MICE WHOSE B CELLS CANNOT PRODUCE THE T15 IDIOTYPE ALSO LACK AN ANTIGEN-SPECIFIC HELPER T CELL REQUIRED FOR T15 EXPRESSION*

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Several studies have shown that T cells serve an important function in regulating the production of antibodies bearing a given idiotype (1–4). T-dependent anti-hapten antibody responses to hapten-carrier conjugates require cooperation of hapten-specific B and carrier-specific T lymphocytes (5). Although this interaction might be expected to activate any B cell binding the hapten, some responses are characterized by the selective activation of certain clones of hapten-specific B cells that produce antibody bearing a particular idiotype. This selective activation appears to involve the participation of a T-helper cell population which is functionally different from carrier-specific T-helper cells and which recognizes idiotypic determinants on B cells (6–8). The preferential activation of idiotype-bearing B cells in these responses may therefore reflect the fact that these B cells have two sources of T-cell help.

The initial generation and expansion of an idiotype-recognizing T-cell population may in turn depend on the quantity of idiotype expressed on lymphocytes or circulating immunoglobulin. In fact, it has been demonstrated that the expression of an allotype influences a population of helper T cells required for the production of that allotype (9). If idiotype-recognizing helper T cells are influenced by the level of idiotype expression in the T-cell donor, then one might predict that mice which do not express an idiotype would have reduced levels of idiotype-recognizing helper T cells. In the present experiments, we have studied the activity of idiotype-recognizing T cells from (CBA/N × BALB/c)F_1 mice. F_1 male mice are hemizygous for the X-linked immune defect (*xid*), one manifestation of which is the inability of their B cells to respond to phosphorylcholine (PC)[1] (10). The F_1 female mice are phenotypically normal and have high levels of naturally occurring anti-PC antibody, most of which expresses the T15 idiotype. In contrast, the defective F_1 male mice have no detectable anti-PC antibody and consequently, no circulating T15 idiotype. The effect of the level of naturally occurring T15 on the expression of idiotype-recognizing T-helper cells was tested in an adoptive transfer system using keyhole limpet hemocyanin (KLH)-primed helper T cells from either F_1 male or F_1 female donors and PC-primed B cells from the F_1 female and measuring the PC-specific plaque-forming cell (PFC)

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[1] *Abbreviations used in this paper:* BGG, bovine gamma globulin; CFA, complete Freund's adjuvant; CGG, chicken gamma globulin; KLH, keyhole limpet hemocyanin; OVA, ovalbumin; PC, phosphorylcholine; PFC, plaque-forming cells.

response to PC-KLH. We found that T cells from F_1 male and female donors were equally effective in providing help for an anti-PC antibody response. Although T cells from the F_1 female mice induced anti-PC responses dominated by the T15 idiotype, the T cells from the F_1 male donors induced a response which was low in the T15 idiotype. The failure of T cells from the F_1 male mice to induce a T15-predominant anti-PC response seemed to be a result of a deficiency of helper T cells necessary for T15 production, because the production of T15 idiotype could be restored by the addition of F_1 female T cells to the helper T cells from KLH-primed male donors. In addition, these F_1 female T cells necessary for idiotype production required carrier priming and re-exposure to the appropriate carrier antigen, but not to PC, to preferentially induce $T15^+$ B cells. These results suggest that the level of expression of T15 on cells or as circulating immunoglobulin is important in generating helper T cells which affect idiotype expression. During a response to PC, these helper T cells, which require carrier-priming and boosting to be activated, can help PC-specific, T15 idiotype-bearing B cells. They can be activated by free carrier, yet their function does not depend on recognition of the carrier linked to the hapten.

It seems clear from these studies that the presence of a population of helper T cells specific for idiotype and carrier results in the selection of PC-specific B cells producing antibodies bearing the T15 idiotype.

Materials and Methods

Mice. 8- to 12-wk-old (CBA/N × BALB/c)F_1 mice were obtained from our own colonies at The Institute for Cancer Research. Because the defect of CBA/N mice is X-linked, the mating of CBA/N female mice to BALB/c males produces F_1 males expressing the defect and F_1 females who are phenotypically normal.

