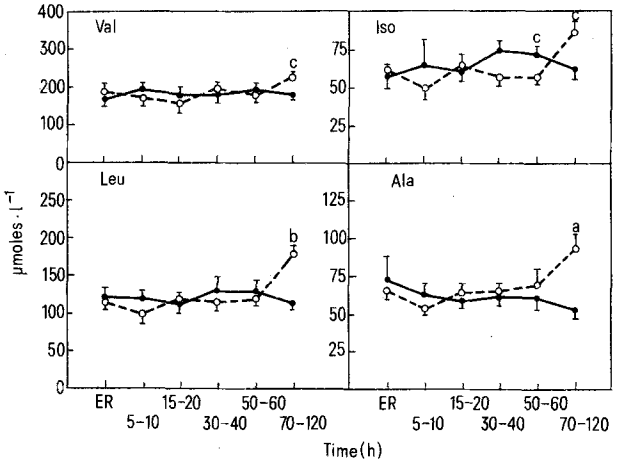


Figure 2. Concentrations of essential amino acids in blood of AMI and non-AMI patients. See legend to figure 1 for details. (Val, valine; Iso, isoleucine; Leu, leucine; Ala, phenylalanine) ($^{a}p < 0.001$; $^{b}p < 0.005$; $^{c}p < 0.05$).

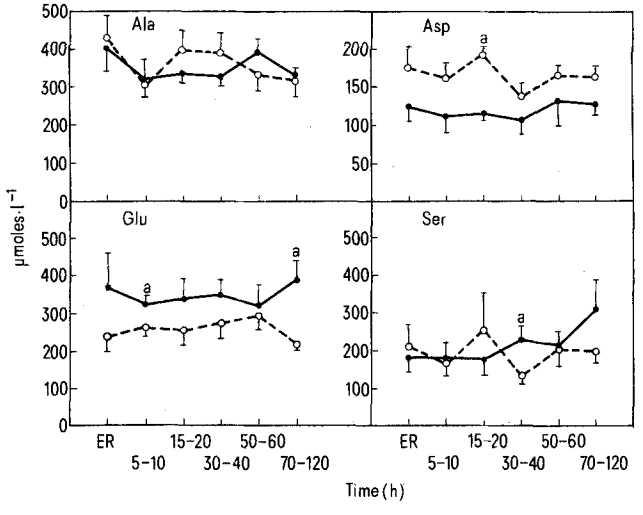


Figure 3. Concentrations of nonessential amino acids in blood of AMI and non-AMI patients. See legend to figure 1 for details. (Ala, alanine; Asp, aspartic acid; Glu, glutamic acid; Ser, serine) ($^{a}p < 0.05$).

supernatant was analyzed for amino acid content on a Beckman amino acid analyzer. Statistical significances were determined by using the Wilcoxon 2-sample test.

*Results.* Levels of taurine in blood samples which were obtained at the ER admittance from AMI patients were significantly elevated as compared to blood taurine levels of patients who had chest pain but who were later diagnosed as not having had an AMI (fig. 1). The taurine levels remained elevated for 50–60 h and returned to control values by the 70–120 time period. Significant differences in the essential amino acid levels in the blood of AMI and non-AMI patients were observed primarily at the 70–120-h time period (fig. 2). Of the nonessential amino acid levels (fig. 3) no differences between AMI and non-AMI patients were observed for alanine; fluctuations in levels were demonstrated for aspartic acid (15–20 h), glutamic acid (5–10; 70–120 h) and serine (30–40 h).

*Discussion.* In this study comparisons of the concentrations of essential and nonessential amino acids in the blood of AMI and non-AMI patients demonstrated only few differences at early time periods and thus differed from the pattern obtained for taurine. We thus conclude that the elevation of taurine levels is unique as denoted by its early rise after myocardial injury.

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## Strain- and age-dependent change in carrier independent helper capacity

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*Summary.* Throughout life the immune system undergoes progressive changes. We report, here, that carrier-independent helper capacity increases during adult life and that the rate of the increase varies in different mouse strains, i.e. is polymorphic.

