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MITOGENIC INDUCTION OF ORNITHINE DECARBOXYLASE IN HUMAN MONONUCLEAR LEUKOCYTES: RELATIONSHIPS WITH ADENOSINE DIPHOSPHATE RIBOSYLTRANSFERASE

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Abstract—1. Relationships between ornithine decarboxylase (ODC) and adenosine diphosphate ribosyl transferase (ADPRT) in human mononuclear leukocytes (HML) were tested by statistical comparisons of their values in a group of 46 people, and by use of inhibitors of ADPRT.

2. ODC was assayed following exposure of HML, for 20 hr, to mitogens [phytohemagglutinin (PHA) and pokeweed mitogen]; ADPRT was measured following exposure of HML to H_2O_2 (100 μM) for 1 hr (activated ADPRT), and in parallel cultures without H_2O_2 (constitutive ADPRT).

3. Significant correlations were found between ODC and ADPRT values; the effects of smoking disturbed the correlations. PHA induction of ODC was negatively influenced by age (standardized β coefficient = -2.95, $P = 0.005$), while age also influenced ADPRT values negatively in non-smokers (for H_2O_2 activated ADPRT, standardized β coefficient = -2.74, $P < 0.008$).

4. Inhibitors of ADPRT, nicotinamide, caffeine and benzamide inhibited the induction of ODC by PHA in a concentration-dependent manner, in the range (0.6–10 mM) known to inhibit ADPRT.

INTRODUCTION

Ornithine decarboxylase (ODC) and adenosine diphosphate ribosyl transferase (ADPRT) are enzymes involved in different ways in the processing and synthesis of DNA. The product of ODC action, putrescine, is converted to other polyamines, which, together, are involved in DNA replication and protein synthesis by means that remain to be fully elucidated (Tabor and Tabor, 1984). ADPRT, in transferring poly ADP-ribose to a variety of proteins, is involved in processes such as cellular proliferation and differentiation, gene expression and DNA repair (Ueda and Hayaishi, 1985; Durkacz *et al.*, 1980; Cleaver and Morgan, 1987).

ODC is widely measured in the study of effects of tumor promoters (O'Brien *et al.*, 1975; Verma and Boutwell, 1977; Weeks *et al.*, 1982), and the enzyme has also been proposed as a marker of familial polyposis, and by inference, as a marker of predisposition to colorectal cancer (Luk and Baylin, 1984). With respect to ADPRT, we have reported associations between clinical conditions and the responses of ADPRT in human mononuclear leucocytes (HML) to oxidative stress. Thus ADPRT levels in HML following incubation with autologous plasma, or with cumene hydroperoxide, could distinguish, in a statistically significant manner, between individuals with or without colonic adenomatous polyps (Markowitz *et al.*, 1988a; Pero *et al.*, 1988a). In further reports we have shown that treatment of HML with H_2O_2 activates ADPRT levels above their constitutive, or background values, but that this response of ADPRT is significantly reduced in cancer patients with tumor *in situ* and in patients with inflammatory bowel disorders (Markowitz *et al.*, 1988b; Pero *et al.*, 1988b).

It is widely reported that H_2O_2 and other forms of oxidative stress cause DNA damage of different kinds, including single strand breaks (Emerit and Cerutti, 1981; Halliwell and Gutteridge, 1984; Birnboim, 1986; Frenkel and Chrzan, 1987; Imlay *et al.*, 1988; Adelman *et al.*, 1988). Activation of nuclear ADPRT by DNA strand breaks is also well known (Ueda and Hayaishi, 1985; Berger *et al.*, 1980; Benjamin and Gill, 1980; Johnstone and Williams, 1982; Jacobson *et al.*, 1983). The novelty of our observations cited above, is that they show correlations between clinical conditions and the response of ADPRT to compounds that cause oxidative stress.