Antigens. PC-KLH and PC-bovine gamma globulin (BGG) were prepared as described previously (11, 4). The degree of substitution was 12 mol of PC per 100,000 daltons KLH and 9 mol of PC per mole of BGG. PC-Ficoll (Pharmacia Fine Chemicals, Div. of Pharmacia Inc., Piscataway, N. J.) was prepared as described by Inman (12) from the aminoethylcarbamylmethyl derivative of Ficoll. PC-Ficoll had 83 aminoethylcarbamylmethyl groups and 57 PC groups per mole of Ficoll.

Anti-T15 Idiotypic Antibodies. The preparation and purification of anti-T15 antibodies has been described previously (4).

Immunizations. (CBA/N × BALB/c)F_1 mice to be used as T-cell donors were immunized with either 20 μg of KLH, ovalbumin (OVA), or chicken gamma globulin (CGG) in complete Freund's adjuvant (CFA). (CBA/N × BALB/c)F_1 female mice to be used as B-cell donors were immunized with 100 μg of PC-BGG in CFA. All donors were used 6 wk after priming.

Fractionation of Spleen Cells. T cells were obtained by passage of primed spleen cells over nylon wool columns as described previously (13, 4). The yield of passed cells was usually between 10 and 20% of the original spleen population. Staining of these cells with fluorescein-ated rabbit anti-mouse immunoglobulin revealed <5% contaminating Ig^+ cells. B cells were obtained after treatment of spleen cells with a 1:20 dilution of hybridoma anti-Thy 1.2 (kindly provided by J. Sprent) for 30 min at 4°C. The cells were subsequently exposed to a 1:4 dilution of guinea pig complement preabsorbed with (CBA/N × BALB/c)F_1 liver cell suspension, for 30 min at 37°C. This procedure was then repeated. These cells were unable to respond to concanavalin A and were unable to generate an adoptive secondary antibody response.

Adoptive transfer. (CBA/N × BALB/c)F_1 male or female mice irradiated with 650 rad were used as recipients. Graded numbers of primed cells, 25 μg PC-KLH, and as appropriate, 10 μg OVA or CGG were injected, i.v., into the irradiated recipients. Their spleens were assayed for PFC 8 d after cell transfer. All groups contained three to five mice. All experiments presented were repeated at least four times with similar results.

Hemolytic PFC Assay. Spleen cells were assayed for direct anti-PC PFC by the modified Jerne hemolytic plaque technique (14) as described previously (4).

Inhibition of Plaque Formation. The proportion of anti-PC PFC shown to be of the T15 idiotype was determined by inhibition of plaque formation using rabbit anti-T15 in the agarose suspension. The relative avidity of the $T15^+$ and $T15^-$ antibody produced was determined by adding different molar concentrations of PC (in the form of PC-Ficoll) into the agarose suspension and the complement source and calculating the number of $T15^+$ and $T15^-$ PFC inhibited at each molar increment of PC. All the PFC were shown to be inhibited by PC-caproic acid paranitrophenyl ester coupled to the protein MOPC 315 or 10^{-4} mol free PC to verify the fact that the antibodies were specific for the hapten PC and not the hapten-carrier bridge region.

Results

Helper Activity for the T15 Idiotype of T Cells from KLH-primed F_1 Female or Defective F_1 Male Donors. It has been well established that the level of idiotype expression by B cells after antigenic stimulation may be influenced by populations of regulatory T cells (1–4). In our laboratory, we have been able to show that the presence and initial priming of these regulatory cells may depend on the level of naturally occurring circulating idiotype (15).

To study the importance of the expression of naturally occurring T15 idiotype in generating T-helper cells necessary for inducing PC-specific B lymphocytes to produce high levels of $T15^+$ anti-PC antibodies, the helper activity of T cells derived from KLH-primed (CBA/N × BALB/c)F_1 female donors was compared to those from F_1 male donors. These two T-cell populations were tested for their ability to collaborate with B cells from PC-primed F_1 female donors. The source of B cells remained the same; only the source of T cells varied.

