

BRIEF COMMUNICATION

Serum immunoglobulin levels in *N. brasiliensis* infection

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SUMMARY

We have measured the levels of IgG1, IgG2a, IgG2c, IgA and IgM in the serum of normal rats, and at various times after infection or re-infection with *Nippostrongylus brasiliensis*, in order to compare these responses with previously measured IgE levels. The results we have to report are that while the levels of the various immunoglobulin classes or subclasses are elevated to a greater or lesser extent, the increments are modest by comparison with the great elevation of total IgE.

INTRODUCTION

In previous publications we have shown that total serum IgE levels are highly elevated following the infection of rats with the nematode *N. brasiliensis* (Jarrett & Bazin, 1974) and have presented time course studies of antibody and total IgE during infection (Jarrett, Haig & Bazin, 1976). Interesting features of IgE elevation in this infection are the magnitude of the response—levels usually rise more than a 100-fold—and the fact that most of the IgE produced is not specific for the parasite.

While a large increase in total serum IgE is a general feature of helminth infection, at least in those species in which it has been possible to measure total IgE, i.e. man (reviewed by Johansson, Bennich & Berg, 1972), dog (Halliwell, 1973) and the rat (reviewed by Jarrett, 1976a), increases of the same order of magnitude have not been reported for other immunoglobulin classes.

We have now examined the sera from one of our time course experiments in greater detail, to ascertain whether or not significant increases occurred in other immunoglobulin (sub)classes, namely IgG1, IgG2a, IgG2c, IgA and IgM. We show here that while the levels of these immunoglobulins increase during infection, the increments are modest by comparison with IgE and occur at different times.

MATERIALS AND METHODS

The animals were outbred female hooded Lister rats (Animal Suppliers Ltd, London), weighing 150–200 g. The parasitological techniques for the culture of *N. brasiliensis* and infection of rats are described by Jarrett *et al.* (1976) and Jennings, Mulligan & Urquhart (1963).

The immunoglobulin estimations reported here were done on serum samples which had been stored at -20°C from an experiment performed 2 years previously for the estimation of total and antibody IgE (see Fig. 2, Jarrett *et al.*, 1976). In this experiment, ten rats which had been immunized with egg albumin and *Bordetella pertussis* were infected with 4000 *N. brasiliensis* larvae 1 month later and were bled 8, 14, 25 and 31 days after infection. They were re-infected with 4000 larvae on the last of these days and further bled on days 6, 12 and 24 after re-infection. Additionally, estimations were performed on the fresh sera of ten unimmunized hooded Lister rats. The results of IgE estimations of the infected rats in the experiment above have been described (Jarrett *et al.*, 1976) as has the method of assay (Jarrett & Bazin, 1974).

Estimation of the levels of the other immunoglobulin classes was by the method of Mancini, Carbonara & Heremans (1965), using antisera monospecific to each Ig (sub)class and normal rat serum with known concentrations as standard. Monoclonal immunoglobulins for the production of antisera were prepared from the sera of LOU/Wsl rats carrying the appropriate immunocytomas. The methods of purification of the immunoglobulins and preparation of the antisera have been described by Bazin, Beckers & Querinjean (1974). The normal rat serum used as standard had been titrated in comparison with purified monoclonal immunoglobulins of each class.

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RESULTS AND DISCUSSION

The serum immunoglobulin levels of normal rats (Table 1) and of egg albumin-immunized rats before infection (Table 2) differed little, with the exception of IgG1, which was raised in the immunized animals.

TABLE 1. Immunoglobulin levels in normal hooded Lister rats

Ig (sub)class	Ig concentration (mg/ml)	
	Mean \pm s.e.*	Range
IgG1	2.80 \pm 0.31	1.31-4.38
IgG2a	10.39 \pm 1.72	3.07-20.73
IgG2c	2.23 \pm 0.45	1.24-5.92
IgA	0.15 \pm 0.06	0.08-0.23
IgM	0.85 \pm 0.10	0.58-1.48
IgE (μ g/ml)	0.81 \pm 0.19	0.11-1.90

* Mean of ten values.