Because of the potential importance of ODC as a biological marker, we are investigating factors that affect, or relate to, the induction of the enzyme in response to growth stimuli. Here we record statistically significant correlations between ODC and ADPRT. We show also that there are parallels in the influence of age on both ODC and ADPRT in HML. Our results show, further, that induction of ODC following PHA treatment of HML is suppressed by well characterized inhibitors of ADPRT.

MATERIALS AND METHODS

L-Ornithine, pyridoxal phosphate and dithioerythritol were from Sigma Chemicals (St Louis, MO). Cellulose cation exchange paper (P-81) was supplied by Whatman

Labsales Inc., Hillsboro, OR. L-[2,3-H]Ornithine (56 Ci/mmol) was from NEN research products. Scintiverse II was from Fisher Scientific, Springfield, NJ, as were all other reagent grade materials. Phytohemagglutinin (M form, PHA) and pokeweed mitogen (PWM), in lyophilized form, were from Gibco Laboratories, Grand Island, N.Y. Cell culture medium (1 × RPMI 1640 without glutamine) was from Flow Laboratories Inc., McLean, VA. This medium was supplemented with antibiotics (50 U penicillin and 50 μg streptomycin per 100 ml, from Gibco Laboratories).

Volunteers were recruited at PMI/Strang Clinic from the normal flow of patients through the clinic, and were free of clinical symptoms of major disease. Blood was collected into heparinized vacutainers, and centrifuged (100 *g*, 15 min) to separate plasma which was then removed. The blood was rediluted to its original volume with physiological saline. Mononuclear leukocytes (HML) were prepared from this blood on a solution of diatrizoate and ficoll, as previously described (Pero *et al.*, 1976). Plasma separated from the cells was centrifuged at 400 *g* for 25 min, and the platelet-depleted plasma (PDP) obtained was available for inclusion in culture media in subsequent experiments.

Culture of HML with mitogens

HML ($3–5 \times 10^6$) were cultured in RPMI medium supplemented with 20% PDP, at a cell density of 2×10^6 cells/ml, with added mitogen, for 20 hr at 37°C. Quantities of mitogen added are recorded in the Results section. The mitogen preparations used were formulated by the manufacturer (see above) and were reconstituted in distilled water, according to the manufacturer's instructions, and then aliquoted and frozen in small quantities. In the experimental details given below, volumes always refer to this reconstituted preparation. When compared with a purified preparation of PHA-M from Sigma Chemical Co., St Louis, MO., addition of the Gibco preparation at 10 μl/ml culture was equivalent to 2 μg of the Sigma preparation/ml culture.

Assay of ODC: this procedure is adapted from that reported by Djurhus (1981) and described in detail elsewhere (Johnson *et al.*, 1989).

Assay of ADPRT: the procedure is adapted from that of Berger (1978) with modifications previously described (Pero *et al.*, 1983). Duplicate samples of 5×10^5 mononuclear leukocytes were cultured in physiologic saline supplemented with 1% PDP for 1 hr at 37°C without additive, or with H_2O_2 (100 μM), as indicated in the Results section. The cells were centrifuged (400 *g*, 10 min) and permeabilized at 4°C by suspension for 15 min in 1.5 ml ice cold buffer (10 mM Tris–HCl, pH 7.8; 1 mM EDTA, 30 mM 2-mercaptoethanol, 4 mM $MgCl_2$). The cells were again centrifuged and resuspended in 50 μl permeabilization buffer, 25 μl reaction buffer (100 mM Tris–HCl, pH 7.8, 120 mM $MgCl_2$) and 15 μl [^{3}H]adenine labeled NAD^+ (27.1 Ci/mmol, diluted to final concentration of 220 μM). Following incubation at 30°C for 15 min the reaction was terminated by addition of 0.5 ml 3 M NaCl at 60°C for 10 min. Protein, including the ADP-ribosylated fraction, was precipitated with ice cold 7% trichloroacetic acid (TCA) and absorbed on nitrocellulose filters (Millipore HAWP 02500). The data were recorded as TCA precipitable cpm/5×10^6 cells in the presence (activated levels) or absence (constitutive levels) of 100 μM H_2O_2.

