
Nonreaginic Anaphylactic Antibodies in Animals

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The antibodies discussed in this article are the homologous antibodies of the IgG class that possess anaphylactic activity. The condition of homology is important; very frequently IgG antibodies produced in one species can sensitize other animal species; but, while they may be useful for many investigative purposes, these sensitizations are not physiologic ones.

IgG anaphylactic antibodies have been demonstrated and well characterized in many mammals: guinea pig,^{1,2} mouse,^{3,4} rat,^{5,6} rabbit,⁷ and sheep.⁸ In man, anaphylactic activity apparently not mediated by IgE has also been described, but this property has not been conclusively assigned to a particular immunoglobulin class. Anaphylactic activity has not been reported in IgA, IgM, or IgD classes of any species.

When IgG anaphylactic antibodies of various species are compared, it appears that they may have very different properties. They all belong to the IgG class but not to the same subclass. The nomenclature and some relevant properties of the antibodies present in some animal species are indicated in Table 1.

The nomenclature adopted by the World Health Organization (WHO) to designate the subclasses of IgG in animals⁹ is mostly based on historical grounds and does not take into consideration the possible structural homologues between subclasses of different species. It is pertinent to remember that homologies, in terms of evolution, may not necessarily exist in all cases because, among other reasons, the different animal species have not been submitted to the same selective pressures. With this notion in mind it will be easy to avoid the confusion that may arise when comparing the nomenclature of antibodies from various species, a problem that frequently appears in the literature, especially when the first descriptions of IgG anaphylactic antibodies were made. Nevertheless, it is clear that, in terms of biologic activity, the existence of IgG anaphylactic antibodies in so many species must reflect a physiologic need and certainly a structural homology whose molecular basis is presently unknown but will probably emerge when the structures responsible for the anaphylactic reaction are discovered and studied in detail.

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Table 1. IgG Anaphylactic Antibodies

Species	Subclass	Electrophoretic mobility	Complement fixation (classical)
Guinea pig	IgG ₄	Fast	No
Mouse	IgG ₁	Fast	No
Rat	IgG _{2a}	Slow	Yes
Sheep	IgG ₂	Slow	No

The electrophoretic mobility indicated in Table 1 is the relative mobility within the various IgG subclasses in each species. This property is often used to characterize these antibodies.

Production

Unlike IgE, which is a minor component of normal serum (around 0.2 µg/ml), the IgG subclasses to which the anaphylactic antibodies belong are present in relatively high concentrations (of the order of milligrams per milliliter) and may increase to extremely high values during the course of immunizations. Thus, in normal guinea pigs the serum concentration of IgG₁ is less than 1 mg/ml but increases to values of 18 mg/ml or more after immunization with protein antigens and complete Freund's adjuvant.¹⁰ In mice, the synthesis of IgG₁ is equally high¹¹ and values of more than 50 mg/ml have been reported in animals parasitized by *Mesocostoides corti*.¹² Important increases in sheep IgG₂¹³ and rat IgG_{2a}¹⁴ have similarly been observed during immunization. The antibody specific for the antigen used to immunize is rarely more than 5 or 10% of this tremendous increase. The rest is antibody directed against unknown determinants resulting from the polyclonal activation induced by the antigenic stimuli. This polyclonal activation is not exclusively of the IgG anaphylactic class; it is observed, at a varying degree, in other immunoglobulins. It is interesting that the increase in IgE in many cases of allergic or parasitic diseases is even higher.¹⁵ For instance, in allergic patients, values of 30 µg/ml, corresponding to 150 times the normal average value of 0.2 µg/ml have been found,^{16,17} and in rats parasitized with helminths, the concentration of IgE may increase from 0.2 to 247 µg/ml, that is, 1230 times. A relative increase of this degree is never observed in the synthesis of other immunoglobulins.¹⁸

