REDUCED FREQUENCY OF CHROMOSOMAL ABERRATIONS AFTER A COURSE OF HYPERBARIC OXYGENATION

A. N. Chebotarev, V. I. Pakhomov, and P. T. Dubynin UDC 616-018.1-092:575.224.23]-02:615.835.12

KEY WORDS: oxygenation; chromosomal aberrations; hyperbaric oxygenation

A series of experimental studies of the mutagenic properties of hyperbaric oxygenation (HBO) has recently been published [1, 2]. It has been suggested that the mutagenic effect of high concentrations of hyperbaric oxygen is linked with a disturbance of the peroxidation system, leading to a raised concentration of peroxide radicals, which have been shown to possess mutagenic properties [8, 10]. However, information in the literature on the mutagenic action of oxygen is contradictory [7]. These contradictions may be connected with differences in the conditions under which experiments with HBO have been conducted. The study of factors leading to manifestation of the mutagenic or, conversely, the protective action of HBO is therefore interesting both from the practical point of view and also in connection with the explanation of the mechanisms of action of hyperbaric oxygen.

In the investigation described below the action of therapeutic doses of HBO on the frequency of chromosomal aberrations in human lymphocytes was studied.

EXPERIMENTAL METHOD

Courses of HBO were given to 7 patients with various diseases and also to 3 clinically healthy volunteers. The patients included 4 with ischemic heart disease, 1 with central carcinoma of the lungs, 1 with carcinoma of the breast, and 1 with multiple sclerosis. The patients were not given substances with mutagenic properties, apart from the one with lung cancer, who received a course of cytostatics. HBO was administered in standard single-seater OKA-MT chambers, individually selected by the method developed previously [4], namely those giving the maximal increase in tissue pO_2 in response to particular conditions of testing. During HBO the pressure in the chamber varied from 1.2 to 1.5 atm and the oxygen concentration from 96 to 98%. The conditions chosen guaranteed that the dose of hyperbaric oxygen received could not give rise to poisoning. The number of daily 50-min HBO sessions varied from 5 to 15. The required oxygen pressure was established in the course of 5 min. The pressure fell during the same period of time before the end of the session. Venous blood for investigation of the frequency of chromosomal aberrations in the lymphocytes was collected before the beginning of the course of HBO and on the day it ended or the following day. Preparation of whole blood cultures, the conditions of culture, preparation of specimens, and analysis of the frequency of chromosomal aberrations were carried out by the usual methods [3]. The cells were cultured in Eagle's medium for 50 h.

EXPERIMENTAL RESULTS

Frequencies of chromosomal aberrations before and after the course of HBO are given in Table 1. Both the total frequency of cells with chromosomal aberrations and the frequencies of breaks in aberrations of chromatid and chromosomal type are shown. In the case of aberrations such as, for example, dicentrics, rings, and chromatid-chromatid exchanges, it was considered that the exchanges took place as a result of two breaks (one for each chromosome). The results show that

All-Union Medical Genetics Research Center, Academy of Medical Sciences of the USSR, Moscow. No. 6 General Hospital, Ministry of Health of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR A. D. Ado.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 111, No. 5, pp. 532-534, May, 1991. Original article submitted October 25, 1990.

