Nuclear Medicine Communications 9, 919–930 (1988)

Human immunological response to mouse monoclonal antibodies in the treatment or diagnosis of malignant diseases

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Received 8 March 1988

Summary

An overview of the literature is presented concerning the formation, detection, incidence and effect of the human immunoglobulin response on immunoscintigraphy. The following conclusions are drawn. The production of human anti-mouse antibodies (HAMAs) is associated with a diminished therapeutic response; adequate prevention of HAMA production is not yet possible; high HAMA titres give rise to rapid clearance of MoAb into the liver and marked reduction of tumour uptake which results in reduced image quality on immunoscintigraphy; alteration of antibody biodistribution is likely to be related to the *in vivo* formation of antibody–antibody complexes which could interfere with image quality when sequential imaging is carried out.

Introduction

Monoclonal antibodies have been used for more than a decade in biomedical research. One of the most exciting and promising areas of research is the use of specific monoclonal antibody radionuclide conjugates for diagnostic imaging (immunoscintigraphy) and therapy for malignant diseases. When these monoclonal antibodies (MoAbs), most of which are developed from mouse hybridomas, are injected into the patient, they are recognized as foreign globulins.

The resulting immune response leads to the development of human anti-mouse antibodies (HAMAs), which can be of practical significance. Once HAMAs have been induced, they are able to neutralize the effects of the MoAbs. Since repeated injections lead to rising HAMA concentrations, the efficacy of this approach may be short-lived. As this is regarded as a major complication of the use of MoAbs for clinical purposes, it is essential to establish the scope of the problem of the production of HAMAs.

0143-3636/88 $03.00+.12

This paper attempts to present an overview of the literature concerning the formation, detection and incidence as well as the effect of the human immunoglobulin antibody response on immunoscintigraphy.

Findings in the literature

Human anti-mouse antibodies (HAMAs)

When a normal individual is exposed for the first time to a foreign antigen, there is a lag phase that may last as long as 10–12 days before antibodies appear in the serum. This primary immune response consists in general of IgM antibodies. Subsequent encounters with the same antigen usually evoke an enhanced secondary or memory response characterized by marked production of IgG [1].

Every antibody has fundamentally the same structure in that it consists of a heavy and a light chain; it also contains a variable and a constant region which may act as antigenic determinants. The antigenic constituents of the variable region of an immunoglobulin are known as its idiotype. The part of the variable region which forms its specific binding site is called its paratope. Thus, it is possible to distinguish between anti-idiotypes directed against idiotypes within the binding site (anti-paratopic) and those directed against idiotypes outside the binding site. Only those binding to the antigen-binding site inhibit the interaction between that binding site and the antigen. Antibodies directed against the constant region are called anti-isotopic antibodies. Jerne [2] postulated a network of interacting antibody molecules and lymphocytes in which idiotypes of antibody molecules are recognized by anti-idiotopic (AB2) antibodies. This AB2 response is probably a very important part of the human immunological response. The immune system may be regulated at least in part by a network of interactions between idiotypes and anti-idiotypes. He also suggested that AB2 antibodies may exert a strong inhibitory effect on B cell clones during the immune response. Of interest is the fact that injection of these AB2 antibodies into the patient can also give rise to HAMAs [3]. Moreover, AB2 antibodies already present in the body have been shown to be potent enhancers or inhibitors of the immune system.

It is known that healthy individuals possess antibodies against various animal proteins [4] and the patients with various malignancies are able to produce a range of antibodies [5]. There are many reports in the literature concerning the presence of pre-treatment anti-mouse antibodies [6–15]. Naturally occurring anti-mouse activity was demonstrated in the serum of 990 of 1008 healthy blood donors by Thompson *et al.* [10]. The aetiology of these pre-existing HAMA levels could be vaccination in the past, animal handling or dietary exposure. Another plausible explanation came from Shawler *et al.* [16], who suggested that such HAMA levels are probably related to the sensitivity of the assay and represent nothing more than background levels caused by non-specific human immunoglobulin. The HAMA levels found by Ritter *et al.* [17] in normal serum were equivalent to the level of endogenous anti-human immunoglobulin (rheumatoid factor) also found in normal serum [18]. It is therefore