Properties

Structure

All IgG anaphylactic antibodies have the same basic structure of 2 H and 2 L chains. The molecular weight is about 160,000 and the molecule exists in the serum as a monomer. The H chains are comprised of four homologous regions and are linked by disulfide bonds whose number varies in various species. There are important differences in the primary amino acid sequence of anaphylactic IgG from different species, as evidenced by their electropho-

retic and chromatographic behavior. Regarding their antigenic determinants, specific antibodies prepared in mammals may show cross-reactions between subclasses of IgG of different species,¹⁹ but there are no unique determinants indicating the anaphylactic capability of the immunoglobulin, that is, the structures responsible for the anaphylactic activity cannot be characterized antigenically. This is not very surprising if one considers that these structures are probably present in all mammals and are not immunogenic for them.

Some carbohydrate exists in the molecule, but the proportion is low, 1% or 2% in weight, as in other IgG subclasses.²⁰

Thermostability

An important difference between antibodies of IgG and IgE classes is their reaction to heat denaturation. Heating at 56°C destroys the activity of the IgE while it has no effect on IgG, a property frequently used as the main criterion for differentiation of the antibodies. The susceptibility of IgE differs among various species: heating for 5 hr is required to inactivate mouse or human IgE, whereas 1 hr is sufficient for rat IgE. Heating affects the Fc region of the molecule and denatures the structures involved in fixation to cellular receptors as well as the antigenic determinants. The capacity to bind to the specific antigen (Fab region) is not modified.²¹

Recent observations indicate that the heat thermolability of the IgE molecule is probably due to some kind of protein-protein interaction of unknown nature. Thermolability can be greatly diminished by simple dilution of the antiserum or by increasing the ionic concentration.^{22,23}

Transmission from Mother to Young

The transmission of immunoglobulins from mother to young plays an important role in neonatal survival because the newborn animal has little capacity to produce antibodies. Transmission may occur via the placenta, yolk sac or amnion, colostrum and milk.²⁴

In some species (man, monkey, rabbit, guinea pig), transmission readily takes place before birth and no antibodies are absorbed through the gut of the suckling baby even if they are present in the colostrum and milk. The opposite situation is found in other species (horse, pig, ox, sheep, goat), where fetal transmission is practically nil and the babies receive the maternal antibodies from colostrum. In a third group of animals (mouse, rat, dog, cat) both prenatal and postnatal passage take place. The information concerning the various types of transmission of each IgG subclass is far from complete. In general, IgE is never transferred, whereas IgG, including anaphylactic subclasses, may pass to the newborn circulation.

Fixation to Cellular Receptors

The outstanding property of anaphylactic immunoglobulins is their ability to induce the release of mediators from mast cells and basophils; this release is mediated by interactions between antibody and cell membrane structures.

In recent years the existence, nature, and role of mast cells receptors for the Fc region of IgE have been studied in great detail, mainly employing in vitro cultured rat basophilic leukemia cells.^{25,26} It has been found that the receptor molecule, the molecular weight of which is about 50,000, has a high affinity for the Fc (association constant 10^9 – 10^{10} liters/mol). The receptors are in close proximity on the cell membrane and it has been calculated that their number is 10^5 – 10^6 per cell. Under normal conditions, 70% of the receptors are occupied by IgE, but this proportion can be much higher in individuals with an elevated IgE serum concentration. Upon contact with the specific plurivalent antigen, aggregation of IgE antibodies takes place, provoking a rearrangement of the receptors on the cell membrane. This rearrangement, in an unknown manner, results in changes in the cell permeability and release of the mediators of anaphylaxis. What triggers the release of the mediators is not the antibody itself but the changes induced in the receptor after aggregation of the antibody. In fact, degranulation can be obtained in the absence of IgE, by treating the cell with antireceptor antibodies that provoke the same steric modifications. This is an important point that shows that the molecule of antibody acts mainly as a transmitter, alerting the cell that antigen is present.²⁷