Statistical analyses were performed on a Data General minicomputer (MV 2000 DC) with a software package from SAS Inc., Cary, NC, using two sided t-testing, regression and general linear modeling procedures.

RESULTS

Choice of culture conditions for treatment of HML with mitogens

The contribution of autologous PDP to the mitogenic response, as measured by induction of ODC, was variable, but generally provided a higher enzyme value than did dialyzed fetal bovine serum (Fig. 1). Because of this, and because PDP is used in cell treatment prior to ADPRT assays, PDP was used in all culture media used for mitogen treatment of HML. The induction of ODC by PHA and PWM was dependent on mitogen concentration (Figs 2 and 3). Other results (e.g. Fig. 1) indicate that the concentration of mitogen producing maximal ODC activity varies from individual to individual. It was not feasible to determine this concentration dependence in cells from a large number of individuals. In order to select a single standard concentration of PHA for cell culture, 10 samples were treated with 2 different mitogen concentrations, 20 and 30 μl PHA per ml culture. As shown (Fig. 4) there is good correlation

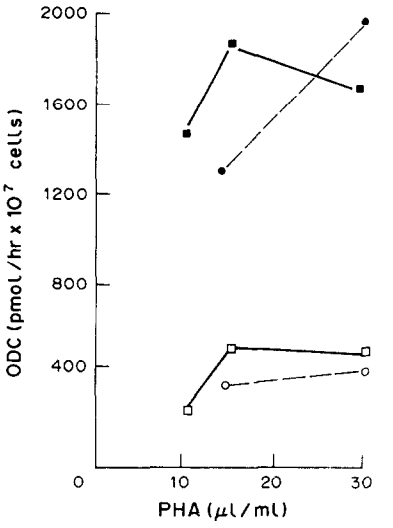

Fig. 1. Effects of autologous plasma and fetal bovine serum on ODC induction by PHA. Cells were cultured at a density of 2.0×10^6 in RPMI medium supplemented with (■, ●) autologous PDP (20%), or with (□, ○) fetal bovine serum (20%), with added PHA (30 μl/ml culture): person A (■, □), person B (●, ○).

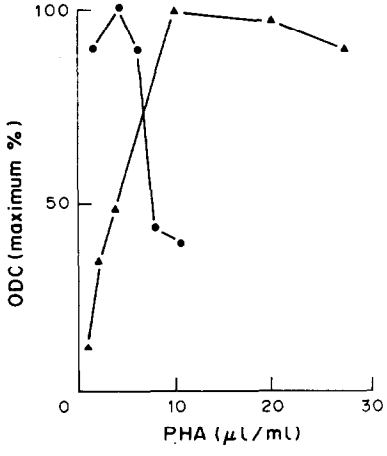

Fig. 2. ODC induction at increasing PHA concentrations. HML from two individuals (●, ▲) were cultured at 37°C, at a cell density of 2.0×10^6/ml in RPMI medium supplemented with autologous platelet poor plasma (PDP, 20%).

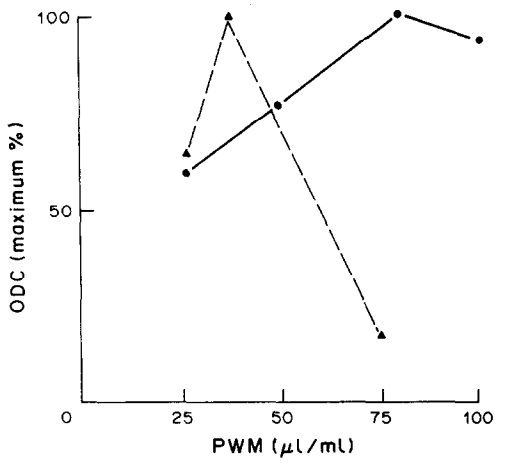

Fig. 3. ODC induction at increasing PWM concentrations. HML from two individuals (●, ▲) were cultured as described in Fig. 2.

between the values obtained with the two levels used: a coefficient of determination (R^2) of 0.91 ($P = 0.0001$) was obtained. Therefore, in the first part of this study, PHA was used in culture media at a level of 30 μl/ml. In order to standardize culture conditions, the same volume of reconstituted PWM preparation was also used.