The relative capacity of regulatory and executive cells of the immune system changes from sexual maturity to old age; the changes are polymorphic. So far, age-related changes of immune regulation have been observed in terms of decline in specific suppressor or amplifier suppressor cells[2], increase in nonspecific non-T[3] and T[4] suppressor cells, and increase in anti-auto-idiotypic antibody[5]. Changes in effector functions have been observed in terms of B-cell subpopulations[6] and of T cells in delayed hypersensitivity[7].

Help for a secondary response to a determinant can be dependent on previous exposure to other determinants of the macromolecule; it can take place if in vitro stimulation occurs in the presence of the sensitizing macromolecule,

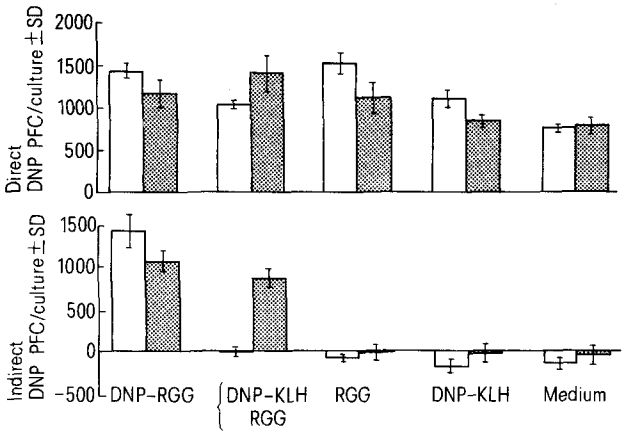


Figure 1. Age-dependent change in help for indirect in vitro plaque-forming response when the carrier of hapten is not identical with carrier used in sensitization. SJL mice, 4 or 28 weeks of age, were injected i.p. with 500 µg DNP-RGG on alum. The animals were sacrificed 8 weeks later. Suspensions of their spleen cells were cultured ($4 \times 10^6$ cells/culture well) in the presence of 0.1 µg antigen, specified on the horizontal axis below the vertical columns. The direct and indirect plaque-forming response was measured with sheep red cells conjugated with dinitrophenylated-human serum albumin. Columns show the number of plaque-forming cells per culture ± 1 SD. Open columns show the response of 4-week-old animals, solid columns indicate the response of 28-week-old animals.

but with hapten conjugated to a different carrier (Th-2 type of carrier independent help). Recently, it has become apparent that there are age-dependent changes not only in suppressor but also in specific[6] and in non-specific helper activity which is independent of the carrier bridge[2,8,9]. We wish to report, here, on the polymorphism of age-dependent changes in the latter.

The nature and age-dependent change of this type of help was tested by direct and indirect plaque formation, with sheep erythrocytes to which haptenated human serum albumin was attached. Direct plaque formation is predominantly due to IgM antibody. The indirect plaque forming response is primarily due to antibody molecules with 2 combining sites and is revealed by the addition to the test system of antibody, directed against IgG. The resulting data are illustrated in figure 1, which shows the in vitro response of spleen cells from SJL/J mice, injected i.p. with 500 µg dinitrophenylated rabbit gamma globulin (DNP-RGG) on alum and treated, 8 weeks later, with various antigens or mixtures of antigens. It will be seen from figure 1 that a hapten-specific indirect plaque-forming response cannot be induced with dinitrophenylated keyhole limpet hemocyanin (DNP-KLH), i.e. to the hapten attached to a carrier to which the animal was not sensitized. In older, but not in young animals the anti-hapten response can be elicited by DNP-KLH if the sensitizing carrier molecule, i.e. rabbit