In the case of IgG anaphylactic antibodies, fixation of the monomer to the mast cell or basophil has not been demonstrated, and it appears that in order to induce anaphylactic phenomena, aggregation by the antigen is previously needed. The fact that aggregated, but not monomeric, IgG acts on the cell can be explained in two ways: (a) following combination with the antigen, the Fc region is modified (transconformed) and new structures, specific for cell receptors, are formed; (b) the fixing structures already exist in the monomeric molecule but the affinity for the receptor is very low and no stable fixation occurs until aggregation by the antigen increases the avidity to an effective value (multiple site interactions). It has been asked whether the postulated IgG receptors are the same as those for IgE. One method of testing this is to try sensitization with IgE in the presence of IgG, or vice versa. If the receptors can bind both immunoglobulins, competition effects should be observed. The experimental evidence indicates that this is not the case when monomeric IgG is used. Using immune complexes formed by IgG_{2a} rat anaphylactic antibodies, it has been found that the interaction of IgE with rat basophilic leukemia cells could be partially inhibited.²⁸ Opposite results have been obtained with mouse IgG₁ and mouse peritoneal mast cells.²⁹ The reasons for this discrepancy are unclear and it cannot be stated conclusively whether the receptor for IgE is the same as that which receives the signal given by aggregated anaphylactic IgG.

Using isolated cell systems, mainly passive in vitro sensitization of peritoneal mast cells,³⁰ it has been shown that one may immediately obtain sensitization after addition of IgG anaphylactic antibodies to the cells, with no fixation time needed, as is the case with IgE. Actually, degranulation of the mast cells can be effected either by adding first the antigen and then the antibodies or by adding a mixture of antigen and antibody. Moreover, mast cells that have been sensitized by addition of IgG can be easily desensitized by simple washing. All these results indicate that no fixation step is required and the activation of the cell is mediated by a complex of antibody and antigen

performed in the fluid phase. The nature of this complex has been investigated in some cases and it appears that there exists an optimal ratio and that complexes formed in antigen excess are not active. On the contrary, when the sensitization is performed with IgE, the reaction is not inhibited by excess antigen.

Biologic Activity

Characterization and quantitative evaluation of the various IgG subclasses can be effected by a number of immunochemical methods, using specific antibodies. Nevertheless, the demonstration of their anaphylactic activity is only possible by means of a biologic test. There exist various ways to perform such a test, all of which are based on the same phenomenon of activation of mast cells and basophils. Activity is evaluated by measuring the intensity of the anaphylactic reaction; this can be done by direct observation of the induced symptoms or indirectly by measurement of the mediators released during the reaction—histamine, 5-HT, slow reacting substance, and the like.

The most frequently employed *in vivo* test is the passive cutaneous anaphylaxis (PCA) originally described by Chase³¹ and developed by Ovary,³² which is the equivalent, in animals, of the Prausnitz–Küstner reaction in humans. General or local anaphylaxis is also employed in some cases. In recent years, *in vitro* tests using isolated cells from the peritoneal cavity or the peripheral blood have been increasingly used.

Sensitizations can be performed by active or passive immunization. Special attention must be given to the conditions of the test in order to differentiate between IgG and IgE. When passive sensitization is used, heating of the antiserum at 56°C is usually sufficient to inactivate the IgE. Other technical criteria are equally useful. In particular, when PCA is employed, the time of incubation (between injection of the antibody and challenge) is short for IgG antibodies (not more than 2–3 hr) and long for IgE (18–48 hr). Persistence of the sensitization of the skin site is also very different: IgE antibodies remain for many days while IgG diffuses very rapidly and has practically disappeared after 24 hr. These times, however, are very relative and depend on the concentration of antibody.