Correlative study of enzyme activities

In the first part of this study, ODC activities were measured following treatment of HML with mitogens. ADPRT activities were measured in cells treated, as described in Materials and Methods, with and without H_2O_2, but, in these cases, without exogenous mitogen. In this way, ODC and ADPRT activities were measured in cells from 46 individuals (Group A), with 24 non-smokers and 22 smokers (Tables 1 and 2). ADPRT values, constitutive and H_2O_2-activated, were measured in a further group of

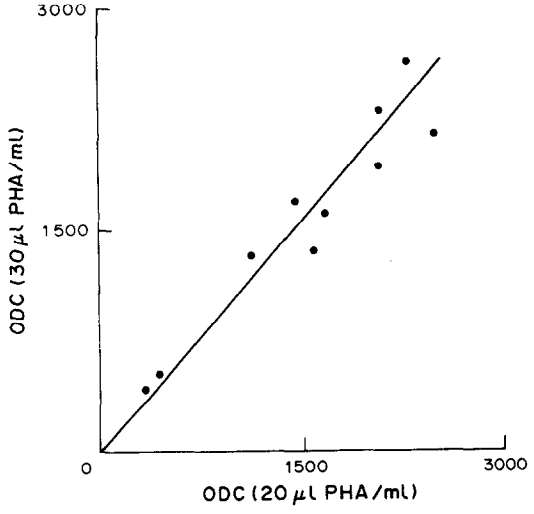

Fig. 4. Correlation of ODC activities induced by two levels of PHA. Cells were cultured as described in Fig. 1, with 30 μl PHA added/ml, or with 20 μl PHA added/ml. The two sets of values were correlated by a least-squares method.

70 individuals (Group B), all non-smokers (Table 2). Table 1 shows ODC values resulting from mitogen treatment of HML. Univariate analysis (Table 1) suggests that age significantly influenced induction of ODC by both PHA and PWM. Multivariate analysis confirms the effect of age on ODC induction by PHA but not by PWM. Neither smoking nor gender influenced ODC induction by PHA or by PWM in this group of study participants.

With regard to ADPRT values, multivariate analysis shows that gender was not a significant influence (Table 2). In the smaller group (Group A), the influence of age on ADPRT was not significant, while in the larger group (Group B, all non-smokers) both constitutive and activated ADPRT values were influenced significantly by age, in a negative fashion (Table 2). Furthermore, in the smaller group, smoking did influence H_2O_2-activated ADPRT positively, but not constitutive ADPRT. Therefore, in the case of H_2O_2-activated ADPRT, the influence of age may be at least partly counteracted by the effects of smoking.

Comparison of ODC and ADPRT activities (Table 3) shows that there are statistical relationships between the two measurements. When PHA was the mitogen, ODC activity related to constitutive ADPRT in the whole group, as well as in smokers and non-smokers. The relationship of PHA-induced ODC to H_2O_2-activated ADPRT was weaker, and was non-significant in smokers. PWM-induced ODC related significantly to both constitutive and H_2O_2-activated ADPRT values, but only in non-smokers. Therefore, the effects of smoking on ADPRT can confound the effects of age on that enzyme, as well as disturb the relationship between ADPRT and ODC.