The results in Table I demonstrate that T cells from both KLH-primed F_1 female

TABLE I

Helper Cells Needed for T15 Idiotype Production are Diminished in the (CBA/N × BALB/c)F_1 Males

Group	(CBA/N × BALB/c)F_1* T-cell donor	Geometric mean (x/+ rel. SE)‡ PFC/spleen			T15
	KLH-primed ($\times 10^{-6}$)	PC-PFC	$T15^+$-PFC	$T15^-$-PFC	
					%
I	0.5♀	2,971 (1.28)	2,487 (1.33)	459 (1.03)	84
	1.0♀	5,217 (1.06)	4,582 (1.05)	631 (1.12)	88
	2.0♀	9,059 (1.04)	7,202 (1.01)	1,832 (1.15)	80
II	0.5♂	1,539 (1.24)	207 (1.86)	1,269 (1.15)	13
	1.0♂	2,914 (1.08)	792 (1.08)	2,109 (1.12)	27
	2.0♂	6,118 (1.06)	1,680 (1.33)	4,228 (1.17)	27
III	2♀ + 2♂	17,505 (1.07)	10,372 (1.13)	7,067 (1.02)	59

* 5 million B cells from PC-primed F_1♀ donors were transferred along with 0.5, 1.0 or 2.0 million T cells from either KLH-primed F_1♀ or F_1♂ donors into irradiated F_1♀ recipients.

‡ The geometric mean (standard error) from each group represents the responses of six to eight individual mice in three replicate experiments. The number of anti-PC PFC shown to be T15 negative was determined by inhibiting T15-positive PFC with rabbit anti-T15 antibodies. The number of T15-positive PFC was determined by subtracting the T15-negative PFC response from the total anti-PC response. The background PFC responses of T and B cells transferred alone was subtracted.

mice and defective F_1 male mice collaborated effectively with B cells from PC-primed F_1 female donors in the response to PC-KLH. This was true at all T-cell numbers tested. It can be seen that the PFC response to PC-KLH induced by T cells from KLH-primed F_1 female donors is dominated by the T15 idiotype (group I). By contrast, the expression of the T15 idiotype was considerably lower when the response was induced by T cells from F_1 male donors (group II). A mixture of the T-helper cell populations induced a response to PC-KLH in which both the total anti-PC response and the expression of T15 idiotype were additive (group III) demonstrating a lack of suppressor effects from either T-cell population. Carrier priming was required for any helper activity regardless of the source of the T cells (controls not shown).

To eliminate the possibility that these differences were a result of hormonal influences, all the experiments reported here were repeated by comparing cells from defective (CBA/N × BALB/c)F_1 male mice with those from the phenotypically normal reciprocal (BALB/c × CBA/N)F_1 male mice, and similar results were obtained.

This set of experiments suggests that T cells from F_1 male mice have a reduced ability to stimulate idiotype-bearing B cells to produce anti-PC antibody, and the presence of the additional helper function for idiotype may require expansion by exposure to immunoglobulin bearing the T15 idiotype.

Characteristics of a Separate Helper Cell Population Necessary for Idiotype Production. Because the difference in the level of help for idiotype production may be a result of a separate helper T cell whose expansion depends on the availability of idiotype, it might be expected that the addition of F_1 female T cells primed with an irrelevant antigen to the T cells from KLH-primed F_1 male donors would provide enough help to induce a dominant idiotype response. The experiments in Table II