reasonable to suppose that pre-existing HAMA levels may merely reflect a facet of the normal immune system. The question of when an HAMA level should be considered indicative of HAMA production should therefore be dependent upon the upper limit of normal. Carrasquillo *et al.* [19] consider a patient HAMA-positive when the percentage binding is at least 3 S.D. greater than the mean for normal individuals. The variety of methods and techniques currently in use for detecting the human anti-mouse antibody response in serum, i.e. radioimmunoassays (RIA), enzyme-linked immunoabsorbent assays (ELISA), haemagglutination tests (HAT) and immunofluorescence assays (IF), makes it difficult to estimate the mean value for normal individuals.

According to the literature, HAMAs are first detected after day 2 [20]. However, the moment of first detection is highly variable, as illustrated by Goodman *et al.* [14] who even found the first detectable HAMA level 233 days after treatment. Therefore, in other studies more patients might have been found to be HAMA-positive if serum samples had been taken at a later stage. The antiglobulin response consists mainly of IgG antibodies, although IgM antibodies have also been observed [7, 9, 11, 15, 21–23]. The rapid elevation of the antiglobulin level reported by several authors [7, 9, 11, 12, 24, 25] is consistent with the kinetics of a secondary immune response, but in general HAMA production occurs 2–3 weeks after MoAb injection; the levels subsequently decrease gradually in the course of several weeks. However, as mentioned above, HAMAs have been detected for up to 300 days after MoAb administration; in fact, in one case an AB2 response persisted [26] for more than 770 days. The fact that antiglobulin levels do not recur indicates that feedback inhibition of the globulin response probably does not occur [9].

The first investigations of the specificity of the antiglobulin response suggested that the response was directed mainly (95%) against the constant region of the MoAb (anti-isotopic), while a minority of the antibodies was directed against the variable region (anti-idiotopic) of the MoAb [7, 11, 27]. However, in man, 50% of the patients receiving the 17-1A monoclonal antibody against a colon carcinoma antigen exhibited an anti-idiotopic AB2 response [28]. Recently, more authors have found that a relatively high percentage of the responses is anti-idiotopic [14, 15, 26, 29]. The difference in results is not yet understood. It is possible that a large percentage of AB2 is followed by anti-anti-idiotopic antibodies (AB3), which could hamper detection of the AB2; another explanation is that the AB2 response is dependent on the type of MoAb. Shawler *et al.* [15] suggest that multiple infusions of a single MoAb will result in a marked specific response, while infusion of two or more monoclonal antibodies may induce only anti-isotopic antibodies.

Variables which influence the development of human anti-mouse antibodies

Of primary interest is the group of variables that determine why some individuals develop an antiglobulin response during immunoscintigraphy or immunotherapy while many others do not. Shawler *et al.* [15] were unable to correlate the lack of response to a large number of clinical parameters, and it still remains difficult to

predict on the basis of clinical data which patients will develop antibodies. Although the development of HAMAs is not related to skin-test positivity [8, 11], the outcome of lymphoproliferative assays [30] or previous therapy, there are some variables that presumably do influence HAMA production.

Table 1. Incidence of HAMAs in patients receiving labelled antibodies for immunoscintigraphy.

Authors	*Year*	*MoAb*	*Pat/HAMA**	*Frequency (%)*
Larson *et al.* [38]	1983	96.5	6/3	50
Carrasquillo *et al.* [50]	1983	96.5	3/3	100
Reynolds *et al.* [51]	1985	96.5	37/12	32
Engelstad *et al.* [22]	1986	96.5	6/3	50
Reynolds *et al.* [32]	1986	T101	20/6	30
Carrasquillo *et al.* [52]	1987	T101	4/0	0
Rosen *et al.* [21]	1987	T101	6/6	100
Reynolds *et al.* [32]	1986	B72.3	30/15	50
Murray *et al.* [13]	1987	ZME018	17/7	41

*No. of evaluable patients in the study/incidence of HAMAs.

