Reviewer’s report

Title: Vaccination with Hemagglutinin or Neuraminidase DNA Protects BALB/c Mice against Influenza Virus Infection in Presence of Maternal Antibody

Version: 1 Date: 1 March 2007
Reviewer: Zichria Zakay-Rones

Reviewer’s report:

General

The significance of maternal antibodies in the protection of the offspring is well known; on the other hand, the presence of maternal antibodies can also interfere with efficient vaccination. One way to overcome this inhibitory effect is by increasing the dose of the antigen, which is not always recommended or feasible. Delaying vaccination until maternal antibodies have waned (at about 3-6 months) may leave a gap of time in which the offspring is not protected. It is therefore of practical importance, as well as interesting, to find a way to vaccinate newborns efficiently, in the presence of maternal antibodies. This specific issue is addressed very clearly in the present work, with the conclusion that a different type of influenza vaccine should be administered to the mother and the offspring in order to obtain protection in the offspring.

The title: Perhaps the concept of how to vaccinate in the presence of maternal antibodies would be better reflected in a title: Vaccination of mice in the presence of maternal antibodies with a heterologous vaccine protects against influenza infection, (optional)

The abstract describes precisely the main findings of the work.

The methods are appropriate for answering the specific aims of the research, and are described clearly. Sufficient details are provided enabling replication of the work.

The experiments are well designed and the results in the text are presented in an informative, understandable and logical manner. The results offer a solution to the question asked in the research - how to overcome the intervention of maternal antibodies in the establishment of immunity following vaccination.

I would like, however, to raise a few points that should be clarified:
1. How many mice were in a tested group? Were the experiments carried out more than once?
2. In tables 1-3 it should be indicated that the antibody results are from the offspring. (I assume it is so, as there are titers of antibodies in the groups of non immunized mothers), nevertheless it should be written in the legends. Anti HA should be added to ELISA in the header as this is what is measured by the Elisa method (as NI is indicated in the next header).
3. The authors describe many schedules of vaccination, yet, in my opinion, one schedule is missing: vaccinating the mothers with DNA vaccines and then the offspring with an inactivated vaccine.
4. It can be assumed that a large part of the results is dependent on the level of the maternal antibodies that the offspring possess, which is dependent on the level in the mother (sometimes it is even more concentrated). This point is mentioned very vaguely and only once in the discussion (p.13 last 2 lines and first 2 lines p. 14).
This test could be done easily by checking the level of antibodies in the offspring before vaccination, or alternatively, checking the level of antibodies in the mothers after vaccination. In the case that sera samples are not available, this point should be at least further discussed.
5. The author should account for the rather poor correlation between survival, viral load and antibody titers. Viral loads in the lungs of offspring from immunized and non immunized mice are the same, so how do the authors account for the difference in survival (table 1); similar data are depicted also in the other tables. Also, the level of antibodies does not correlate with the protection, which may be because antibodies to HA have been measured by ELISA rather than by the HI assay which correlate to neutralizing antibody.
6. An observation that was neglected, perhaps because it is not directly within the frame of the research, is the protection provided by neuraminidase that correlates to viral load and is dependent on the level of NI antibodies. It is worthy of mention in the discussion as it corroborates claims that neuraminidase should be included in the vaccine.

In conclusion: The manuscript is dealing with an important and significant issue in the field of vaccination. The manuscript is written in a clear, acceptable way and the conclusions are based on the data obtained in the experimental work. The discussion relies on a large body of data in the literature, but lacks consideration of some of the data collected by the authors (see remarks above). Some of the corrections are minor, while others call for more experimental work, if possible. If further experiments are not feasible,
then the researchers should at least consider the issues in the discussion so that the work can be published.

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Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)

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Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

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Discretionary Revisions (which the author can choose to ignore)