Mechanistic basis for enzyme correlations

The possibility that ODC induction following PHA treatment of cells might be dependent on ADPRT was tested by including inhibitors of ADPRT (nicotinamide, benzamide, caffeine) in incubation media. It is shown (Fig. 5) that these inhibitors have a marked effect in countering the induction of ODC by PHA: induction of ODC could be suppressed by over 80% by nicotinamide and benzamide, at relatively high concentrations (10 mM). Nevertheless, significant suppression could be observed at relatively low levels of these two inhibitors, while considerable suppression was achieved with caffeine at concentrations in the regions of 1 mM. At such concentrations these compounds have been shown to inhibit ADPRT activity effectively (Sims *et al.*, 1982).

We have also examined the effects of ADPRT inhibitors on ODC induction by adding the inhibitor, nicotinamide and caffeine, at different times relative to addition of PHA to cultures. Most complete inhibition was achieved when inhibitors were added at the same time as mitogen (Table 4). The effects of the inhibitors were notably less when added 3 or 7 hr after PHA.

It has been reported that some inhibitors of ADPRT also inhibit the production of superoxide radical by the NADPH oxidase system of monocytes (components of HML) and of polymorphonuclear cells. It has also been shown that ODC induction by treat-

Table 1. ODC activity (pmol/hr·10^7 cells) in mitogen treated HML

		Mean		Min		Max		SE	
Group	*n*	PHA	PWM	PHA	PWM	PHA	PWM	PHA	PWM
Total	46	684	496	36	4	2585	2369	92	88
Females	30	762	433	54	47	2585	2369	129	109
Males	16	544	607	36	4	1267	1793	105	151
⩾50 years	30	484[A]	331[C]	54	4	1517	1973	75	81
<50 years	16	1046[B]	794[D]	36	47	2585	2369	193	182

Univariate analysis: for [A] vs [B]—t = 2.7 ($P = 0.013$); for [C] vs [D]—t = 2.3 ($P = 0.03$).
Multivariate analysis to test the effect of age on ODC values: PHA induced ODC—standardized coefficient = −0.30, $P = 0.005$; PWM induced ODC—not significant at 0.05 level.

ment of HML with PHA could be inhibited by a variety of anti-oxidants, including Vitamin E, mannitol, catalase and superoxide dismutase (Johnson, 1989). Furthermore, the induction of ODC was also inhibited by diphenylene iodonium, an inhibitor of NADPH oxidase (Johnson, 1989). The correlations noted above between ADPRT and ODC might arise, therefore, because of a dependence of both activities on oxidant mechanisms. Consequently, we have tested the activation of ADPRT in HML by PHA. The results (Figs 6 and 7) show clearly that the action of PHA involves a mechanism that activates ADPRT in a manner similar to the action of oxidants such as cumene hydroperoxide and H_2O_2 (Markowitz *et al.*, 1988a, b; Pero *et al.*, 1988b).

DISCUSSION

We have studied ODC in human mononuclear leukocytes, using them as a readily available source of human cells. In this report we explore the possible relationship of ODC with another enzyme, ADPRT.

ADPRT is of interest here because of its role in the cell cycle (see below) and because both ODC and ADPRT are known to be affected by tumor-promoting phorbol esters. Further actions of such tumor promoters include production of superoxide radical, the induction of DNA strand breaks, and the inhibition of DNA repair synthesis (O'Brien *et al.*, 1975; Poirier *et al.*, 1975; Emerit and Cerutti, 1981; Singh *et al.*, 1985; Birnboim, 1986). Furthermore, there is a growing body of evidence relating oxidant-activated ADPRT, with clinical conditions, including the presence of adenomatous polyps in the colon (Markowitz *et al.*, 1988a), inflammatory bowel disorders (Markowitz *et al.*, 1988b), and the presence of malignant tumor in a variety of tissues (Pero *et al.*, 1988b).

Statistically significant correlations were found between ODC and ADPRT in mononuclear leukocytes of 46 individuals: mitogen-stimulated ODC correlates with both constitutive and H_2O_2-activated ADPRT. Age has been shown to influence both these enzyme measurements negatively: this is in keeping with observed effects of age on other lymphocyte functions (Schwab and Weksler, 1986; Proust *et al.*, 1987). There is, therefore, a suggestion of a biological relationship between the two enzymes. Such a relationship might arise because of a requirement for one enzyme in the induction of the other.

