the first stop, 31.1 ± 7.8 at the second, 31.4 ± 7.8 at the third and 30.7 ± 7.8 at the end of decompression and its coefficient of body prsesure 2.2. The last figures were the largest throughout this study and the number of bubbles was also the largest. Thus the number of bubbles and its coefficient of body pressure increased correspondingly with the increase of the bottom time in this group. The number of bubbles formed by decompression was already as much as 28.8 ±8.8 at the first stop and it did not increase so markedly in the following decompression in this group. These results suggest that the Standard Decompression Schedule should be modified at least in this part. While the last part of the decompression schedule between 9 to 0 meter is almost applicable, the nonstop decompression between from the 30-meter to the 9-meter depth must be taken into account and a more prolonged decompression time is required. The final number of bubbles at the 30-meter depth-40-minute bottom time exposure was 15.3±3.9 in this study and that of the above-mentioned Mano's work was 109.0±6.7, showing that the former was one-seventh of the latter. The number of bubbles in the same decompression schedule of R.N.P.L. (Royal Naval Physiological Laboratory) Table amounted to 74.9±5.7 in Mano's work, which corresponds to 10.5 ± 3.2 of this study. The R.N.P.L. Table is considered to be the most reliable in the world, so the number of bubbles of 10.5± 3.2 is regarded as the actual safety level against the incidence of decompression sickness. Accordingly, the part of the Standard Decompression Schedule by the 30meter depth 130-minute bottom time exposure should be modified.

D) Results by 40-meter depth exposure

Three different schedules were examined under this pressure, their bottom times being 15 minute, 46 minute and 100 minute, respectively. The number of bubbles by the 15-minute bottom time exposure was 7.3±3.0 after nonstop decompression and its coefficient of body pressure was 1.4, that by the 46-minute bottom time exposure was 14.3 ± 6.6 at the first stop, 15.9 ± 6.2 at the second and finally 18.6±5.6 at the end of decompression and its coefficient of body pressure was 1.9, and that by the 100-minute bottom time exposure was 10.6±4.7 at the first stop, 11.6 ± 4.9 at the second, 12.2 ± 4.7 at the third, 12.6 ± 4.6 at the fourth and finally 12.9±4.5 at the end of decompression and its coefficient of body pressure was

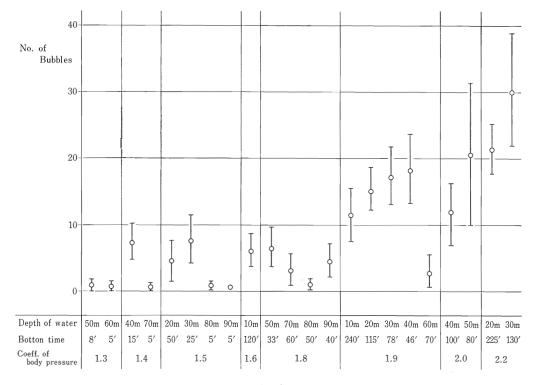


Fig. 4.

2.0. Thus, the number of bubbles increased gradually according to the stage of decompression. But the number of bubbles by the 46-minute bottom time exposure was significantly larger than that by the 100-minute bottom time exposure (p<0.05), although the coefficient of body pressure of the two schedules was the same.

E) Results by the 50-meter depth exposure
The nonstop decompression schedule is
not applicable in the case of exposure to
the pressure at above the 40-meter depth
of water (5 ATA) in the Standard Decompression Schedule.

Three different schedules were also examined under *this pressure*, their bottom times being 8 minute, 33 minute and 80 minute, respectively. The number of bubbles by the 8 minute bottom time exposure was 0.5 (0 to 2) at the 3-meter depth stop for 2 minutes and finally 0.8 (0 to 3) at the end of decompression and its coefficient of body pressure was 1.3, that by the 33-minute bottom time exposure was 5.4 ± 2.7 at the first stop, 5.9 ± 2.7 at the second and finally $6.4\pm$

2.9 at the end of decompression and its coefficient of body pressure was 1.8 and that by the 80-minute bottom time exposure was 13.8 ± 11.4 at the first stop, 18.4 ± 10.9 at the second, 18.8 ± 10.9 at the third, 19.5 ± 10.5 at the fourth, 20.4 ± 11.0 at the fifth and finally 20.9±10.8 at the end of decompression and its coefficient of body pressure was 2.0. The number of bubbles in this group showed a slight increase but it was significantly negligible in the part from the second to the final stop. The total decompression time by the 80-minute bottom time exposure amounted to 180 minute which was 2.25 times as compared with the bottom time. The number of bubbles was fairly large in this group and this increase in the number of bubbles took place mainly during the first and the second stop. Therefore, a more prolonged and deeper first stop is recommended.

