Table 2. Mortality of CBA and hybrid (C57BL × CBAT6T6)F_1 mice joined in parabiosis

Pretreatment of partners CBA	(C57BL × CBAT6T6)F_1	Number of pairs	Died (No. of days after parabiosis) (C57BL × CBAT6T6)F_1	Both	CBA
N	N[a]	15	15 (2×9, 10, 2×11, 2×15, 16, 17, 2×18, 19, 21, 22, 23)	0	0
	T[b]	15	15 (3×8, 2×9, 3×10, 11, 12, 13, 15, 17, 20, 21)	0	0
IR[c,d]	N	15	10 (11,12,15,17,18, 19, 21, 22, 24, 57)	5 (10, 14, 15, 18, 31)	0
	T	15	13 (10, 2×11, 12, 13, 3×14, 15, 17, 19, 24, 27)	2 (25, 28)	0
T	N	15	9 (9, 10, 12, 16, 20, 24, 26, 31, 32)	5 (6, 2×7, 9, 15)	1 (8)
	T	15	6 (18, 23, 30, 31, 35, 51)	9 (2×6, 2×7, 10, 13, 15, 17, 27)	0
TIR	N	15	0	14 (9, 16, 2×17, 2×21, 23, 24, 25, 27, 2×30, 31, 56)	1 (40)
	T	15	1 (25)	9 (10, 11, 12, 13, 2×14, 15, 20, 26)	5 (10, 19, 2×20, 46)

[a] Normal. [b] Thymectomized. [c] Irradiated (900 R). [d] Reconstituted with 10^7 syngeneic bone marrow cells.

type were found in spleen, lymph nodes, bone marrow and thymus when present. These results suggest that inadequate immunological protection plays in secondary disease a more important role than GVH reaction does. CBA mice used for parabiosis studies were normal N, irradiated and reconstituted with syngeneic bone marrow IR, thymectomized T and mice thymectomized irradiated and reconstituted with syngeneic bone marrow cells TIR. They were joined to normal N or thymectomized T F_1 hybrides of the same sex.

As shown in table 2, all hybrid partners succumbed in about 3 weeks when parabiosed with normal CBA parent. All hybrid partners were also killed when parabiosed to CBA IR mice, but some of the parental partners died on the same day, as did the hybrid mice. Mortality of parental partners was further increased by prior thymectomy. Finally, when parental CBA mice were both thymectomized and irradiated TIR, their mortality was greater than that of their hybrid partners. Also the highest frequency of deaths of both parabiotic partners was observed in groups in which the immune status of partners was impaired by thymectomy and/or irradiation. It appears that immunologically crippled parental partners succumbed due to a continuous immunological stimulus, which, exceeding the capacity of the immune system, led these partners to immunological exhaustion and consequent death[1,8].

Statistical analysis of IR, T or TIR CBA mice parabiosed with normal or thymectomized hybrid partners showed significant differences ($p < 0.001$ for each comparison). This suggests that the immunological status of hybrid partners also influences the outcome of the parabiosis.

Dexamethasone-induced neutrophilia. Negative correlation with increased plasma adrenaline concentrations

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Summary. Maximal increments in adrenaline and dexamethasone (DXM) plasma concentrations were observed c15 (T_{50} 40 min) and 30 (T_{50} 210–240 min) minutes after an i.v. DXM dose (6 mg/m² BSA) in man. There appears, however, to be no direct interaction between these agents in the development of induced neutrophilia, which occurs c240 min postinjection.

Glucocorticoids in general[2] and dexamethasone (DXM) in particular[3], induce a peripheral blood neutrophilia in man with conventional pharmacological dosage. The neutrophilia is maximally developed 4–6 h following either i.v. or oral administration and is associated with a profound lymphocytopenia[2]. It has been suggested that mobilization of the marginated granulocyte pool (MGP) may account for the increased circulating granulocyte pool (CGP), because of the presence during neutrophilia of cells with low alkaline phosphatase (AP) activity[3], presumably emerging from sequestered organ sites and

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2 J. M. Mishler, in: Experimental Hematology. Leucapheresis and Plasmapheresis, vol. 5, 15, suppl. Ed. J. M. Goldman and J. M. Mishler, Munksgaard 1977.

3 J. M. Mishler and P. M. Emerson, Br. J. Haemat. *36*, 253 (1977).

4 J. W. Athens, O. P. Haab, S. O. Raab, A. M. Mauer, H. Ashenbrucker, G. E. Cartwright and M. M. Wintrobe, J. clin. Invest. *40*, 989 (1961).

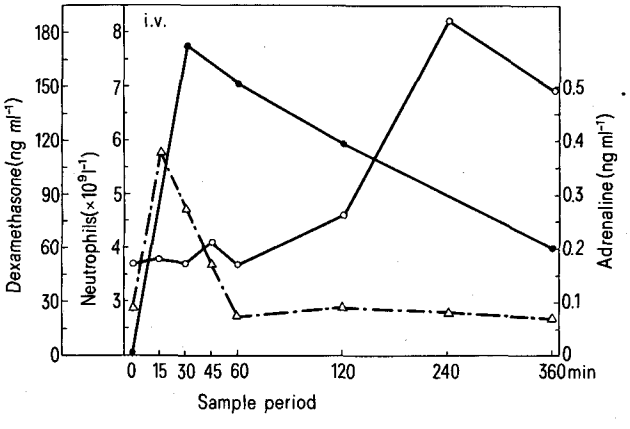
small venules. Labelling studies[4], on the other hand, have established that adrenaline-induced neutrophilia is is manifested by an elevated CGP at the expense of the MGP, and during maximum peripheral counts, the AP activity of cells is low[5]. Aside from the actual kinetic development of neutrophilia, both DXM and adrenaline invoke similar peripheral cell effects; thus it was of interest to determine if DXM increased adrenaline levels during neutrophilia, as its principal mechanism of action.