An important point for the quantification of the reaction is that there is no synergistic or inhibitory effect due to the simultaneous presence of IgE and IgG. The reaction obtained when both are present is the addition of the reactions that would be produced independently by each antibody.³³

Complement Fixation

The possibility that complement fixation participates or is essential for the anaphylactic reaction has been very much studied and discussed. Guinea pig and mouse IgG₁ do not fix complement by the classical pathway, but when they are in aggregated form they do fix it by the alternative pathway. Aggregated rat IgG_{2a} myeloma fixes complement by the classical pathway.

In general, it can be said that *in vitro* experiments do not show any consistent correlation between complement fixation and anaphylactic activity. The situation is not so clear *in vitro* because the role of the alternative pathway

of fixation is not clearly understood and because many of the physiologic consequences of the anaphylactic reaction can be mediated by mechanisms in which the participation of some complement components is possible.

Heterologous Sensitizations

Although anaphylactic reactions can be produced in all species with homologous antibodies, passive sensitization with heterologous antibody is not always possible. Guinea pig, for example, can be sensitized by antibodies from rabbit, man, or sheep, but not from horse or fowl. The basis for this species difference is not known. In addition, studies on heterologous sensitizations have shown that the anaphylactic IgG antibody is active only in the homologous species. When heterologous sensitization is possible, the antibody is an IgG subclass that is not anaphylactic for the donor. These facts are difficult to frame within any present hypothesis on sensitization but have necessitated redefinition of the anaphylactic antibody as the one that mediates anaphylactic phenomena within the species.

Heterologous sensitizations are really experimental artifacts without physiologic significance. The very fact that they are possible, however, demonstrates the existence of functional similarities between species.

It is interesting that most of the studies done on passive sensitization until about 15 years ago used heterologous antibodies, generally rabbit antibody in guinea pigs. The results obtained helped elucidate many of the aspects of the anaphylactic reaction, but the conclusions drawn concerning the nature of the antibodies involved were completely wrong.

Interference of Nonspecific Immunoglobulins

The process of sensitization is hindered by the simultaneous presence of immunoglobulins that are not specific for the antigen used. This phenomenon has been observed in all systems studied *in vitro* and *in vivo* and constitutes one of the more striking and constant features of sensitization. The interference is evident not only when sensitization is carried out with mixtures of antibody and nonspecific immunoglobulin³⁴ but also when tissues are loaded with immunoglobulin before or after sensitization either *in vivo* or *in vitro*.³⁵ Conversely, washing the immunoglobulin out of the tissues facilitates the sensitization process.³⁶

Quantitative studies using various models of passive sensitization have shown the following:

1. the interference is proportional to the concentration of immunoglobulin;
2. interference is manifested by a decreased rate of sensitization, but,
3. provided that the incubation time is long enough, sensitization still occurs even at high relative immunoglobulin concentrations;
4. interference is produced by homologous immunoglobulins of the same subclass as the anaphylactic antibody; and
5. interference can be produced by heterologous immunoglobulins belonging to the subclass of the heterologous sensitizing antibody.

The mechanism of interference is not really understood in the case of IgG immunoglobulins. For many years, the prevailing hypothesis on the mechanism of sensitization was that, in order to mediate anaphylactic reactions, the antibody had to be "fixed" to tissue receptors. In keeping with this hypothesis, interference was supposed to be a "competition," between antibody and a nonspecific immunoglobulin, for the receptor. This concept had to be changed after it was recognized that fixation was a property of IgE but not of the monomer IgG antibody, as discussed before. The "competition" hypothesis proved to be correct only in the case of IgE. In addition, quantitative aspects of the interference phenomena are in conflict with a simple competitive hypothesis. An alternative explanation assumes that interference is due to an interaction between the molecules of immunoglobulin, antibody or not. The "association" hypothesis³⁷ is supported by a number of experimental results but the direct evidence confirming the existence of the postulated interaction is still lacking.