F) Results in the range from the 60-meter to the 90-meter depth exposure

Two different schedules were examined by each pressure exposure at more than the

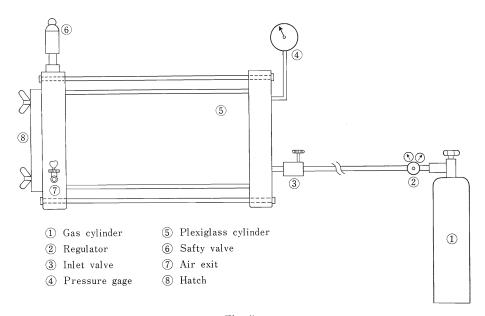


Fig. 5.

60-meter depth, namely, the minimum bottom time of 5 minute and the maximum one of the each depth pressure. The final number of bubbles at the end of decompression was less than 5.0 in all of the decompression schedules by the 60-meter to 90 meter depth pressure.

By the 60-meter depth exposure, the number of bubbles by the 5-minute bottom time exposure was 0.3 (1 to 2) at the end of decompression and its coefficient of body pressure was 1.3 and that by the 70-minute bottom time exposure was 1.1 (0 to 4) at the first stop and finally 2.6 (0 to 10) at the end of decompression and its coefficient of body pressure was 1.8.

By the 70-meter depth exposure, the number of bubbles by the 5-minute bottom time exposure was 0.3 (0 to 1), which corresponded to that by the 60-meter depth-5-minute bottom time exposure, and its coefficient of body pressure was 1.4 and that by the maximum 60-minute bottom time exposure was 1.6 (0 to 7) at the first stop and finally 3.1±2.3 at the end of decompression and its coefficient of body pressure was 1.8.

The results by the 80-meter depth exposure were noteworthy. The number of bubbles by the 5-minute bottom time exposure was 0.4 (0 to 2) at the end of decompression and its coefficient of body pressure was 1.5 and that by the maximum 50minute bottom time exposure was 0.3 (0 to 2) at the first stop and finally 0.6 (0 to 3) at the end of decompression and its coefficient of body pressure was 1.8. The number of bubbles in this series was all below 1.0 and the coefficient of body pressure was 1.8 which corresponded to that by the 70-meter depth-60-minute bottom time exposure with the number of bubbles being 3.1±2.3. The number of bubbles by this pressure exposure was significantly less (p <0.05) than that by the 10-meter depth-120-minute bottom time exposure (5.8 \pm 2.6) with the coefficient of body pressure of 1.6.

By the 90-meter depth exposure also, we reached the same conclusion as by the 80-meter depth exposure. The number of bubbles by the 90-meter depth-40-minute bottom time exposure was 4.5±2.6 which was significantly larger (p<0.01) than that by the 80-meter depth-50-minute bottom time exposure, but the coefficient of body pressure of these two schedules was of the same level of 1.8.

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As mentioned above, the results of the exposure at above the 60-meter depth were all below 5.0, which was far below that of the R.N.P.L. Table, therefore the Standard Decompression Schedule was proved appropriate at least in the decompression schedules at above the 60-meter depth. But the columns of the decompression schedules should be modified below for the 50-meter depth exposure, especially the columns of the long bottom time.

DISCUSSION

Many attempts to detect the "silent bubbles" were carried out in order to foreknow the incidence of decompression sickness before it occurs. For example, Buckles^{22,23)} contrived a method of using laser beams for detecting the bubbles in the blood and Smith²⁴⁾ and Spencer²⁵⁾ invented an apparatus harnessing the Doppler phenomenon of the ultrasonic waves, in which a receiving set of ultrasonic waves transferred into sounds is equipped and the bubbles are identified by the apparatus.