In the first place, it has been observed [31] that HAMAs are seldom encountered in patients with B cell malignancies but are frequently found in patients with T-cell or solid tumours. The variation in the incidence of HAMAs (see Tables 1 and 2) probably depends on the immunocompetence of the subjects. This is illustrated by the fact that out of six patients with chronic lymphocytic leukaemia none exhibited an immune response to MoAb T101 (an anti-human T-cell monoclonal antibody) whereas five out of ten patients with cutaneous T-cell lymphoma had measurable HAMA activity [15]. Moreover, the immune system of patients with less advanced disease might be expected to be more competent so that the likelihood that HAMAs will develop would be greater. Theoretically, it is feasible that healthy humans should have a 100% response rate to murine MoAbs. In the second place, mouse whole antibody is more immunogenic than the Fab fragment [32]. However, it has been shown that repeated administration of the murine Fab fragment will also lead to a high frequency of HAMA positivity [15, 19, 20, 24, 26]. In addition, the development of HAMAs may be dose-dependent [9, 20, 21, 28, 30, 33]. Eight out of nine patients who were given less than 200 mg MoAb developed HAMAs compared with only one out of nine receiving higher doses, suggesting that larger doses of MoAb could induce tolerance for murine immunoglobulin [20]. Essentially the same observation was reported by Oldham *et al.* [9], who found measurable increases in anti-globulin response after administration of 50 mg doses and a loss of demonstrable antiglobulin at higher doses. Herlyn *et al.* [26] were not able to confirm this correlation but found instead

Table 2. Incidence of HAMAs in patients undergoing immunotherapy.

Authors	*Year*	*MoAb*	*Pat/HAMA**	*Frequency (%)*
Miller *et al.* [53]	1981	Leu-1	1/1	100
Miller *et al.* [7]	1981	Leu-1	1/0	0
Miller *et al.* [27]	1983	Leu-1	7/4	57
Sears *et al.* [24]	1982	17-1A	4/3	75
Koprowski *et al.* [28]	1984	17-1A	29/10	34
Sears *et al.* [20]	1984	17-1A	18/9	50
Sears *et al.* [29]	1985	17-1A	20/10	50
Herlyn *et al.* [26]	1986	17-1A	42/21	50
Herlyn *et al.* [26]	1986	17-1A	37/32	86
Sears *et al.* [33]	1986	17-1A	65/35	54
Sindelar *et al.* [25]	1986	17-1A	25/23	92
Lobuglio *et al.* [39]	1986	17-1A	20/17	85
Frödin *et al.* [23]	1986	17-1A	8/8	100
Douillard *et al.* [60]	1986	17-1A	20/11	55
Steplewski *et al.* [61]	1986	17-1A	4/3	75
Dillman *et al.* [54]	1982	T101	2/0	0
Dillman *et al.* [56]	1983	T101	6/4	66
Dillman *et al.* [57]	1983	T101	2/0	0
Bunn *et al.* [58]	1983	T101	5/4	80
Foon *et al.* [12]	1984	T101	13/0	0
Dillman *et al.* [36]	1984	T101	8/2	25
Schroff *et al.* [11]	1985	T101	24/7	29
Shawler *et al.* [15]	1985	T101	16/5	31
Bertram *et al.* [30]	1986	T101	13/3	23
Rosen *et al.* [21]	1987	T101	5/5	100
Miller *et al.* [40]	1982	Anti-idio	1/0	0
Meeker *et al.* [3]	1985	Anti-idio	11/5	45
Ball *et al.* [55]	1983	PM81, PMN29	3/1	33
Linch *et al.* [37]	1983	UCHT 1,2	1/1	100
Carrasquillo *et al.* [19]	1984	48.7	8/5	62
Goodman *et al.* [14]	1985	96.5; 48.7	4/4	100
Oldham *et al.* [9]	1984	9.2.27	8/3	38
Houghton *et al.* [59]	1985	R24	12/12	100
Press *et al.* [31]	1987	1F5	4/1	25

*No. of evaluable patients in the study/incidence of HAMAs.

a positive correlation between the number of injections and the occurrence of HAMAs. In addition to the dose of MoAb and the number of injections, the time interval between injections (treatment schedule) has also been associated [19, 20] with HAMA production. Finally, differences in response may be related to pre-existing antiglobulin level [11] or the route of administration. Reynolds *et al.* [32] suggest that there is no apparent difference in HAMA development when an antibody is given subcutaneously

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