



**AGRO-BIO**  
The Proteomic &  
Immune Response

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# Protocol for the production of chicken polyclonal antibodies Total IgY

Version 2010/01

EN

Agro-Bio has developed a time-optimized immunization protocol, yielding quality antibodies, needing low amounts of antigen. We recommend the use of 2 animals minimum to follow the protocol of immunization in order to overcome the differences obtained due to the genetic variability between each animal. The chicken are excellent for producing antibodies, their immune response is comparable to mammal response and is fast (< 30 days).

"The amount of antibody produced in one month is 18 times greater than rabbit in the same period" (Gassman et al. 1990).

IgY present advantages that may be decisive in the choice of immunization protocol:

- IgY seem similar to natural IgG fragmented F(ab')<sub>2</sub>,
- IgY have no affinity for mammal Fc receptor,
- IgY do not activate mammal complement proteins.

TOTAL IgY	
<b>Species:</b>	EOPS Chicken (Exempt from specific pathogenic organisms) (white Leghorn (PA12) or golden Leghorn (LD1 and LD2)).
<b>Antigens:</b>	Peptides, proteins, bacteria, virus or any other inactivated or nontoxic organism whose antigenicity and immunogenicity have been demonstrated. Concentration advised > 1 mg / mL.
<b>Antigen quantities:</b>	To obtain good titers of antibodies, AGRO-BIO uses for a protocol 35 days: <ul style="list-style-type: none"> <li>- 750 µg by chicken,</li> <li>- 3.10<sup>8</sup> PFU or CFU by chicken.</li> </ul>
<b>Injections:</b>	The injections are carried out by intramuscular route, in FCA for the first one whereas the other injections are made in incomplete Freund's adjuvant.



## Immunization protocol

Day	0	10	20	35
<b>Injection</b>	1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	
<b>Volume of serum</b>	Pre-immune <sup>(1)</sup>			eggs <sup>(2)</sup>

<sup>(1)</sup> Pre-immune serum enables to determine whether the animals have naturally occurring antibodies that can recognize non-specifically the antigen of interest.

<sup>(2)</sup> The eggs layed after the last injection are collected to extract the corresponding hyper immune IgY. The 35<sup>th</sup> day (=D35) IgY are extracted by precipitation.

Within two weeks following the dispatch of your order, you will receive a provisional timetable listing the various stages of your service.

### Included in our protocol:

- The purchase of animals, the quarantine period and transfer to experimentation,
- The guard of animals during the protocol,
- The injections and IgY extractions,
- The 10-day guard while awaiting your decision on the fate of animals,
- The sacrifice of animals without recovery of serum.

### Not included in our protocol:

- The charges,
- The guard of animals for a period exceeding 10 days (you have 10 days starting from last collection reception date to inform us of your decision to continue or not your protocol. After this deadline and in the absence of response from you, we are forced to sacrifice your animals).

Cf. General terms of sale.

Agro-Bio is approved by the prefecture of Loir-et-Cher for experimentation on vertebrate animals.  
Accreditation number: B 41-285-4