But it is impossible to detect the number of bubbles directly either by laser beams or by ultrasonic waves. These are suitable to determine the individual difference in each exposure, but it is difficult for each worker to use the apparatus and to become skilled to identify the difference in the sounds under various conditions (Evance²¹⁾).

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The method of this experiment was initially contrived by Le Messurier26) who made a report entitled "Preformed Nuclei in the Etiology of Decompression Sickness", emphasizing that gelatin was the most efficient for bubble formation in the study of the etiology of decompression sickness. Thereafter, Beckman¹⁵⁾ and Yount et al.²⁷⁾ proved the efficiency of this method. D'Arrigo,28) one of their colleagues, reported on the improved method of bubble formation in the agarose gel. Unlike the gelatin composed of a complex mixture of charged peptide chain, agarose is an uncharged, relatively inert and homogeneous polysaccharide and is more suitable for the experiment on bubble formation in the gel (Veis²⁹⁾). Mano³⁰⁾ suggested that the agarose gel method was useful for studying the etiology of decompression sickness. Although this agarose gel method is based merely on a physiological phenomenon, it can be applied also to the clinical cases. The incidence of decompression sickness is not only influenced by the physiological changes in the surroundings but also by the peculiarities and the body condition of the individual workers. In other words, the risk of becoming sick is different according to the person even when exposed to the same pressure condition. The number of bubbles was quite different, nevertheless the samples were all prepared with a homogeneous agarose solution pipetted into the counting cells with the same shape and pressurized under the same pressure condition (D'Arrigo²⁸⁾; Mano and D'Arrigo^{17,30)}). And the number of bubbles was also different among the agarose gel samples of different lots., even if they were manufactured in the same factory by the same processes.

Although some skilled techniques are required for making agarose gel samples, the counting cells are small in size and portable, and moreover it is easy for the users to count the number of bubbles correctly (Veis²⁹⁾). But it is impossible to make the individual differences and daily changes of the body condition clear by this method which covers the physical phenomenon only, the homeostasis of the organism not being taken into account.

Assuming that $N_{0.5\%}$ shows the number of bubbles in a decompression schedule with the incidence of decompreision sickness rating of 0.5%, and the number of bubbles in an unknown decompression schedule of less than $N_{0.5\%}$, the incidence of decompression sickness in that schedule can be considered to be less than 0.5%, and in case the number of bubbles is over $N_{0.5\%}$, the incidence is presumed to be more than 9.5%. Namely, if $N_{\text{X}\%}$ (such as $N_{\text{0.5}\%},~N_{\text{1}\%},$ $N_{10\%}$, $N_{50\%}$, $N_{100\%}$ and so forth) shows that the incidence of decompression sickness is X% in that schedule, $N_{x\%}$ can be an indicator of the incidence of decompression sickness, by which a counterplan to prevent the onset of decompression sickness can be taken before the appearance of the symptoms of the sickness.

The reliability of the Standard Decompression Schedule has been discussed only through the practical use of the table without an accurate and theoretical basis. The incidence rate of decompression sickness in the divers and compressed air workers in Japan has been reported to be 0.46% by Nashimoto et al.,31) 3 to 5% by Mann et al.32,33,34) and 4.5 to 28% by Morita et al.35,36) Such a difference in the incidence might have resulted from the misuse of the table and it might be impossible to prevent the sickness completely even if divers might adhere consistently to the table. From these

facts, the Standard Decompression Schedule, especially the columns in the decompression schedules for the 10-meter depth to the 50-meter depth exposure, should be corrected.

It is generally considered that as the coefficient of body pressure becomes higher, the number of bubbles increase. And it seemed in this study also that the greater the coefficient of body pressure was, the larger became the number of bubbles, but the latter did not change in proportion to the former in some schedules.

If the coefficient of body pressure shows correctly the quantity of N_2 gas remaining at the end of decompression, all the decompression schedules of the Standard Decompression Schedule is correct, because it is entirely within the range of 1.1 to 2.2 (Nashimoto and Mano⁷). It is also presumed that the smaller the coefficient of body pressure is, the less is the incidence of the sickness.

