reacted with unrelated subjects, as have all 100 pairs of unrelated normal subjects studied so far. These results with siblings and our limited data on fraternal twins suggest that siblings and fraternal twins do not differ in their behavior in the mixed leukocyte reaction.

These results are compatible with the laws of inheritance: siblings have, on the average, 50 percent of their chromosomes in common, so that they are more similar to one another than they are to nonrelated members of their species. However, the number of chromosomes that they have in common can theoretically vary from 0 to 100 percent, and an appreciable number of sibling pairs will lie towards one or the other of the two extreme ends of this scale. On the other hand, a child always inherits 50 percent of his chromosomes from his mother and 50 percent from his father.

Thus, in a situation such as the selection of a donor for a kidney homograft, one of the recipient's brothers or sisters might be much more compatible than either parent, while another sibling would be relatively incompatible (*8*). If the mixed leukocyte reaction proves to be effective in predicting histocompatibility, it may be particularly useful as an indicator of compatibility between siblings. The pairs which react most strongly may be those whose genetic similarity lies towards the "completely dissimilar" end of the scale, and those who show no reaction may be placed closer to the "completely identical" extreme.

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Antigenic Behavior of Moloney Lymphomas: Independence of Virus Release and Immunosensitivity

Abstract. *Mouse lymphomas induced by Moloney virus were compared with regard to their ability to elicit humoral antibodies against Moloney cells, to sensitize against Moloney isografts, and to respond to established isograft resistance. The first two properties were parallel, while the third was independent. The former, but not the latter, is attributed to the release of infectious virus.*

Lymphomas induced by the Moloney virus are capable of inducing specific resistance against the transplantation of other Moloney lymphomas to genetically compatible, isologous hosts (*1, 2*). Humoral antibodies reacting with Moloney lymphoma cells can be detected in the serum of resistant animals by the cytotoxic or the indirect fluorescent antibody test (*2*). The resistance can be induced by at least three methods: by inoculating homografts from Moloney lymphomas that fail to grow or regress after temporary growth, by isografting subthreshold numbers of Moloney lymphoma cells, or by inoculating homogenates containing Moloney virus (*2*). All lymphomas tested released virus even after serial passage. A single dose of irradiated tumor cells, incapable of multiplication but competent to release virus, induces formation of antibody to the cells, the antibody lasting throughout most of the lifetime of the recipient animal (*3*). In contrast, treatment of the x-irradiated cells with hydroxylamine caused complete inactivation of its capacity to induce antibody formation.

While all Moloney lymphomas tested induced a specific immunological response against themselves and other Moloney lymphomas as judged by formation of antibody and by resistance to transplantation, the degree of susceptibility to the rejection response of sensitized hosts varied considerably. A number of lymphomas were not transplantable to isologous recipients at all, even if large numbers of cells were inoculated, unless the recipients were irradiated; with large cell numbers others were transplantable to irradiated and nonirradiated hosts alike, while small inoculums often failed to take. The threshold dose in untreated recipients could be diminished by several orders of magnitude by total body irradiation of 400 roentgens. Lymphomas of the latter transferable type were highly susceptible to the rejection response of sensitized hosts. They did not grow progressively even when large numbers of cells were grafted and the hosts were irradiated prior to challenge. On the other hand, certain lymphomas were characterized by a much lower threshold dose, by only small differences in the takes of small inoculums in irradiated (compared to unirradiated) hosts, and by only a minor increase of the threshold cell number or only a slight prolongation of the latency period upon inoculation into specifically presensitized hosts.

Thus, while all tested tumors released virus, they showed very great variations with regard to their sensitivity to the virus-induced rejection response (VIR). From results with the mouse antibody-production (MAP) test for quantitative assay of the virus (*3*), it appeared that the same number of irradiated tumor cells from different lines released different, but for each line fairly constant, amounts of virus, as judged by the antibody titers 35 days after inoculation. The question arose whether differences in the sensitivity to virus-induced rejection are related to the degree of virus release. An experimental study of this question may define the mechanism of the rejection response.

Since the Moloney agent is an RNA virus that matures by budding from the cell membrane, antiviral antibodies may combine with it on the surface of releasing cells, with complement binding and cell lysis as a consequence. If this were the case, a correlation is expected between virus release and sensitivity to virus-induced rejection in different tumors. Alternatively, the two phenomena may be independent, as in the polyoma system (*4*). Sensitivity to such rejection would then be determined by new cellular antigens, and would appear in virus-induced neoplastic cells and not be related to virus release as such. It is also possible to postulate a dualistic scheme where virus maturation and release occur in cells *not* primarily responsible for tumor proliferation. Dividing neoplastic cells would be characterized by a more "moderate" interaction of virus and cell, where genetic informa-

tion derived from the virus nucleic acid is integrated with the host cell at some self-perpetuating template that determines the appearance of new cellular antigen or antigens and sensitivity to virus-induced rejection, together with the neoplastic behavior which may or may not be another expression of the same cellular change that is expressed in the form of antigenticity.

We now report studies on a number of selected Moloney lymphomas designed to disclose any correlation between virus release and sensitivity to virus-induced rejection.

