Heterogeneity of Mononuclear Phagocytes

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HETEROGENEITY OF MACROPHAGES AS REVEALED BY STUDIES ON THEIR OXIDATIVE METABOLISM

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We have examined the ability of macrophages and polymorphs (PMN), from different sources and from different animal species, to release superoxide anion (O_2^-) and H_2O_2 during phagocytosis of serum opsonized heat killed B. mycoides. Table 1 shows that: 1) all the phagocytes examined exhibit an increased O_2 uptake upon phagocytosis; 2) in guinea pig (GP) peritoneal PMN and macrophages (casein elicited) the increased O_2 uptake is associated with a marked release of O_2^- and H_2O_2, while in resident and BCG activated alveolar macrophages of rabbit (R) this release is hardly detectable as previously reported (Biggar et al., 1976, Tsan, 1977;, Yamaguchi et al., 1980); 3) the O_2^- and H_2O_2 released account for only a fraction of the O_2 taken up by all the cells examined; 4) the poisoning of catalase and peroxidase with NaN_3 results in a stoichiometric recovery of H_2O_2 with respect to the oxygen consumed in GP and R PMN, indicating that in these cells all the O_2 consumed is reduced to H_2O_2; 5) the recovery of H_2O_2 with respect to the oxygen consumed is about 50% in human blood PMN and peritoneal macrophages of R (casein elicited) in the presence of NaN_3, indicating that NaN_3-sensitive mechanisms are operative also in these cells; 6) NaN_3 does not appreciably modify the release of H_2O_2 from peritoneal macrophages of GP and alveolar macrophages of R.

Two possible explanations for the low recovery of O_2^- and H_2O_2 with respect to the oxygen consumed by some types of the phagocytes examined are as follows:

i) The O_2 consumed is not reduced or is partially reduced to O_2^- and H_2O_2 and

ii) O_2^- is reduced to O_2^- and H_2O_2 but the rate of degradation

TABLE 1

O_2 UPTAKE BY, AND O_2^- AND H_2O_2 RELEASE FROM PHAGOCYTOSING MACROPHAGES AND POLYMORPHS

		-NaN_3				+NaN_3		
PMN		O_2	H_2O_2	O_2^-		O_2	H_2O_2	O_2^-
Human Blood (HB)	(6)	171±22	35± 9	70±16	(3)	265±47	150±17	80±26
GP Peritoneal Exudates (GPPE)	(4)	173±35	41±10	90±10	(4)	216±38	228±60	79±11
R Peritoneal Exudates (RPE)	(4)	164±25	22±4	42±6	(4)	218±26	201±29	42±10
Macrophages								
GP Peritoneal Elicited (GPPEL)	(5)	230±28	81±15	163±23	(3)	239±33	101±17	151±36
R Peritoneal Elicited (RPEL)	(4)	66±17	17±3	6±1	(4)	66±17	39±10	10±3
R Alveolar Resident (RAR)	(4)	66±13	0	0.4±0.2	(4)	53±13	0.3±0.1	0.8±0.3
R Alveolar BCG-Activated (RAA)	(4)	98±14	6±1	5±1	(7)	81±13	7±1	6±1

The values reported, expressed as nmoles/4 min/1.5×10^6 cells, are the means ± SEM of the differences between phagocytosing and resting cells. Number of experiments is given in parenthesis. O_2^- and H_2O_2 were measured under the same experimental conditions used for the assay of O_2 consumption (Rossi et al., 1980).

TABLE 2

ENZYME ACTIVITIES OF DIFFERENT PHAGOCYTES

	HB-PMN(5)	RPE-PMN(6)	GPPE-PMN(3)	GPPEL-Mϕ(2)	RPEL-Mϕ(4)	RAR-Mϕ(5)	RAA-Mϕ(8)
GSH-peroxidase	1.8±0.4	0.6±0.2	0.6±0.2	5.0	12.4±4.0	25.4±3.6	75.3±13.7
Catalase	14.5±3.2	4.0±0.3	6.7±3.1	20.8	4.6±2.6	31.3±1.2	26.2±3.1
Peroxidase	365.0±65.0	32.3±12.5	115.0±28.0	19.0	6.0±3.0	6.3±1.9	2.0±0.3
SOD	0.44±0.05	0.13±0.03	0.18±0.01	0.69	0.56±0.06	1.63±0.34	0.97±0.17

The means (Units/10^6 cells) ± SEM are reported. The number of experiments is given in parentheses. The experimental details and the definition of Units are reported in Rossi et al., 1980.

of these molecules is faster than the rate of their release.

Cytochalasin B (CB) induces a marked release of both O_2^- and H_2O_2 from RAR-macrophages, indicating that even in these cells oxygen is, at least in part, reduced to O_2^- and H_2O_2 (Rossi et al., 1980 and unpublished data). Table 2 reports the levels of several H_2O_2-degrading enzymes and of the O_2^--degrading enzyme superoxide dismutase (SOD) in the phagocytes examined in this study. The data show that 1) peroxidase activity is high in PMN and low in macrophages; 2) catalase activity is higher in macrophages (with the exception of RPEL-macrophages); 3) macrophages have higher GSH-peroxidase (NaN_3-insensitive H_2O_2-degrading enzyme) than PMN; 4) macrophages have a higher content of SOD than PMN.

In conclusion, these results indicate that

i) in all the phagocytes examined the O_2 consumption is associated with O_2^- and H_2O_2 production;
ii) both macrophages and PMN release only part of the O_2^- and H_2O_2 produced;
iii) most of the O_2^- and H_2O_2 produced is enzymatically degraded within the cells;
iv) the mechanisms of H_2O_2 degradation differ in different types of phagocytes (Higgins et al., 1978). In RAR-macrophages, RAA-macrophages and in GPPEL-macrophages H_2O_2 is degraded mainly through NaN_3-insensitive pathways, while in RPE-PMN and GPPE-PMN H_2O_2 is almost totally disposed of by catalase and peroxidase. In HB-PMN and RPEL-macrophages both NaN_3-sensitive and NaN_3-insensitive mechanisms are probably operative;
v) the relationship between O_2^- release and SOD level varies, indicating that O_2^- release is controlled by additional factor(s) beyond SOD level.

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