As to the relationship between the quantity of the N₂ gas in the body and the number of bubbles formed by decompression, Yount explained that when the preexisting gas nuclei-forming microbubbles grew larger and were united together to form larger bubbles, the N2 gas remained in proportion to the number of bubbles at the end of decompression. If this explanation proves true, the smaller coefficient of body pressure corresponds to the smaller number of bubbles. And in different decompression schedules, the same coefficient of body pressure would show the same number of bubbles at the end of decompression.

But in this study, the number of bubbles by the 40-meter depth-15-minute bottom time exposure was 7.3±3.0 at the end of decompression and that by the 70-meter depth-5-minute bottom time exposure was

0.3 (0 to 2) at the end of decompression, while their coefficient of body pressure was the same of 1.4. No interrelationship was seen in the number of bubbles of these two decompression schedules.

In the four decompression schedules, when the coefficient of body pressure was 1.5, the number of bubbles at the end of decompression was 4.5 ± 2.8 by the 20-meter depth-50-minute bottom time exposure, 7.2 ±3.5 by the 30-meter depth-25-minute bottom time exposure, 0.4 (0 to 2) by the 80-meter depth-5-minute bottom time exposure and 0.1 (0 to 1) by the 90-meter depth 5-minute bottom time exposure. The number of bubbles in the latter two exposure cases was different from that of the former two (p<0.01).

In the other four decompression schedules, when the coefficient of body pressure was 1.8, the number of bubbles at the end of decompression was 6.4±2.9 by the 20-meter depth-50-minute bottom time exposure, 3.1±2.3 by the 70-meter depth-60-minute bottom time exposure, 0.6 (0 to 3) by the 80-meter depth-50-minute bottom time exposure and 4.5±2.6 by the 90-meter depth-40-minute bottom time exposure and no interrelationship was seen.

In the five decompression schedules, when the coefficient of body pressure was 1.9, the number of bubbles was 10.8 ± 4.4 by the 10-meter depth-240-minute bottom time exposure, 15.1 ± 3.1 by the 20-meter depth-115-minute bottom time exposure, 17.1 ± 4.6 by the 30-meter depth-78-minute bottom time exposure, 18.6 ± 5.6 by the 40-meter depth-46-minute bottom time exposure and 2.6 (0 to 10) by the 60-meter depth-70-minute bottom time exposure.

In this group, the number of bubbles was similar except at the level of the 60-meter depth-70-minute bottom time exposure, but the difference seemed large.

In the two decompression schedules, when the coefficient of body pressure was 2.0, the number of bubbles was 12.9±4.5 by the 40-meter depth-100-minute bottom time exposure and ,20.9±10.8 by the 50-meter depth-80-minute bottom time exposure. These levels could hardly be concluded to be identical.

Lastly, in the two decompression schedules, when the coefficient of body pressure was a maximum of 2.2, the number of bubbles was 21.8±3.6 by the 20-meter depth-225-minute bottom time exposure and 30.7±7.8 by the 30-meter depth-130-minute bottom time exposure. These levels were the highest throughout this study, but they were different from each other, while the coefficient of body pressure was the same.

It must be determined that these inconsistencies between the number of bubbles and the coefficient of body pressure might be attributed either to the bubble formation method adopted in this study or to the "Haldane ratio principle" itself, or to the calculation of the coefficient of body pressure. The difference in the number of bubbles in the decompression schedules having the same coefficient of body pressure may be attributable to the calculation of the coefficient of body pressure, because the coefficient of body pressure was calculated from the "120-minute half-saturation time ratio of the tissues", without any experimental basis.

As mentioned above, Mano and D'Arrigo¹⁷⁾ examined the decompression tables of the various countries using agarose gel. They examined the decompression schedule by the 30-meter depth-40-minute bottom time exposure in each table. The number of bubbles was 109.6±6.9 in their work which would be equal to 15.9±3.9 in this study and the coefficient of body pressure was 1.7. If the number of bubbles was de-

termined only by the exposed pressure and bottom time, the maximum number of bubbles of 30.7±3.8 in this work would be equal to 218.7±14.0 in their work, this result would be showing that the Standard Decompression was inadequate for the divers and compressed air workers. If the application of the decompression schedule is safe for the divers, a shorter total decompression time is more advantageous for them. If the uppermost limit of the coefficient of body pressure for safety diving is 2.2, the decompression schedule with the coefficient of body pressure under that level requires a shorter decompression time, and a longer decompression time would be required in a decompression schedule with a coefficient of body pressure above that level, and all the decompression schedules with the same coefficient of body pressure would involve the same risk of decompression sickness.