Blocking Antibodies

The anaphylactic reactions are mediated by very small quantities of antibody. Fatal shock can be induced in the guinea pig after intravenous sensitization with 0.03 mg of antibody; in PCA, the quantity of IgG₁ that sensitizes a skin site is 0.02 µg and even less in the case of IgE. In active sensitization it is very difficult to evaluate the concentration of antibody mediating the reaction but there is no doubt that it is also very small, and that the same is true in human allergic patients. It is to be expected, then, that the corresponding quantities of antigen are also very small and that factors affecting the combination of antibody and antigen must be determinant for the development of the anaphylactic shock. One of these factors is the dose of antigen available for combination at the site where the reaction takes place. In general, individuals actively sensitized possess not only anaphylactic antibodies but also antibodies belonging to other classes and subclasses of IgG. If the challenging dose of antigen is small, it may happen that the antigen available to combine with the anaphylactic antibody is not sufficient to elicit the reaction. The term "blocking antibody" is used to designate all the nonanaphylactic antibodies that compete for the antigen and may have an inhibitory effect on the anaphylactic reaction. This competition for antigen is perhaps one of the most important factors controlling the triggering of allergic reactions in man and is one of the main mechanisms invoked to explain the desensitization obtained by repeated inoculations of small doses of antigen. These injections would preferentially induce the synthesis of nonanaphylactic antibodies that combine with the majority of the challenging antigen and neutralize it.

Competition for antigen is probably more critical for IgE than for IgG because, as previously discussed, the IgE concentration is much smaller than that of other immunoglobulins. Also, blocking antibodies are present in the circulation; this may favor the neutralization of the antigen before it reaches the target cells where the active IgE is fixed.

In order to be effective, blocking antibodies must be present at a high relative concentration and must have a high affinity for the antigen. These

conditions are usually fulfilled by the conventional IgG and IgA humoral and mucosal antibodies. In recent years it has become evident that other types of antibodies can also act as blocking antibodies; they are the so-called nonprecipitating or coprecipitating antibodies initially described by Heidelberger and Kendall in 1935.³⁸ These antibodies combine with the antigen but are unable to form insoluble complexes. The label coprecipitating is applied to those antibodies that combine with the antigen already complexed with normal precipitating antibodies and coprecipitate with them. Nonprecipitating antibodies are present in practically all antisera, constituting 5–10% of the total antibody population. Purification of nonprecipitating antibodies is performed by adding successive small doses of antigen to the antiserum. After each addition, the resulting precipitate is eliminated by centrifugation. The quantity of antigen used in each addition is only a small fraction (usually one-twentieth) of the quantity corresponding to the equivalence point. After a certain number of additions, no more precipitate is formed and the nonprecipitating antibodies remain in the supernatant, from which they can be recovered and isolated by immunoabsorption.

Nonprecipitating antibodies have been found in all animal species studied, namely rabbit, guinea pig, sheep, goat, rat, cow, horse, and man, and they are present in all IgG subclasses. Their physiochemical and antigenic properties are identical to those of the corresponding precipitating antibodies.

Studies done with purified anti-DNP nonprecipitating antibodies have demonstrated that the molecule has two antigen binding sites that have different affinity.³⁹ The association constant of one of the sites has a value comparable to that of the precipitating antibody present in the same antiserum, while the value for the other site is 50–100 times less. For this reason, the molecule behaves practically as univalent, which is why insoluble complexes cannot be formed with antigen. The structural basis for the different affinity of the two sites is not clear at present. Sites with different affinity have also been demonstrated in the IgM molecule.^{40,41}

An interesting point concerning nonprecipitating antibodies is that because of their inability to form large complexes, their biological properties are different from those of the precipitating antibodies. In general, they are either incapable of mediating most physiological functions, or their capacity for doing so is very diminished. Nonprecipitating antibodies do not fix complement by the classical or the alternative pathway, and they do not stimulate *in vitro* phagocytosis, although they are cytophilic for the macrophages and they have no opsonizing capacity *in vivo*. Regarding their ability to induce anaphylactic phenomena, nonprecipitating antibodies are much less active than precipitating antibodies in heterologous sensitizations, the only sensitizations so far studied in detail.⁴²