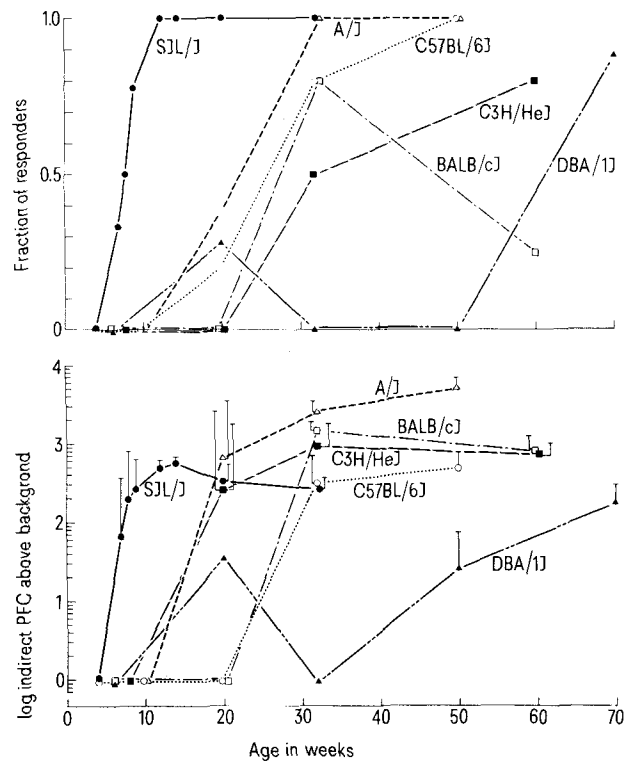


Figure 2. Th-2 type of helper effect in the indirect PFC response to dinitrophenyl. Mice were immunized with 100 µg of DNP-conjugated KLH on alum at the ages shown on the horizontal axis and sacrificed 8 weeks later. Suspensions of spleen cells were cultured for 4 days, in the presence of RGG coupled to the same hapten and KLH. The upper panel shows the fraction of responding animals, plotted on the vertical axis. Responders are defined as animals whose indirect PFC response exceeded by 2 SD that of nonresponding young animals.The lower panel shows the average indirect PFC plotted on the vertical axis. Vertical bars show 1 SD above mean.

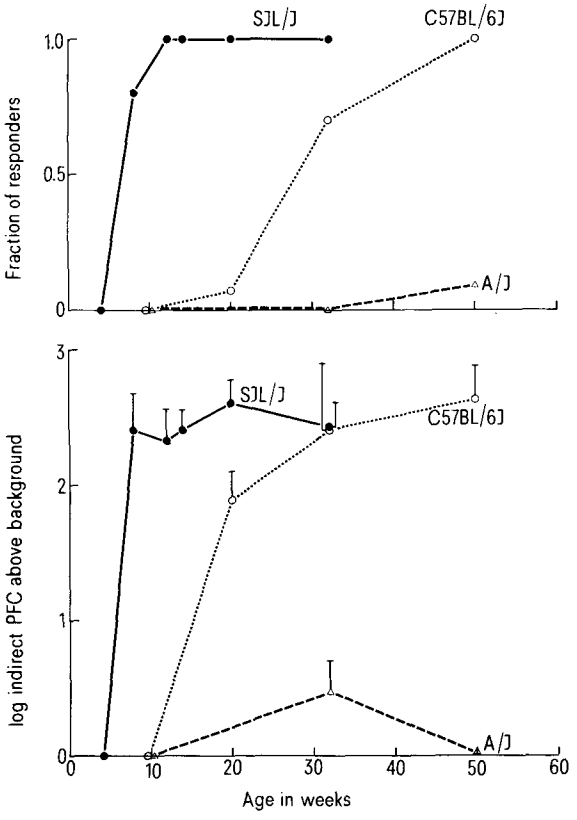


Figure 3. Th-2 type of helper effect in the indirect PFC response to para-azobenzene sulfonic acid. Mice were immunized with 100 µg of PAS-conjugated KLH on alum at the ages shown on the horizontal axis and sacrificed 8 weeks later. Suspensions of spleen cells were cultured for 4 days, in the presence of RGG coupled to the same hapten and KLH. The upper panel shows the fraction of responding animals, plotted on the vertical axis. Responders are defined as animals whose indirect PFC response exceeded by 2 SD that of nonresponding young animals. The lower panel shows the average indirect PFC plotted on the vertical axis. Vertical bars show 1 SD above the mean.

gamma globulin (RGG), is added to the tissue culture. This response could be elicited with various different macromolecules used for sensitization or as carriers.