TABLE II

Activation Requirements for Helper Cells Needed for T15 Production

Group	(CBA/N × BALB/c)F_1* T-cell donor		Antigen ± free carrier‡	Geometric mean (×/÷ rel. SE)§ PFC/spleen			T15
	KLH-primed	OVA-primed		PC-PFC	$T15^+$-PFC	$T15^-$-PFC	
							%
I	♀		PC-KLH	10,050 (1.16)	8,611 (1.21)	1,984 (1.23)	81
II	♂		PC-KLH	6,393 (1.03)	1,679 (1.16)	4,775 (1.05)	26
III	♂	♀	PC-KLH	9,170 (1.04)	2,840 (1.12)	6,298 (1.01)	31
IV	♂	♀	PC-KLH + OVA	10,441 (1.07)	7,369 (1.08)	3,067 (1.05)	71
Controls							
V	♀	♀	PC-KLH	12,144 (1.25)	9,472 (1.33)	2,523 (1.07)	78
VI	♂	♂	PC-KLH + OVA	6,505 (1.05)	1,673 (1.16)	4,744 (1.13)	26
VII	♂		PC-KLH + OVA	7,011 (1.15)	2,003 (1.22)	4,986 (1.23)	29
VIII	♂	♀	PC-KLH + CGG	10,065 (1.40)	3,613 (1.33)	6,404 (1.45)	36
IX		♀	PC-KLH + OVA	1,196 (1.21)	1,167 (1.22)	12 (2.42)	99

* F_1♀ recipients received 5 million B cells from PC-primed F_1♀ donors and 2 million T cells from each T-cell donor.

‡ Each recipient received 25 μg PC-KLH ± 10 μg OVA or CGG.

§ See footnotes to Table I.

again demonstrate that T cells from KLH-primed F_1 male donors have far less ability to help F_1 female B cells produce the T15 idiotype than T cells from KLH-primed F_1 female donors (groups I and II). If T cells from F_1 male mice primed with KLH are combined with T cells from F_1 female mice primed to OVA, an anti-PC response predominated by the T15 idiotype is produced if the system is boosted with PC-KLH plus OVA (groups III and IV). It can be seen that although helper cells for T15 idiotype production existed among the helper T cells of the inappropriately OVA-primed F_1 female population, they required activation by the addition of free OVA to induce a T15-dominated response to PC-KLH. The failure of T cells from KLH-primed F_1 male donors to induce a substantial $T15^+$ anti-PC response was neither changed by the presence of free carrier alone nor by T cells from OVA-primed F_1 male donors in the presence of OVA (groups VI and VII). The addition of T cells from carrier-primed F_1 female donors to T cells from KLH-primed F_1 male donors in the presence of PC-KLH and an inappropriate free carrier was unable to induce a T15-dominated anti-PC response (groups VIII). These results indicate that not only do T cells from the F_1 female donors include a population of T cells which can preferentially induce idiotype-bearing B cells to produce antibody, but also that the T cells necessary for idiotype production once activated by antigen can act on a B cell without the need for the hapten to be physically linked to the carrier. Furthermore, the T-cell population needed for a dominant production of idiotype is ineffective in inducing a $T15^+$ anti-PC response alone (group IX), although the small anti-PC response generated is 99% $T15^+$.

The Influence of Helper T Cells on the Avidity of the Antibody Response of $T15^+$ and $T15^-$ B Cells. Previous studies have shown that the response to PC on a variety of carriers is characterized by the production of antibodies of restricted avidity (16–19). Because our data demonstrates that a population of T cells could selectively activate B cells producing T15, we were interested in demonstrating that the selection of $T15^+$ B cells by a T-cell subset corresponded with a shift to production of anti-PC antibody having restricted avidity. The results in Fig. 1 (panels A and B) illustrate the PC inhibition profiles of $T15^+$ and $T15^-$ antibody responses induced by T cells from KLH-primed F_1 male and F_1 female donors, respectively. In all cases shown, 80–90% of all $T15^+$ anti-PC antibodies are inhibited with 10^{-7} and 10^{-8} mol PC. This result illustrates, in this system, that the $T15^+$ B cells producing antibodies to PC are of restricted avidity. By contrast, the inhibition profiles of the $T15^-$ anti-PC responses are considerably more diverse and are, on the average, of higher avidity.

The results illustrated in panels C and D clearly confirm that the transfer of helper cells from OVA-primed F_1 female donors along with T cells from KLH-primed F_1 male donors induces an anti-PC response to PC-KLH characterized by dominant T15 production which is of restricted avidity. As before, the functional activation of these helper cells needed for idiotype production depends on reexposure to the priming antigen.