It is then clear that nonprecipitating antibodies can act as blocking antibodies not only for anaphylactic reactions but also for other effector functions. Evidence is accumulating to show that nonprecipitating antibodies are increased and probably play a role in some chronic and parasitic diseases. It is possible that by covering the surface of the pathogen, they protect it against cytotoxic effects so that blocking nonprecipitating antibodies are, in fact, "facilitating" antibodies. It is a matter of speculation whether the synthesis of

these nonprecipitating antibodies is enhanced by the invader and constitutes a mechanism of escape from immune defenses.

Physiologic Functions

The anaphylactic reaction, in the form currently studied in the laboratory, is a harmful reaction, frequently fatal. However, anaphylactic antibodies have been preserved in evolution and are present in all mammals, and probably in many other more primitive species. Consequently, one might ask what is the normal, useful, physiologic function of these antibodies. It can be hypothesized that the release of mediators constitutes a first line of defense in the sense that the local and short-lived reaction induced modifies the microcirculation in the tissues and facilitates the transport of substances and cells that contribute to the repair of the lesion or to neutralization of the foreign invader. In normal individuals the level of anaphylactic antibodies is kept low and the inflammatory reaction is correspondingly weak. Also, it should be kept in mind that the production of anaphylactic antibodies, especially of IgE class, is difficult to stimulate and that the various techniques and immunization schedules employed to induce them in the laboratory do not reproduce situations normally encountered in nature. It is tempting to conclude that the synthesis of anaphylactic antibodies is normally kept very low, just adequate to permit a moderate reaction convenient for defense. One exception to this general rule is the case of spontaneously allergic individuals. In humans with clinical allergies, the level of IgE is greatly increased; this increase is not only manifested for antibodies specific for the allergen but also for the total immunoglobulin class. In addition, allergic conditions are frequently familial and it has been also observed that patients who avoid contact with the causative allergen (by moving to a different location, for instance) become easily sensitized to another substance. These features strongly suggest that the allergic condition is related to a malfunction of the regulatory mechanisms of anaphylactic antibody synthesis or mediator release. Another situation in which production of IgE, specific and nonspecific, is greatly enhanced is that of parasitic helminth infections. Recent studies on the immunologic aspects of parasitosis have disclosed that anaphylactic antibodies play an important defensive role. It has been shown that IgE mediates activation of macrophages and induces killing of schistosomes of *Schistosoma mansoni*, and that anaphylactic IgG antibodies are able to stimulate cytotoxicity by other cellular types (see references in ref. 43). The presence of high concentrations of anaphylactic antibodies in parasite-infested individuals also helps maintain a permanent state of hypersensitivity and a particular population of effector cells; both conditions may be important in the control of the parasitic infection.⁴⁴

Another fact that deserves consideration is the existence of two classes of antibodies producing anaphylaxis. The reason for this duplication could be their different distribution. IgE has to be regarded as a cytophilic antibody and IgG as a circulating antibody. They may, in some way, complement each other in regard to the physiologic function which they perform.