ODC and ADPRT are both induced during the mitogenic response; these inductions are considered essential for the progression of lymphocytes through the cell cycle. ODC induction precedes S phase: in human fibroblasts (Ritling *et al.*, 1986; Kaczmarek *et al.*, 1987) and human lymphocytes (Kaczmarek *et al.*, 1987) induction of ODC is a G_1 event. In bovine lymphocytes the synthesis of ODC mRNA reportedly reached a maximum 6 hr from the time of application of mitogen; thereafter, increases in ODC activity were related to the efficiency of mRNA translation (White *et al.*, 1987). In a macrophage-like cell line ODC mRNA was induced more rapidly, occurring 2 hr after stimulation with bacterial lipopolysaccharide (Shurtleff *et al.*, 1988).

Increases in ADPRT during cell cycling have also been reported. There is general agreement that the enzyme activity is low in quiescent cells (the constitutive level), and, according to one report, there is no enzyme synthesis at least within 8 hr of exposure to mitogen (Scovasi *et al.*, 1987). In a further study, between 24 and 48 hr following mitogenic stimulation of human lymphocytes, ADPRT activity increased *ca* 3-fold, and at 72 hr had increased by an additional

Table 2. ADRPT activities in HML treated with H_2O_2 (activated) and in untreated controls (constitutive)

		ADPRT (cpm/5 × 10^6 cells)					
		Constitutive			H_2O_2-activated		
Group	*n*	Mean ± SE	Min	Max	Mean ± SE	Min	Max
A	46	710 ± 57[1]	298	1955	3456 ± 288	372	9601
Female	30	708 ± 71	298	1955	3584 ± 385	372	9601
Male	16	713 ± 98	308	1538	3238 ± 426	731	7102
B	70	694 ± 26[2]	93	2209	2617 ± 98	122	8709
Female	41	668 ± 34	139	2209	2532 ± 137	431	8709
Male	29	723 ± 40	93	2124	2713 ± 140	122	8131

[1] Effect of smoking on activated ADPRT—standardized β coefficient = 2.09, $P = 0.04$.
[2] Effect of age on activated ADPRT—standardized β coefficient = −2.74, $P = 0.008$; constitutive ADPRT—standardized β coefficient = 2.06, $P = 0.044$.

Table 3. Multivariate analysis of variance to test the relationship between PHA induced ODC and ADPRT

Mitogen	Group	Partial correlation coefficients and probability values for ODC related to: Constitutive-ADPRT	Partial correlation coefficients and probability values for ODC related to: H_2O_2 activated-ADPRT
PHA	Total ($n = 46$)	0.51 (0.003)	0.30 (0.04)
	Non-smokers ($n = 24$)	0.51 (0.01)	0.54 (0.006)
	Smokers ($n = 22$)	0.51 (0.011)	NS*
PWM	Total ($n = 46$)	NS	NS
	Non-smokers ($n = 22$)	0.41 (0.04)	0.49 (0.02)
	Smokers ($n = 24$)	NS	NS

*NS = not significant.

factor of 2 (Rochette-Egly, 1980); changes in enzyme levels during the first 24 hr period were not described in that study. It is also reported that, in pig lymphocytes treated with PHA, there was overall a 2.9-fold increase in ADPRT with one quarter of that increase occurring in the first 24 hr, prior to DNA synthesis (Lehmann *et al.*, 1974). There is agreement also, that although ADPRT levels are low in quiescent lymphocytes, the enzyme is readily activated by DNA strand breaks (Durkacz *et al.*, 1980; Ueda and Hayaishi 1985; Cleaver and Morgan, 1987). We have shown that ADPRT activity increased within 1 hr of treatment with mitogen, earlier than the reported time of induction and *de novo* synthesis of the enzyme. This is consistent with the induction of chromosomal strand breaks early in the mitogenic response, with consequent activation of ADPRT.