Discussion

The experimental results presented above lead us to three main conclusions. First, dominant production of the T15 idiotype in the antibody response to PC-proteins requires two helper T cells, one of which is a conventional antigen-specific helper T cell and another cell which regulates the level of idiotype expression. Second, the

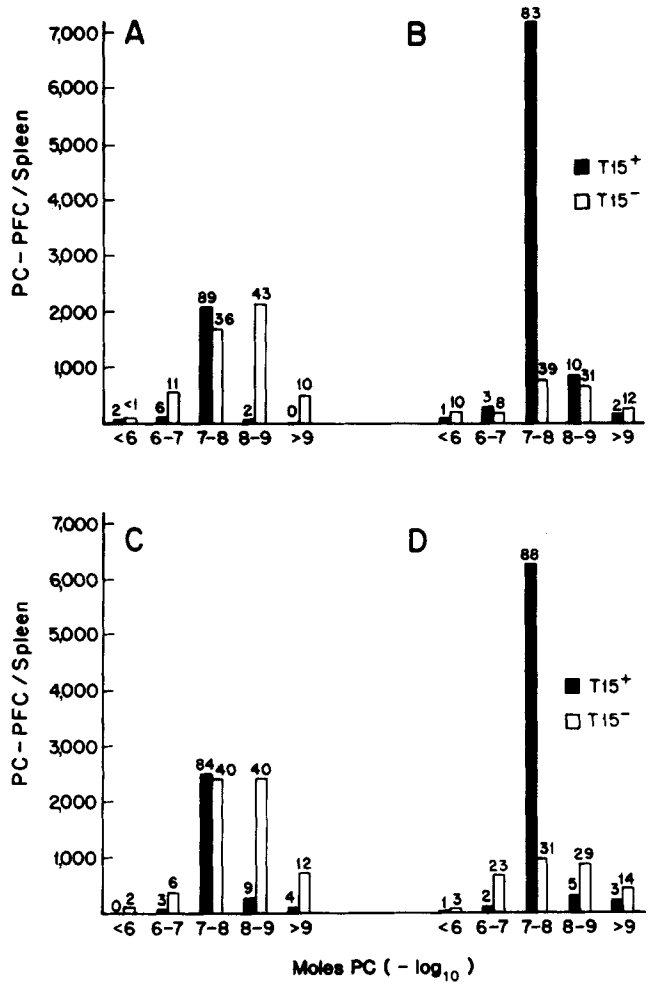

FIG. 1. The relative avidity of the $T15^+$ and $T15^-$ anti-PC antibody produced was determined by adding different molar concentrations of PC into the agarose suspension along with rabbit anti-T15. The number of $T15^+$ and $T15^-$ PFC inhibited at each molar increment of PC was calculated. All inhibition profiles illustrate responses of B cells from PC-primed F_1 female donors to PC-KLH induced by T cells from KLH-primed F_1 male donors (panel A), T cells from KLH-primed F_1 female donors (panel B), T cells from KLH-primed F_1 male donors and OVA-primed F_1 female donors (panel C), and T cells from KLH-primed F_1 male donors and OVA-primed F_1 female donors in the presence of free OVA (panel D). The percentage of $T15^+$ and $T15^-$ PFC inhibited at each molar increment of PC is shown at the top of each bar.

helper T cell which is required for dominant production of the T15 idiotype also is antigen (carrier) specific, but its activation does not require that PC be covalently coupled to the carrier. Third, the activity of the helper T cell required for dominant production of the T15 idiotype seems to be directly correlated with the level of circulating T15 idiotype in the T-cell donor.

These experiments originated with the observation that F_1 male offspring of the X-linked immune deficient mouse strain CBA/N crossed with BALB/c fail to respond to PC on any carrier and have no detectable T15 idiotype in their circulation (10).