TABLE 2. Immunoglobulin levels in *N. brasiliensis* infected hooded Lister rats

Days after:	Ig concentration (mg/ml)					
	IgG1	IgG2a	IgG2c	IgA	IgM	IgE (μ g/ml)†
Infection						
-1	4.47 \pm 0.84	11.49 \pm 1.32	3.27 \pm 0.38	0.12 \pm 0.01	0.92 \pm 0.04	1.28 \pm 0.20
8	3.51 \pm 0.41	9.11 \pm 1.62	3.69 \pm 0.70	0.12 \pm 0.02	0.79 \pm 0.11	2.03 \pm 0.26
14	7.54 \pm 1.41	7.88 \pm 1.32	4.29 \pm 0.88	0.10 \pm 0.01	0.76 \pm 0.08	227 \pm 20.04
18	11.95 \pm 1.67	10.92 \pm 0.83	5.82 \pm 0.78	0.14 \pm 0.01	1.00 \pm 0.05	170 \pm 20.16
25	9.11 \pm 1.85	13.72 \pm 1.93	4.31 \pm 0.60	0.33 \pm 0.11	0.90 \pm 0.06	71.60 \pm 10.02
31	8.70 \pm 3.02	13.55 \pm 3.12	4.26 \pm 0.85	0.34 \pm 0.09	0.96 \pm 0.17	30.60 \pm 3.41
Re-infection						
6	14.92 \pm 1.59	10.52 \pm 1.07	4.51 \pm 0.68	0.21 \pm 0.04	1.42 \pm 0.21	331 \pm 28.78
12	17.81 \pm 4.05	11.59 \pm 2.90	5.16 \pm 0.83	0.32 \pm 0.06	1.36 \pm 0.15	222 \pm 31.29
24	12.29 \pm 1.99	11.39 \pm 0.75	4.28 \pm 0.26	0.43 \pm 0.11	0.89 \pm 0.06	67.6 \pm 7.47

* All results mean \pm s.e.; mean of seven to ten values for all Igs except IgG1. For the latter, means of five values except on day 8 where the mean is of eight values.

† IgE in μ g/ml, not mg/ml.

On day 8 after *N. brasiliensis* infection the levels of IgG1, IgG2a, and to a lesser extent IgM, had actually fallen below pre-infection levels in most animals. A similar decrease in immunoglobulin levels (IgG1, IgG2 and IgA) during the first week of *Trichinella spiralis* infection of mice has been recorded by Crandall & Crandall (1972).

Later in the infection the levels of the various immunoglobulins rose to some extent. The largest increase, apart from IgE, occurred in the level of IgG1: the mean level rose to a maximum 18 days after the first infection and 12 days after re-infection, increasing by a factor of 2.7 and 4, respectively, over the pre-infection level. The other notable feature of this response was a considerable variation between individual values: the range was 6.78-32.00 mg/ml on day 12 after re-infection, while it was 2.75-7.68 mg/ml before infection.