References

1. White RG, Jenkins CG, Wilkinson PG: The production of skin sensitizing antibody in the guinea pig. *Int Arch Allergy* 22:156–165, 1963
2. Benacerraf B, Ovary Z, Bloch KJ, Franklin EC: Properties of guinea pig 7S antibodies. I. Electrophoretic separation of two types of guinea pig 7S antibodies. *J Exp Med* 117:937–949, 1963
3. Fahey JL, Wunderlich J, Mishell R: The immunoglobulins of mice. I. Four major classes of immunoglobulins: 7S γ 2, 7S γ 1, γ 1A (β 2A) and 18S γ 1M-globulins. *J Exp Med* 120:223–242, 1964
4. Nussenzweig RS, Merryman C, Benacerraf B: Electrophoretic separation and properties of mouse antihapten antibodies involved in passive cutaneous anaphylaxis and passive hemolysis. *J Exp Med* 12:315–328, 1964
5. Morse HC, Bloch KL, Austen KF: Biological properties of rat antibodies. II. Time course of appearance of antibodies involved in antigen-induced release of SRS-A. Association of this activity with rat IgG α . *J Immunol* 101:658–663, 1968
6. Bach MK, Bloch KL, Austen KF: IgE and IgG antibody-mediated release of histamine from rat peritoneal cells. I. Optimum conditions for in vitro preparation of target cells with antibody and challenge with antigen. *J Exp Med* 133:752–784, 1971
7. Lindquist K: A unique class of rabbit immunoglobulins eliciting passive cutaneous anaphylaxis in homologous skin. *Immunochemistry* 5:525–542, 1968
8. Esteves MB, Sant'Anna OA, dos Santos Annes VC, Binaghi RA: Characterization and properties of an anaphylactic 7S antibody in sheep. *J Immunol* 112:722–727, 1974
9. Bulletin of the World Health Organization. 56(5):815–817, 1978
10. Binaghi RA: Production of 7S immunoglobulins in immunized guinea pigs. *J Immunol* 97:159–164, 1966
11. Barth WF, McLaughlin CL, Fahey JL: The immunoglobulins of mice. VI. Response to immunization. *J Immunol* 95:781–790, 1965
12. Mitchel GF: Responses to infection with metazoan and protozoan parasites in mice. *Adv Immunol* 28:451–511, 1979
13. Margni RA, Castrelos OD, Paz CB: The sheep immune response. Variations in the anti-hapten and anti-carrier antibodies in the γ 1 and γ 2 immunoglobulin fractions. *Immunology* 24:781–789, 1973
14. Binaghi RA, Sarandon de Merlo E: Characterization of rat IgA and its non-identity with the anaphylactic antibody. *Int Arch Allergy* 30:589–596, 1966
15. Waldmann TA, Polmar SH, Balestra ST, Jost MC, Bruce RM, Terry WD: Immunoglobulin E in immunologic deficiency diseases. *J Immunol* 109:304, 1972
16. Johansson SGO, Medlin T, Vahlquist B: Immunoglobulin levels in Ethiopian preschool children with special reference to high concentrations of IgE. *Lancet* i:1118, 1968
17. Bennich H, Johansson SGO: Structure and function of human immunoglobulin E. *Adv Immunol* 13:1, 1971
18. Jarret E, Bazin H: Elevation of total serum IgE in rats following helminth parasite infection. *Nature* 251:613–614, 1974
19. Esteves MB, Binaghi RA: Antigenic similarities among mammalian immunoglobulins. *Immunology* 23:137–145, 1972
20. Oettgen HF, Binaghi RA, Benacerraf B: Hexose content of guinea pig γ 1 and γ 2 immunoglobulins. *Proc Soc Exp Biol Med* 118:336–342, 1965
21. Ishizaka K, Ishizaka T, Menzel AEO: Physicochemical properties of reaginic antibody. VI. Effect of heat on γ E-. *J Immunol*.
22. Prouvost-Danon A, Binaghi RA, Abadie A: Effect of heating at 56°C on mouse IgE antibodies. *Immunochemistry* 14:81–84, 1977
23. Prouvost-Danon A, Abadie A, and Da Ponte JV: Heat inactivation of rat and mouse IgE. *Mol Immunol* 17:247–253, 1980
24. Brambell FWR: The Transmission of Passive Immunity from Mother to Young. Amsterdam, North Holland 1970