Benzamide, caffeine and nicotinamide have been widely characterized and used as inhibitors of ADPRT (Durkacz *et al.*, 1980; Rochette-Egly *et al.*, 1980; Ben-Hur *et al.*, 1984; Ueda and Hayaishi, 1985). Our results clearly show that these inhibitors suppressed the induction of ODC by PHA. The effects of the inhibitors were greatest when added to cell cultures simultaneously with PHA, rather than one or more hours later. This may implicate ADPRT in early events leading to the induction of ODC. We note, however, that the effects on ODC induction of the ADPRT inhibitors used are not in the same order in which they inhibit ADPRT. Thus caffeine was a more effective suppressant of ODC induction than nicotinamide, while caffeine inhibited ADPRT rather less than nicotinamide did (Sims *et al.*, 1982). This may suggest that ODC induction is influenced by the inhibitors through a mechanism separate from ADPRT inhibition.

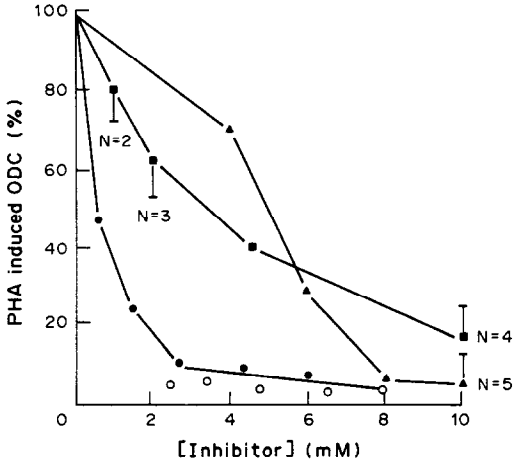

Fig. 5. Effects of inhibitors of ADPRT on induction of ODC by PHA. Cells were incubated as described in Fig. 1, with the addition of 20 μl PHA/ml culture medium, and with nicotinamide (▲), or benzamide (■), or caffeine with subject 1 (○) or caffeine with subject 2 (●). Standard error bars are included where more than one blood sample was tested. *N* indicates the number of samples used to obtain the mean values plotted.

In the study of statistical correlations between ADPRT and ODC, different modes of enzyme activation were employed, one with oxidative stress (ADPRT activated by H_2O_2), and one with exogenous mitogen (ODC induced by PHA). An important possibility is that the observed correlations arose not because the induction of ODC is dependent on ADPRT, but because both activities are modified by oxidant conditions. It is clear that activation of ADPRT by H_2O_2 is a response to oxidant, while it has been shown (Johnson, 1989) that the induction of ODC by PHA can be significantly modified by antioxidants such as Vitamin E, mannitol, catalase and superoxide dismutase. In further work, to be published elsewhere, it has been shown that exogenous H_2O_2 can contribute significantly to the induction of ODC (D. B. Johnson, unpublished observations). As reviewed above, both ODC and ADPRT are induced by tumor-promoting phorbol esters, compounds that are known to generate oxidant states. It should also be noted that some inhibitors of ADPRT, notably nicotinamide, have other functions, including the inhibition of superoxide radical production by the NADPH oxidase system of monocytes and macrophages (Troll *et al.*, 1987).