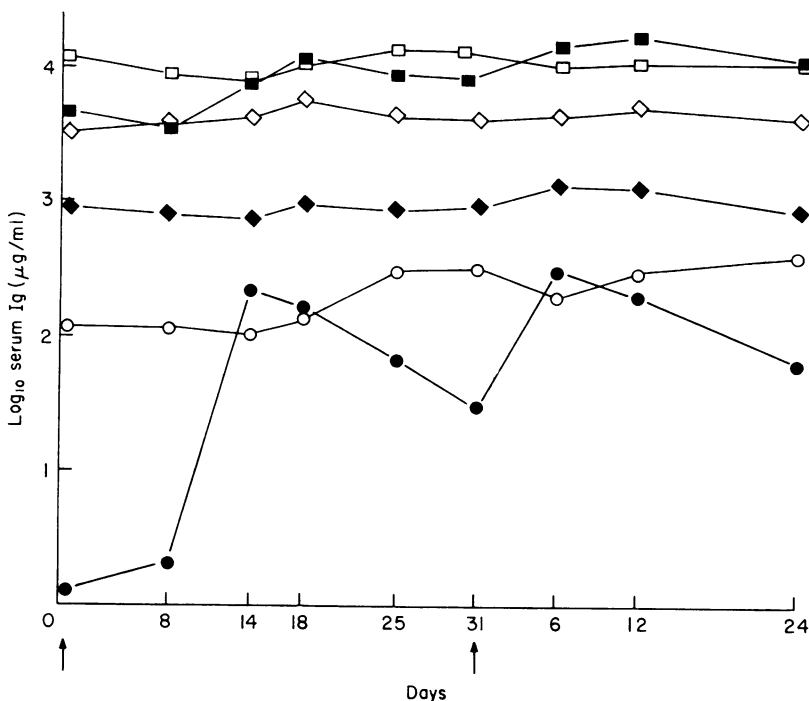


FIG. 1. Immunoglobulin levels in *N. brasiliensis* infection to show relative increments. See Table 2 for details. The first arrow shows the infection point, the second the re-infection point. (■)IgG1; (□)IgG2a; (◇)IgG2c; (◆)IgM; (○)IgA; (●)IgE.

IgG2a and IgG2c levels rose by a smaller factor. Two of the ten animals were deficient in IgG2c, and in one of these rats the level was below the sensitivity of the assay (i.e. < 126 µg/ml) on all the days: in the other rat the highest level reached (on day 18 after infection) was 0.6 mg/ml. All the other immunoglobulin values of these rats were quite normal.

It can be seen from Fig. 1 that the increments of total serum IgE in *N. brasiliensis* infection are greater by far than those of the other immunoglobulin classes. In this feature, as in the potentiation of unrelated antibody responses (Jarrett, 1976b), the stimulus of helminth infection is selective for IgE.

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REFERENCES

- BAZIN, H., BECKERS, A. & QUERINJEAN, P. (1974) Three classes and four (sub)classes of rat immunoglobulins: IgM, IgA, IgE and IgG1, IgG2a, IgG2c. *Eur. J. Immunol.* 4, 44.
- CRANDALL, R.B. & CRANDALL, C.A. (1972) *Trichinella spiralis*: immunologic responses to infection in mice. *Exp. Parasit.* 31, 378.
- HALLIWELL, R.E.W. (1973) The localisation of IgE in canine skin an immunofluorescent study. *J. Immunol.* 110, 422.
- JARRETT, E.E.E. (1976a). Production of IgE and reaginic antibody in rats in relation to worm infections. *Molecular and Biological Aspects of the Acute Allergic Reaction* (ed. S. G. O. Johansson, K. Strandberg and B. Uvnas), p. 105. Plenum, New York.
- JARRETT, E.E.E. (1976b) IgE production in infection with helminth parasites. *Current Titles in Immunology Transplantation and Allergy*, 4, 529.
- JARRETT, E.E.E. & BAZIN, H. (1974) Elevation of total serum IgE in rats following helminth parasite infection. *Nature (Lond.)*, 251, 613.
- JARRETT, E.E.E., HAIG, D.M. & BAZIN, H. (1976) Time course studies on rat IgE production in *N. brasiliensis* infection. *Clin. exp. Immunol.* 24, 346.
- JENNINGS, F.W., MULLIGAN, W. & URQUHART, G.M. (1963) Variables in X-ray inactivation of *N. brasiliensis* larvae. *Exp. Parasit.* 13, 367.
- JOHANSSON, S.G.O., BENNICH, H. & BERG, T. (1972) The clinical significance of IgE. *Progress in Clinical Immunology 1*, (ed. R.S. Schwartz), p. 157. R. S. Grune and Stratton Inc, New York.
- MANCINI, G., CARBONARA, A.O. & HEREMANS, J.F. (1965) Immunochemical quantitation of antigens by single radial immunodiffusion. *Immunochemistry*, 2, 235.