Female offspring, which do not express phenotypically the CBA/N defect, respond well to PC and have abundant T15 idiotype in their serum. The present experiments demonstrated that the ability of helper T cells from KLH-primed (CBA/N × BALB/c)F_1 male and female mice to induce B cells from F_1 female donors to respond to PC-KLH differed. Although helper T cells from both donors were equally effective in generating anti-PC antibody responses, T cells from the F_1 male donors failed to induce an anti-PC response which was dominated by the T15 idiotype. This inability of T cells from KLH-primed F_1 male donors to generate a T15-dominated response could be corrected by adding F_1 female T cells primed with OVA and boosting the recipients with a mixture of PC-KLH and OVA. These results suggest that the F_1 male mice lacked a helper T cell required for dominant production of the T15 idiotype. Nevertheless, F_1 male T cells primed with KLH do provide a helper T cell which is required for substantial production of anti-PC antibody (Table II), and the activation of this helper T-cell population requires covalent linkage of the hapten and carrier. The F_1 female cells primed with OVA provide a helper T cell which is also specific for its priming carrier. Moreover, mixtures of these two sets of cells induce large numbers of T15 idiotype-positive anti-PC antibody-forming cells. Thus, both helper T cells show precise specificity for the priming antigen. One requires hapten and carrier to be present on the same molecule. The other would appear to be specific for idiotype, because it induces selective activation of T15-bearing B cells, and does not require physical linkage of hapten and carrier. It is interesting to note that preferential activation of T15-positive B cells in this system does not represent the selection of B cells of the highest avidity, because the anti-PC response induced by F_1 male helper T cells alone actually has more high avidity PFC than that induced by F_1 female helper T cells.

The deficiency of helper T cells for T15 idiotype production in F_1 males correlates with their lack of circulating T15 idiotype. Although it is possible that this finding is a result of the expression of the CBA/N defect directly at the T-cell level, this seems unlikely for two reasons. First, F_1 male T cells have been shown to express T15 idiotype in levels similar to those found in F_1 female T cells (20). Second, similar results are obtained using helper T cells from mice depleted of B cells and circulating T15 idiotype by injections of anti-μ chain antibody from birth.[2]

If one accepts that the synergistic action of two helper T cells elicits optimal production of T15 idiotype in the T-dependent antibody response to PC, then it is interesting to determine whether either helper T cell acting alone can activate a B cell. Although this question cannot be definitively answered, our data would suggest the following. Helper T cells from F_1 males do elicit a good response to PC, and some of the cells produce T15 idiotype. It is our interpretation that this result represents the activity of conventional helper T cells which can activate B cells capable of binding PC-carrier independent of which idiotype the B cells bear. The 30% of $T15^+$ idiotypic B cells activated would represent the frequency of T15-positive precursors in the PC-binding B-cell pool. Alternatively, one could argue that F_1 males do have a limited number of T15 recognizing helper T cells, whose activity might result from

[2] Bottomly, K., C. Janeway, B. J. Mathieson, and D. E. Mosier. Absence of an antigen-specific helper T cell required for the expression of the T15 idiotype in mice treated with anti-μ antibody. Manuscript submitted for publication.

exposure to the T and B cells bearing the T15 idiotype found in these mice (20, 21). The helper T cells derived from OVA-primed F_1 female mice by themselves elicit very small anti-PC responses. Such antibody as is produced is, as expected, entirely of the T15 idiotype. Clearly, the synergistic behavior of these two sets of helper T cells gives rise to optimal idiotype production. We could conclude therefore that the helper cell population with conventional specificity is effective by itself in B-cell activation, whereas the anti-idiotypic helper cell set is relatively ineffective when acting alone.

Data from other systems have also suggested that there exist two types of antigen-specific helper T-cell populations which act synergistically to induce optimal antibody responses[3] (22, 23). In these systems, one of the helper T-cell populations appears not to require that hapten and carrier to be physically linked. Similarly, the requirement for two synergizing helper T cells for the optimal production of idiotype-bearing antibody in vivo (6) and in vitro (7, 8) has also been demonstrated. The studies of Woodland and Cantor (6) have shown a requirement for two helper T cells in the production of idiotype-positive antibody. However, in their system, only one of the helper T cells was antigen specific. They showed that KLH-primed mice depleted of those helper T cells required for idiotype production by various means could be reconstituted with helper T cells from mice primed with BGG. Although not deliberately stimulated with antigen, these cells may have become activated by BGG present in the fetal calf serum through which they were processed during adoptive transfer. If this were so, then their results would be more compatible with our findings.

