Use of alternative adjuvant systems to produce neutralizing IgY antibodies against *Bothrops alternatus* venom

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Introduction

The use of adjuvant is a key point in the production of antivenoms and currently immunization schemes mostly consider Freund adjuvant. Anyhow, there are other adjuvants that may have a better performance, particularly when the antivenoms are produced by using other platforms different from horses, such as laying hens and egg yolk antibodies (IgY).

The aim of this study was to evaluate the performance of a commercial Montanide™ adjuvant to produce an IgY-based antivenom against *Bothrops alternatus*.

Methodology

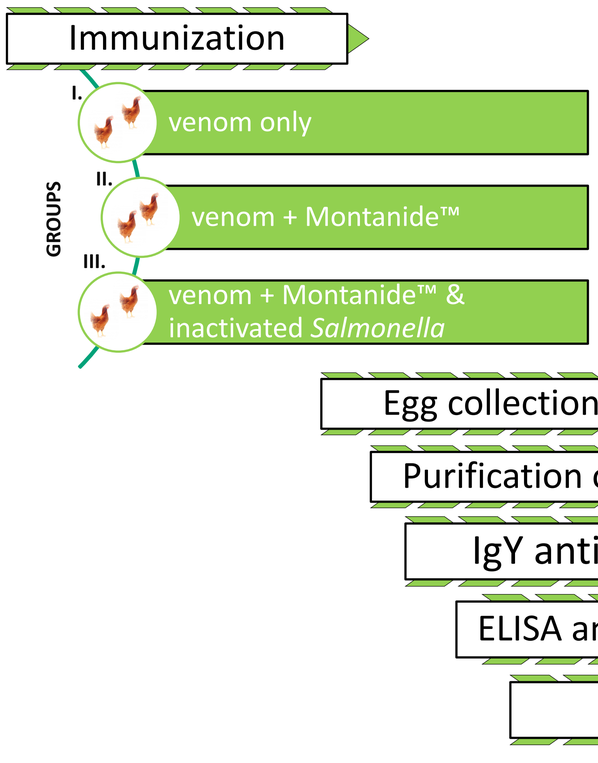

Figure 1. Overview of the methodology. Experiments were approved by the IACUC from the CICVyA-INTA (Procedure Nr. 20/2012). Median effective dose (ED_{50}) assay was performed according to WHO guidelines (2017).

Results

IgY immunoreactivity

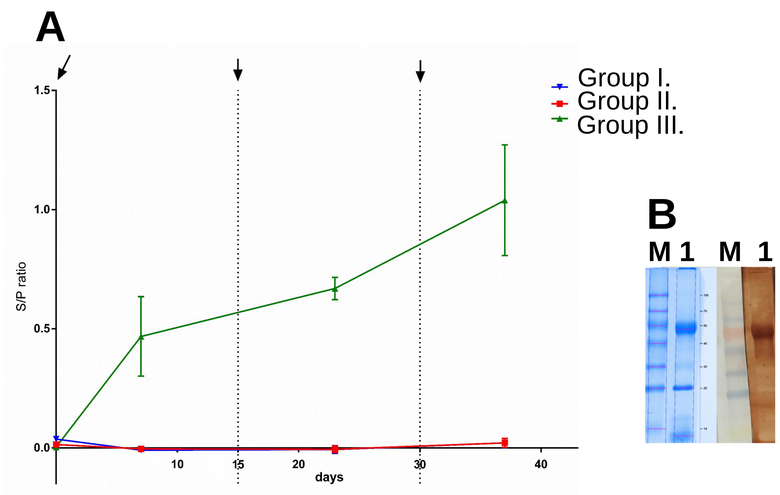

Figure 2. A: Relative level of IgY antibodies measured by ELISA. B: SDS-PAGE and Western-Blot of the bothropic venom; Lane M: marker, Lane 1: venom.

In vivo neutralization

	LD_{50} [µg/mice]	Challenge Dose	ED_{50} [µg/mL]
Chicken 1	28.28	3 x LD_{50} = 84.80 µg	500
Chicken 2			< 200

Table 1. Neutralization of lethality of IgY antivenom obtained from eggs of group III (venom + Montanide™ & inactivated *Salmonella*).

Conclusions

Montanide™ commercial adjuvant could be used to produce IgY based-antivenoms against *B. alternatus* but an immune-stimulant component such as inactivated Salmonella in the first injection is needed to elicit the response.

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