25. Kulczycki A Jr, Iserksy C, Metzger H: The interaction of IgE with rat basophilic leukemia cells. I. Evidence for specific binding of IgE. *J Exp Med* 139:600–615, 1974
26. Kulczycki A Jr, Metzger H: The interaction of IgE with rat basophilic leukemia cells. II. Quantitative aspects of the binding reaction. *J Exp Med* 140:1676–1695, 1974
27. Ishizaka T, Ishizaka K: Triggering of histamine release from rat mast cells by divalent antibodies against IgE receptors. *J Immunol* 120:800–805, 1978
28. Halper J, Metzger H: The interaction of IgE with rat basophilic leukemia cells. VI. Inhibition by IgG₄ immune complexes. *Immunochemistry* 13:907–913, 1976
29. Daëron M, Prouvost-Danon A, Voisin GA: Mast cell membrane antigens and Fc receptors in anaphylaxis. II. Functionally distinct receptors for IgG and IgE on mouse mast cells. *Cell Immunol* 49:178–189, 1980
30. Vaz N, Prouvost-Danon A: Behaviour of mouse mast cells during anaphylaxis in vitro. *Progr Allergy* 13:111–173, 1969
31. Chase MW: Studies on the sensitization of animals with simple chemical compounds. X. Antibodies inducing immediate type skin reaction. *J Exp Med* 86:489–497, 1947
32. Ovary Z: Immediate reactions of the skin of experimental animals provoked by antigen-antibody interaction. *Progr Allergy* 5:459–508, 1958
33. Binaghi RA, Perrudet-Badoux A: Activité additive des anticorps IgE et IgG₄ dans la réaction anaphylactique (PCA) chez le rat. *Ann Immunol (Inst Pasteur)* 127C:49–56, 1976
34. Binaghi R, Liacopoulos P, Halpern BN, Liacopoulos-Briot M: Interference of non-specific gamma globulins with passive in vitro anaphylactic sensitization of isolated guinea pig intestine. *Immunology* 5:204–210, 1962
35. Liacopoulos-Briot M, Binaghi RA, Halpern BN: Inhibition de l' anaphylaxie chez de cobaye hypergammaglobulinémique. *CR Soc Biol Fr* 162:679–683, 1968
36. Liacopoulos, P, Liacopoulos-Briot M, Binaghi RA, Perramant F: Corrélation entre la "dé-sorption" des gammaglobulines et la sensibilisation anaphylactique. Etudes sur des organes isolés sensibilisés passivement in vitro et in vivo. *CR Soc Biol Fr* 155:220–225, 1961
37. Binaghi RA: The sensitization of tissues, and interference of non-specific gamma-globulin, in Austen KF, Becker EL (eds): *Biochemistry of the Acute Allergic Reactions*. Oxford, Blackwell, 1968, pp 53–73
38. Heidelberger M, Kendall FE: A quantitative theory of the precipitin reaction. III. The reaction between crystalline egg albumin and its homologous antibody. *J Exp Med* 62:697–720, 1935
39. Margni RA, Cordal ME, Leoni J, Hajos SE, Veira S, Manghi M, Bazurro M: Nonprecipitating antibodies isolated by immunoadsorption. *Immunochemistry* 14:299–303, 1977
40. Onoue K, Grossberg AL, Yagi Y, Pressman D: Immunoglobulin M antibodies with ten combining sites. *Science* 162:574–576, 1968
41. Oriol R, Binaghi RA, Coltorti E: Valence and association constant of rat macroglobulin antibody. *J Immunology* 106:932–937, 1971
42. Margni RA, Perdigon G, Abatangelo C, Gentile T, Binaghi RA: Immunobiological behaviour of rabbit precipitating and nonprecipitating (coprecipitating) antibodies. *Immunology* 41:681–686, 1980
43. Capron ARG, Capron M, Dessaint JP: ADCC as primary mechanisms of defense against metazoan parasites. *Progr Immunol* 4:782–793, 1980
44. Binaghi RA: Mechanism of the immune defense anti-*Trichinella spiralis*, in Israel L, Lagrange PH, Salomon JC (eds): *Cancer Immunology and Parasite Immunology*. Paris, INSERM, 1981, pp 461–63