The concept that B cells and their products may determine the types and quantity of regulatory T cells found in mice is supported by findings in other systems (24, 25). In particular, Janeway and coworkers (24) demonstrated the absence of a synergizing T cell needed for an optimal anti-DNP antibody response in mice suppressed from birth with anti-μ antibody and consequently having no B cells. Likewise, our own preliminary data[2] suggest a deficiency in helper T cells for the T15 idiotype in mice similarly suppressed with anti-μ antibody. Therefore, we propose that helper T cells which selectively activate idiotypic B cells are themselves initially selected for by the idiotypes present in the mice from which they are obtained.

We envision that the antigen-specific T-cell population defined here, which regulates the production of the T15 idiotype, selects B cells bearing the T15 idiotype by means of an anti-idiotypic receptor. Various studies have shown the selective activation of B cells bearing particular isotypes (26), allotypes (9), or idiotypes (6–8). Furthermore, Woodland and Cantor (6) have succeeded in depleting helper T cells required for optimal production of idiotype on plastic surfaces coated with idiotypic molecules. If the helper T cell required for optimal production of the T15 idiotype does bear an anti-idiotypic receptor, then this would allow this T cell to signal B cells selectively without the requirement for physical association to hapten and carrier. The simplest explanation for these findings is to postulate that these helper T cells bear two receptors, one for their association with T15-bearing B cells and the other for their activation with the carrier antigen. This dual specificity is similar to that seen with PC-specific suppressor T cells generated by pretreatment with anti-T15 antibody and carrier priming (4). It seems possible that the two helper T-cell

[3] Janeway, C. A., D. L. Bert, and F-W. Shen. Cell cooperation during in vivo anti-hapten antibody responses. V. Evidence for two distinct Ly 1^+23^- helper T cells with distinctive specificities. Manuscript submitted for publication.

populations needed for a predominantly T15-positive anti-PC antibody response are, in fact, two different specificity types of the same population of T cells; both having specificity for antigen (carrier) and self determinants, either self MHC determinants (27, 28) or alternatively self idiotypic determinants (29). It is unlikely that our findings can be accounted for by the presence of two synergizing T cells in the F_1 female, OVA-primed population, one specific for idiotype and one specific for OVA. If this latter explanation of our results were correct, it is difficult to understand why the antigen-specific cell would not be found in the KLH-primed F_1 male T-cell population. Our studies suggest that the helper T cells needed for T15 idiotype production are induced by the presence of idiotype itself. Although the expansion of individual components of the system appears to depend on anti-idiotype-idiotype interactions (30), the activation of the individual members of this network appears to require, in addition, exposure to antigen.

Summary

The X-linked CBA/N defect in B cell function precludes an antibody response to phosphorylcholine (PC). Accordingly, (CBA/N × BALB/c)F_1 male mice are unresponsive to PC and lack circulating immunoglobulin bearing the T15 idiotype characteristic of BALB/c anti-PC antibody. In contrast, (CBA/N × BALB/c)F_1 female mice respond to PC and >80% of the anti-PC antibody is $T15^+$. No T-cell abnormalities are known to be associated with the CBA/N mutation. These experiments compared the ability of helper T cells from either (CBA/N × BALB/c)F_1 male ($T15^-$) or F_1 female ($T15^+$) mice to help F_1 female B cells respond to PC and to influence the level of T15 expression. The results indicate that although F_1 male T cells collaborated with F_1 female B cells just as efficiently as F_1 female T cells for the total anti-PC response, the percentage of T15 expression induced by F_1 male T cells fell dramatically. The (CBA/N × BALB/c) F_1 male thus appear to lack a helper T-cell subset required for dominant idiotype production. This helper T cell defect could be repaired by adding F_1 female T cells primed to a second carrier to F_1 male T cells and restimulating the cell mixture with PC coupled to the antigen used to prime the F_1 male cells plus free second carrier. This result implies that conventional helper T cells derived from the F_1 male donor can collaborate with a distinct helper T-cell subset from the F_1 female donor which recognizes both carrier and idiotype to induce an anti-PC antibody response dominated by the T15 clonotype.

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