The results show (Table 1) that lymphoma YHA released appreciable amounts of virus as measured by the MAP test and it was also capable of immunizing against the isografting of other Moloney lymphomas. On the other hand, it was rather resistant to virus-induced rejection. Lymphoma YDAB showed a similar behavior, although its sensitivity to virus-induced rejection was somewhat higher than with YHA. The lymphoma YHA is completely resistant (*2*) to the cytotoxic effect in vitro of serums from mice resistant to Moloney lymphoma in spite of the fact that the tumor cells showed a good response in the fluorescent-antibody test. The behavior of lymphoma YLD was different. Judged by the MAP test and the capacity to induce rejection it released considerable amounts of virus. It was highly sensitive to virus-induced rejection, however, and it maintained this property unchanged in the course of the 28 serial passages so far studied. It was also sensitive to the cytotoxic effect in vitro during the whole observation period. Lymphoma Y7A behaved similarly to lymphoma YLD during the five passages observed.

Lymphoma YAA occupied an intermediary position. During the first six transfer generations it behaved like lymphoma YLD but it changed subsequently and became more like lymphoma YHA. It continued to release large amounts of virus, but its sensitivity to virus-induced rejection was replaced by complete resistance after the 7th to 8th transfer generation.

All other Moloney lymphomas tested so far (18 different lines) released considerable amounts of virus, even though the MAP test indicates considerable quantitative differences between them. The most conspicuous difference between the tumor lines concerns the degree of their sensitivity to virus-induced rejection, however. Some are highly sensitive while others are quite resistant. Complete resistance is yet to be found; in fact, lymphoma YHA, the most resistant tumor so far, is somewhat sensitive, showing a prolonged latency period in already sensitized mice when very small cell numbers are inoculated. Some tumors sensitive to virus-induced rejection became resistant in the course of serial passage while others remained unchanged. Insensitivity or change to insensitivity was not correlated with a lesser release of virus; on the contrary lymphomas YHA and YDAB, the most resistant lines, seemed to release larger quantities of virus than the other lines did (Table 1).

Table 1. Schematic summary of main findings with some Moloney lymphomas. VIR, virus induced rejection; TG, transfer generation.

35-day MAP value*	Latency in MAP-test† (days)	Ability to immunize against			Response to VIR		
		Lymphoma (cell and No.)	Transfer generation	Resistance	Sensitizer	Challenge cell dose	Rejection
		YHA (C3H), passages 4–12 TG					
0.88–0.98	< 28	YLD (10^6)	2–10	+	YLD or YAA	10^5	± or —
		YLD (10^6)	16–28	+			
		YAA (10^6)	4–6	+			
		YAA (10^6)	7–9	±		10^6	—, rarely +
		YAA (10^6)	10–30	—			
		YLD C57 leaden, passages 2–28 TG					
0.64–0.95	< 29	YAA (10^6)	2–5	+	YHA	10^6	+, stable
		YAA (10^6)	6–10	± or —	YAA		+, stable
		YAA (10^6)	11–30	—			
		YAA; A, passages 4–6 TG					
0.64	35	YLD (10^6)	6–30	+	YHA	10^6	+
					YLD	10^6	+
		YAA; A, passages 7–27 TG					
0.51–0.59	35	YLD (10^6)	6–29	+	YHA	10^6	gen. 7–9: ±
					YLD	10^6	gen. 10–30:—
							gen. 9–29:—
		YDAB; A×DBA/2F$_1$; passages 2–6 TG					
0.70	20	YLD (10^6)	11–14	+	YHA or YLD	10^4	+, ±, or —
		YLD (10^6)	26–31	+		10^5	+, ±, or —
						10^6	—
		Y7A C57Bl; passages 1–5 TG					
0.84	35	YLD (10^4)	16	+	YHA	10^5	+
		YLD (10^6)	32		YHA	10^6	+
					YDAB	10^6	+
					YAA	10^6	+

* Fluorescent index (*2*), obtained by testing the pooled serum of 2 A×C57Bl F_1 mice 35 days after the subcutaneous inoculation of 5×10^6 lymphoma cells that had been irradiated with 6000 r. Values exceeding 0.3 are considered positive. Target cells: YLD. † Number of days prior to the attainment of a fluorescent index of about 0.5 in the serum of 2 A×C57Bl F_1 mice after the single subcutaneous inoculation of 5×10^6 lymphoma cells irradiated with 6000 r.

It would thus appear that the capacity of a tumor to induce the formation of antibodies specifically reacting with Moloney target cells in the fluorescent test is related to its capacity to sensitize against the grafting of other Moloney lymphomas, both actions probably being dependent on the same quality, namely, virus release. Sensitivity to virus-induced rejection is an independent property, not dependent on virus release. It is perhaps due to the development of new cellular antigen or antigens, determined by genetic information derived from the virus, and not necessarily related to the intracellular multiplication of infectious particles. A moderate, nonreleasing type of interaction may be envisaged, perhaps of the type found in polyoma and apparently also in the Rous system (*5, 6*). If this is the case, it should be possible to isolate nonrereleasing lines of lymphoma cells that are still susceptible to virus-induced rejection.

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