Materials and methods. 3 normal healthy male subjects (ages 21–30 years) participated in the present study. Dexamethasone sodium phosphate (Organon Laboratories Ltd, U.K.) 6 mg/m² BSA was administered i.v. through a 19-gauge butterfly needle inserted in a forearm vein, between 08.00 to 09.00 h on the study day. Each subject had previously fasted the night before and continued fasting during the entire sampling period while resting in a supine position. Whole blood samples were taken prior to (baseline) and 15, 30, 45, 60, 120, 240, and 360 min postinjection. Laboratory indices measured at each sampling interval included plasma DXM[6] and adrenaline (including noradrenaline)[7] concentrations and total leucocyte count with differential scoring for neutrophils.

Results. 15 min following the i.v. DXM injection, the plasma concentrations of both DXM and adrenaline had risen markedly (figure). The elevated adrenaline levels subsided rapidly, and at 1 h postinjection, simulated pre-infusion concentrations. The noradrenaline concentrations also increased during the 1st 30 min, from 0.21 to 0.43 ng ml^{-1}, thereafter the levels declined to prefusion values. The DXM levels decreased in a slower fashion, with a plasma T_{50} of approximately 210–240 min. The peripheral neutrophil count, on the other hand, did not commence to rise until approximately 120–240 min postinjection; well beyond the intervall in which the highest concentration of adrenaline was observed. The neutrophil count reached its maximum value 240 min postinjection and thereafter started to subside.

Discussion. Glucocorticoid-induced neutrophilia has in the past been explained in the following manner: 1. cellular egress from the CGP may be moderately or severely reduced, 2. an influx of new cells may in some cases come from the bone marrow reserve (BMR), but 3. an intravascular shift of blood pools (e.g. from the MGP to the CGP) does not occur[8,9]. It has been tacitly assumed that all glucocorticoids behave in a similar fashion, that is, they impair or inhibit neutrophil movement from the intravascular to the extravascular compartments. Thus on this assumption the neutrophilia which follows glucocorticoid administration simply reflects the accumulation of cells possessing an increased intravascular T_{50}. More recent studies have suggested that DXM does not impair movement of neutrophils into induced dermal lesions[10] and that an intravascular shifting of blood pools (MGP → CGP) may occur[3], as evidenced by cells possessing low AP activity and by the absence of increased numbers of immature (band) cells during maximum neutrophilia. The absence of increased numbers of immature cells at maximum neutrophilia, suggests but does not prove, that the BMR is not involved significantly in the observed CPG elevation. The low AP activity of cells during DXM-induced neutrophilia, may signify re-entry or shifting of 'older' mature cells from the MGP to the CGP. The belief that the MGP is a depot for the storage of 'older' neutrophils is suggested by the observation that AP content[11] and leucocyte membrane resistence[12], which are both inversely related to neutrophil age, are both decreased following adrenaline-induced neutrophilia[5,12].

Even though similarities exist between the peripheral blood effects induced by both DXM and adrenaline, there appears to be no direct interaction between these 2 agents during the development of neutrophilia, as evidenced by the present study. The exact mechanism of DXM-induced neutrophilia would seem to warrent additional investigation, in order to elucidate the mechanism of action.



The plasma concentrations of DXM (●) and adrenaline (△) are plotted in reference to the peripheral blood neutrophil (○) count, in 3 healthy male subjects administered 6 mg/m² DXM i.v. Each point represents the mean of 3 determinations, except for DXM plasma levels, in which the mean of 2 values were plotted.

5 J. M. Mishler and A. A. Sharp, Scand. J. Haemat. *17*, 78 (1976).
6 M. Hichens and A. F. Hogans, Clin. Chem. *20*, 266 (1974).
7 H. Hörtnagl, C. R. Benedict, D. G. Grahame-Smith and B. McGrath, Br. J. clin. Pharmac. (in press).
8 J. W. Athens, O. P. Haab, S. O. Raab, D. R. Boggs, G. E. Cartwright and M. M. Wintrobe, J. clin. Invest. *41*, 1342 (1962).
9 C. R. Bishop, J. W. Athens, D. R. Boggs and G. E. Cartwright, Clin. Res. *15*, 130 (1967).
10 W. P. Peters, J. F. Holland, H. Senn, W. Rhomberg and T. Banerjee, New Engl. J. Med. *282*, 242 (1972).
11 D. M. Williams, Br. J. Haemat. *31*, 371 (1975).
12 E. Storti, E. Lusvarghi, G. F. Grignaffini and A. Sgandurra, Haematologia *1*, 27 (1967).