If the coefficient of body pressure indicates correctly the residue of N2 gas after decompression, it can be calculated from the exposed pressure and the bottom time, and it may also be calculated from the number of bubbles in the agarose gel samples which are exposed to the same pressure condition. Decompression sickness is caused not only by the change in the atmospheric pressure but also by the reaction of the human body to the changes of the exposed pressure. If the incidence of the sickness is chiefly influenced by the coefficient of body pressure of the decompression schedule only, the coefficient of body pressure must be the same in all decompression schedules. Therefore, it is improper that the decompression schedules of the various countries have multiple coefficients of body pressure. The number of bubbles was significantly different from each other among those decompression schedules with the same coefficient of body pressure. This result shows that the calculation of the coefficient of body pressure is not necessarily accurate. In other words, those tables which are based on the "Haldane ratio principle", such as the Japanese Standard Decompression Schedule, the U.S. Navy Table, the French Navy Table and so forth, must be discussed from the following points:

- 1) Replanning of the decompression schedule as it seems to be improper, and of
- 2) The procedures for the calculation of the coefficient of body pressure.

According to Yount,111 the "Haldane ratio principle" itself has to be modified, and those tables based on that principle must also be re-examined. The number of bubbles exceeding 10.0 at the first stop would not increase, when the coefficient of body pressure is above 2.0, in those decompression schedules such as by the 20-meter depth-225-minute bottom time exposure, 30-meter depth-130-minute bottom time exposure, 40-meter depth-100-minute bottom time exposure and 50-meter depth-80minute bottom time exposure. This result shows that the microbubbles were readily formed before the first stop, because of the too large decompression ratio. It is recommended to take the first deeper stop and to ascend more slowly in order to prevent the formation of the microbubbles. The decompression ratio of 2:1 (adopted by J. S. Haldane) proved still large, as microbubbles were formed in the tissue lowering the tissue pressure and looking to take the balance of the pressure between the tissues and the environment. The pressure gap between the tissues and environments may be narrowed by the formation of microbubbles but it may cause osteonecrosis in the future. The cause of this disease was first regarded as a result of neglecting the decompression table, but it can not be neglected that the table itself contains some inadequate decompression schedules. Kawashima et al.¹¹⁾ reported that the incidence of osteonecrosis was 26.9% (72/268) in the divers who had never suffered from "bends". These divers were not considered to have a particular constitution of being insusceptible to "bends" even when they rose up to the water surface rapidly. Usually Japanese divers do not take sufficient decompression time and do not adhere to the decompression table faithfully for economical reasons or sometimes because of the social background of neglecting to control their health but only such reasons will not always be attributable to the high incidence of the disease (38% to 67.7%) (Kawashima et al.10)).

By the 30-meter depth-130-minute bottom time exposure, the number of bubbles was \bar{x} =28.9 at the first stop (nonstop decompression to the 9-meter depth) and it increased to 31.4 at the 3-meter depth stop, and eventually it was \bar{x} =30.7 at the end of decompression, which was not likely to increase but to decrease in number. It is most important for the prevention of decompression sickness, including osteonecrosis, to suppress the number of bubbles to the lowest possible level.

Mano et al.^{1,37)} reported that the number of bubbles was $\bar{\mathbf{x}}$ =61.1±1.1 by the 100-feet (30-meter) depth-40-minute bottom time exposure in their study with the Model I table, where the same ascending speed of the U.S. Navy Table was taken at the beginning 30-feet (9-meter) but a slower speed of 3 ft/min. was taken after that depth till the first stop. This number of bubbles was for 8.6 ± 0.6 in this study, which was less than 15.3 ± 3.9 of the Standard Decompression Schedule, thus proving that the Model I was more reliable than the Standard De-

compression Schedule.

Bubble formation with agarose gel provides a useful method of examining the decompression schedules because the incidence of decompression sickness including osteonecrosis may be decreased.

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