Therefore, the statistial correlations reported above between ODC and ADPRT may arise as a result of the dependence of ODC induction on AD-

Table 4. Suppression of ODC induction by inhibitors of ADPRT

Time of addition of inhibitor following PHA (hr)	*n*	ODC activity (% of control) ±SE
0	5	8.0 ± 2.3
1	3	13.7 ± 5.1
3	2	21.5 ± 7.8
20	5	100.0

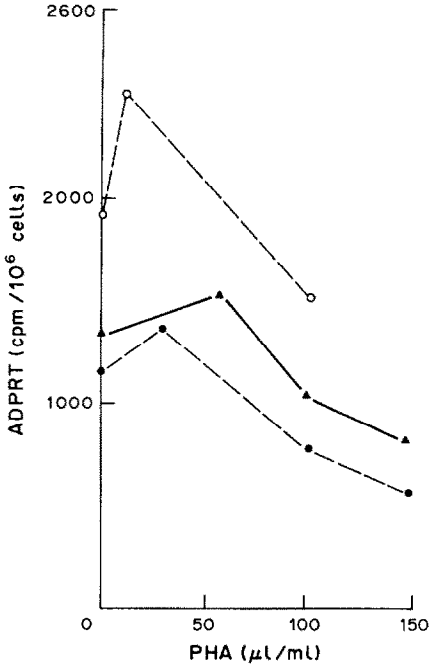

Fig. 6. Effects of PHA and ADPRT activity in HML from three individuals. Cells were cultured in RPMI supplemented with 20% PDP, for 1 hr, at the concentrations of added mitogen shown.

PRT activity, or alternatively, because of a common oxidant-dependent mechanism affecting both enzymes.

SUMMARY

1. Factors affecting, or relating to, induction of ODC in HML were studied. Enzyme induction was by PHA and by pokeweed mitogen.

2. In a group of 46 subjects (30 female, 16 male; 22 smokers, 24 non-smokers) neither gender nor smoking habits affected induction of ODC, while age did affect enzyme induction by PHA but not by pokeweed mitogen. The PHA response was significantly reduced with subject age.

3. Adenosine diphosphate ribosyl transferase activities were measured in HML, following incubation for 1 hr in physiological saline containing 1% autologous plasma (constitutive ADPRT levels) or following incubation under the same conditions but with added H_2O_2 (100 μM) (H_2O_2-activated ADPRT levels).

4. Multivariate analysis showed that H_2O_2-activated ADPRT levels correlated significantly with ODC induced by PHA (partial correlation coefficient 0.54, $P = 0.006$), and by pokeweed mitogen (partial correlation coefficient 0.49, $P = 0.02$) in non-smokers, but not in smokers.

5. In the same group of 46 subjects, ADPRT levels were shown to be affected significantly by smoking habits (standardized β coefficient 2.09, $P = 0.04$) but not by age. In a further group of 70 subjects, all non-smokers, it was shown that age affected ADPRT levels. Standardized β coefficients and probability values were, for the effect of age on constitutive ADPRT, -2.06, $P = 0.044$, and on H_2O_2-activated levels of ADPRT, -2.74, $P = 0.008$. Therefore, the affects of smoking included a confounding of the effects of age, as well as a disturbance of the relationship between ODC and ADPRT.

6. The induction of ODC was inhibited by nicotinamide, caffeine and benzamide, compounds well characterized as inhibitors of ADPRT. This, together with the relationships described above, may indicate a dependence of ODC induction of ADPRT.

7. Alternatively, the correlations may arise because of a basic mechanism underlying both ODC and ADPRT measurements. Such a mechanism could be a dependence on an oxidant mechanism in both the activation of ADPRT and the induction of ODC.

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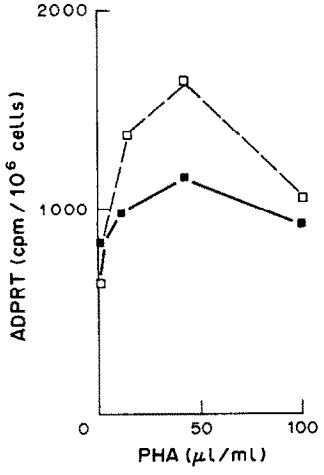

Fig. 7. Effects of PHA on ADPRT activity in HML. Cells from two individuals (neither is represented in Fig. 5) were cultured in RPMI supplemented with 5% PDP, for 1 hr, at the concentrations of added mitogen shown.

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