TABLE 1. Frequency of Chromosomal Aberrations before and after Course of HBO

Serial No.	Number of HBO sessions	Pressure, atm	Number of cells analyzed	Fraction of aberrant cells, %	Chromatid breaks			Chromosomal breaks		
					total	per 100 cells	χ^2	total	per 100 cells	χ^2
1	0		136	8,82	11	8,09		6	4,41	
		1,2					1,5			
	10		300	6,00	16	5,33		3	1,00	
2	0		174	4,67	5	2,87		4	2,30	
		1,2					3,42			0,23
	7		300	8,00	21	7,00		5	1,67	
3	0		300	4,33	8	2,67		8	2,67	
		1,2					2,27			0,69
	10		300	2,67	3	1,00		5	1,67	
4	0		206	33,01	75	36,41		66	32,04	
		1,5					1,09			4,46
	13		54	31,48	25	46,30		8	14,82	
5	0		300	7,67	14	4,67		19	6,33	
		1,5					0,03			4,48
	5		300	5,67	15	5,00		8	2,67	
6	0		300	6,67	13	4,33		10	3,33	
		1,2					0,73			5,33
	9		300	3,00	9	3,00			0,67	
7	0		300	6,00	11	3,67		15	5,00	
		1,2					3,11			3,19
	10		244	2,87	3	1,23		5	2,05	
8	0		267	21,72	10	3,75		117	43,82	
		1,2					0,63			3,78
	9		147	18,37	8	5,44		46	31,29	
9	0		300	4,67	4	1,33		12	4,00	
		1,2					2,84			6,63
	15		450	4,22	15	3,30		5	1,11	
10	0		300	7,00	11	3,67		14	4,67	
		1,2					0,89			3,20
	13		300	4,00	7	2,33		6	2,00	
Total:	Before HBO		2583	9,87	162	6,67		271	10,49	
							16,16			37,27
	After HBO		2695	5,86	122	4,53		93	3,45	
							10			10
Number of degrees of freedom							10			10
Probability							0,90			0,99995

no significant change was found in the frequency of breaks in aberrations of chromatid type ($p = 0.9$), whereas the frequency of aberrations of chromosomal type was reduced on average threefold after the course of HBO ($p = 0.00005$).

Therapeutic HBO thus reduces the frequency of chromosomal aberrations of chromosomal type but leaves the frequency of aberrations of chromatid type virtually unchanged. When cells with chromosomal aberrations divide, on the one hand partial elimination of the aberrant cells may take place, or on the other hand aberrations of chromatid type may change into aberrations of chromosomal type. The fact that the frequency of aberrations of chromatid type before and after HBO shows virtually no change may be taken as evidence that HBO does not lead to the formation of lesions inducing chromosomal aberrations of chromatid type, or likewise aberrations of the chromosomal type. The decrease in frequency of the latter after a course of HBO may be the result of the higher degree of proliferation of the cells in vivo and, consequently, of their elimination.

The results of this investigation contradict those of [6], in which HBO was found to have a mutagenic effect. One probable reason for this difference may be the fact that the authors cited used medium 199 and not Eagle's medium for cell culture. Medium 199 contains a small amount of folic acid, the absence of which leads to increased fragility of chromosomes in specific sites [5, 9].

Differences in the effect of hyperbaric oxygen may also be connected with differences in the degree of oxygen saturation of the tissues. In a high concentration oxygen can depress the system eliminating peroxide radicals, thereby giving a mutagenic effect. Oversaturation with oxygen in this way leads to oxygen poisoning. In our case we used therapeutic doses of HBO, not leading to poisoning. These doses may have a stimulating action on the antioxidative protection system, leading to a decrease in the concentration of peroxide radicals and, consequently, to a decrease in the frequency of chromosomal aberrations.

LITERATURE CITED

1. E. P. Gus'kov, T. P. Shkurat, and M. V. Kamynina, Genetika, **21**, No. 10, 1693 (1985).
2. E. P. Gus'kov and T. P. Shkurat, Genetika, **19**, No. 8, 1362 (1983).
3. Technical Recommendations on Testing New Therapeutic Preparations for Mutagenic Properties [in Russian], Moscow (1981), pp. 37-39.
4. V. I. Pakhomov, E. A. Petrova, V. N. Kostyunin, and N. A. Getman, Ter. Arkh., No. 12, 104 (1988).
5. A. T. L. Chen, J. A. Reidy, J. L. Annest, et al., Environ. Molec. Mutagen., **13**, No. 4, 319 (1989).
6. E. P. Guskov, T. P. Shkurat, E. I. Shimanskaja, and S. I. Guskova, Mutat. Res., **241**, 341 (1990).
7. H. Joenje, Mutat. Res., **219**, No. 4, 193 (1989).
8. C. J. Moody and H. M. Hassan, Proc. Nat. Acad. Sci. USA, **79**, 2855 (1982).
9. T. A. Reidy, H. Zhou, A. T. L. Chen, et al., Mutat. Res., **225**, No. 4, 175 (1989).
10. C. E. Vaca, J. Wilhelm, and M. Haums-Ringdahl, Mutat. Res., **195**, 137 (1988).