The polymorphism of the phenomenon was explored by immunizing mice at various ages with DNP-KLH and then exposing spleen cells from these mice to DNP-RGG and KLH. Differences in age at which carrier-independent helper capacity is acquired were studied with 6 different inbred strains of mice, and are shown in figure 2. The age at which activity was observed varied markedly and occurred at the earliest age in SJL and at the latest in DBA/1. Age-dependent changes were also observed with another hapten, para-azobenzene sulfonic acid (PAS; fig. 3). In this case, the response depended on the availability of hapten-primed B cells, but even if they were available, the indirect plaque-forming hapten response was lower than that observed in the indirect plaque-forming response to DNP. The carrier-independent helper capacity of SJL was again detected relatively early. It will be noted that the development of helper capacity in A/J was greatly delayed by comparison to its appearance in C57BL/6 (fig. 3), whereas in the indirect PFC response to DNP (fig. 2) the carrier-independent helper capacity of C57BL/6 and A/J became demonstrable at comparable ages.

The difference between PAS and DNP need for hapten-primed B cells is presumably attributed to 'naturally' induced DNP sensitization, as a concomitant of aging.

In short, we have observed a helper capacity which is evoked by an antigen-specific event, but can provide help to memory B cells, irrespective of the carrier. This helper capacity increases with age at a rate that shows remarkable polymorphism.

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## *O,p'*-DDT causes growth of an androgen-dependent gland in *Coturnix* quail[1]

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*Summary.* *O,p'*-DDT injected into adult castrated male quail *(Coturnix coturnix japonica)* stimulated growth of the proctodeal (foam) gland, a structure that is androgen but not estrogen responsive, indicating that this pesticide constituent, in addition to its known estrogenic actions, is also androgenic.

*O,p'*-DDT, which constitutes about 15–20% of commercial DDT[2], has been shown to have estrogenic actions at several target organs; it causes growth of immature mouse uteri, chick oviducts, and quail oviducts[3,4], and causes morphological feminization of avian embryos[2,5]. It binds to cytosol estrogen receptors[6], and may also bind to androgen receptors, because it has been shown to inhibit binding of 5α-dihydrotestosterone (DHT) in the rat prostate[7].

The proctodeal gland, unique to the genus *Coturnix*, is a foam-producing protuberance posterior to the cloaca that can be externally measured[8,9]. It regresses in castrates, and is restored to intact size by treatment with testosterone (T)[10]. In vitro, the proctodeal gland of mature males primarily metabolizes T to DHT[11]. In vivo, DHT or its propionate (DHTP) are as effective or more effective than T or TP at stimulating growth of the gland[11,12], while estradiol is totally ineffective[12,13], suggesting that growth is mediated by DHT receptors.

For the present study, adult male quail housed on a 16 h light: 8 h dark light:dark cycle were bilaterally castrated under Nembutal anesthesia; 1 week later (day 1) they were examined to ensure that all proctodeal glands had regressed, and were randomly assigned to 1 of 4 treatment groups. Each bird was injected in the pectoral muscle twice daily for 16 days; the 2 injections were 0.5–2 h apart. 1 group received injections of the sesame oil vehicle. A 2nd group received oil injections on days 1 and 2, and thereafter received oil followed by 0.5 mg 5α-DHTP (Steraloids). A 3rd group received 1.0 mg *o,p'*-DDT (Aldrich) followed by oil on days 1 and 2, and 1.0 mg *o,p'*-DDT followed by 0.5 mg 5α-DHTP thereafter. A 4th group received 1.0 mg *o,p'*-DDT followed by oil each day. These procedures were designed to facilitate detection of any antiandrogenic as well as androgenic actions of *o,p'*-DDT.

Proctodeal glands were measured according to standard procedures[9] on day 4 and every other day thereafter. At the end of the experiment each bird was weighed, killed, and examined to confirm the absence of testicular tissue. All procedures were then repeated with new males, and the data from both experiments were combined.

*O,p'*-DDT had no deleterious effect on overall condition. Weights are shown in the table; none of the means are significantly different from each other. As the figure indicates, DHTP markedly stimulated proctodeal gland growth,

Body weights of the males at the end of the experiment

| Treatment | N | Weight (g, $\bar{X} \pm$ SEM) |
|---|---|---|
| Oil | 11 | 133±3 |
| DHTP | 11 | 154±4 |
| *o,p'*-DDT+DHTP | 11 | 150±4 |
| *o,p'*-DDT | 13 | 138±4 |

DHTP = 5α-dihydrotestosterone propionate.