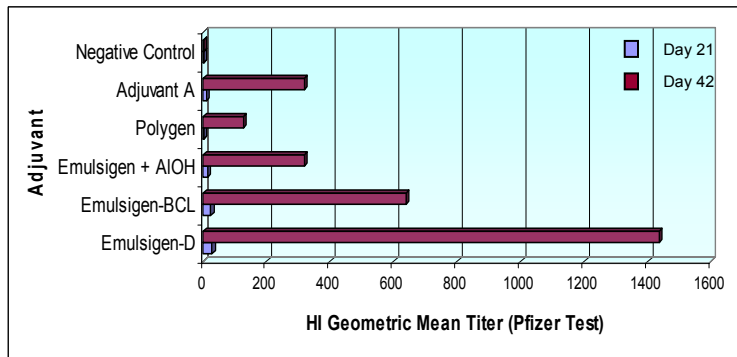
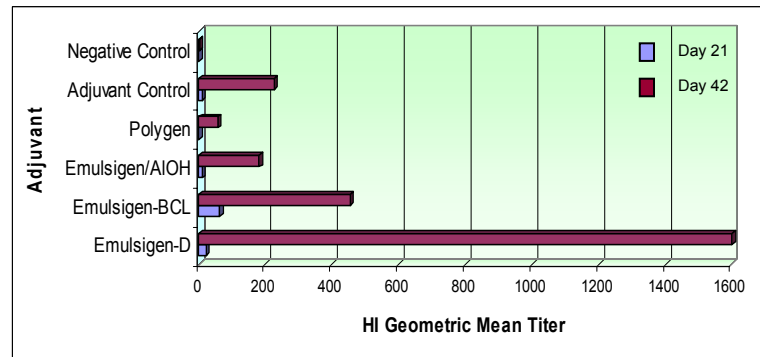


# COMPARISON OF ADJUVANTS FOR STIMULATION OF HI ANTIBODY TO SIV

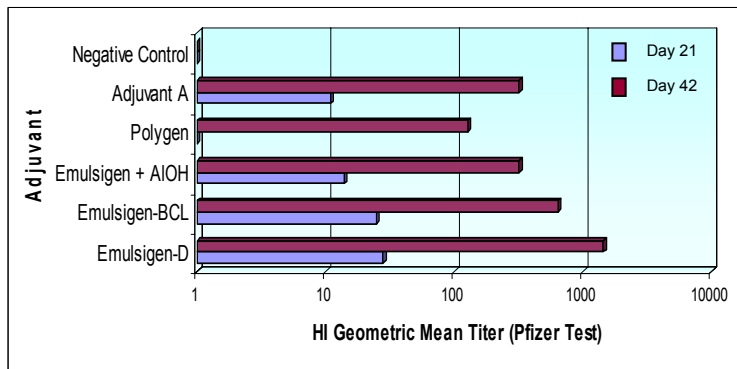
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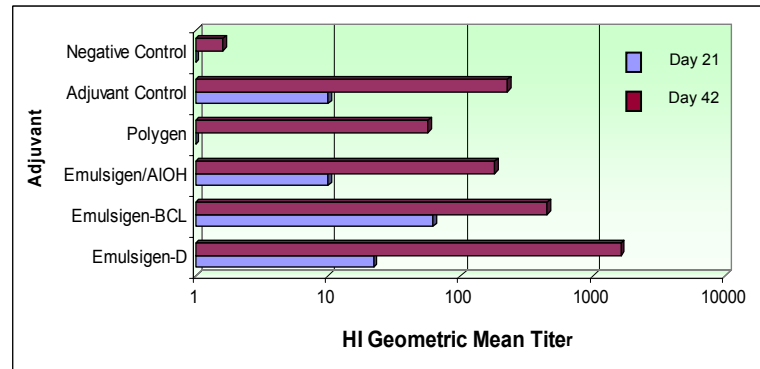
**Figure 1a** Adjuvant stimulation of HI antibody to H1N1 - Arithmetic Scale



**Figure 2a** Adjuvant stimulation of HI antibody to H3N2 Arithmetic Scale



**Figure 1b** Adjuvant stimulation of HI antibody to H1N1 -- Logarithmic Scale



**Figure 2b** Adjuvant stimulation of HI antibody to H3N2 --Logarithmic Scale

## Introduction

Swine influenza virus (SIV) causes an acute, endemic infectious disease of pigs. Bivalent vaccines (H1N1 and H3N2) are currently used for controlling this disease. However, it has been reported that inactivated SIV vaccines do not consistently provide complete protection to virus challenges in vaccinated pigs<sup>1</sup>. One way to improve protection provided by SIV vaccines is to use more potent adjuvants that stimulate higher immune responses. **This study was conducted to determine the best adjuvant for use with SIV vaccines.**

## Materials and Methods

A commercial vaccine containing inactivated freeze-dried H1N1 and H3N2 SIV antigens was used to prepare five test vaccines by adding the recommended amount of either the manufacturer's adjuvant (Adjuvant Control) or one of four other adjuvants as part of the diluent. Test vaccines thus contained a constant amount of antigen making any variability in immune response directly related to the effect of the adjuvant. Immune responses were evaluated using hemagglutination inhibition (HI) testing conducted by the Veterinary Diagnostic Laboratory at Iowa State University. All sera were coded so that the results were blinded. Thirty six seronegative three week old pigs, housed at the University of Nebraska, were randomly assigned to each of six groups. Groups 1 to 4 were given vaccines containing EMULSIGEN<sup>®</sup>-D, EMULSIGEN<sup>®</sup>-BCL, EMULSIGEN<sup>®</sup> with Al(OH)<sub>3</sub> (EMULSIGEN<sup>®</sup>/AIOH) and POLYGEN<sup>™</sup>, respectively (all supplied by MVP Laboratories, Inc., Omaha, NE, USA). Group 5 pigs received the Adjuvant Control (supplied by the manufacturer of the freeze-dried SIV antigens). Pigs in group 6 were given PBS only (negative controls). On Day 0, all of the vaccinated pigs were injected intramuscularly with a 2.0 ml dose. On Day 21, all pigs were given a second 2.0 ml dose. All pigs were bled on Days 0, 21, and 42.

## Results

Data are graphically presented on arithmetic and logarithmic (Log) scales (Figures 1a, 1b, 2a and 2b). The Log scale better illustrates that there is a HI response at 21 days post vaccination.

On Day 0, all 36 pigs were seronegative to both subtypes. Group 6 pigs (negative control group) remained seronegative throughout the study. The Day 42 H1N1 Geometric Mean titers (GMT) were 1,437, 640, 320, 127 and 320 for groups receiving EMULSIGEN<sup>®</sup>-D, EMULSIGEN<sup>®</sup>-BCL, EMULSIGEN<sup>®</sup>/AIOH, POLYGEN<sup>™</sup> and the Adjuvant Control, respectively (see Figure 1).

The Day 42 H3N2 GMTs were 1,613, 453, 180, 57 and 226 for the groups receiving EMULSIGEN<sup>®</sup>-D, EMULSIGEN<sup>®</sup>-BCL, EMULSIGEN<sup>®</sup>/AIOH, POLYGEN<sup>™</sup> and the Adjuvant Control, respectively (see Figure 2).

## Discussion

This study compared EMULSIGEN<sup>®</sup>-based adjuvants containing added immunostimulants, a copolymer adjuvant (POLYGEN<sup>™</sup>) and an Adjuvant Control as to their ability to stimulate HI titers in pigs vaccinated with SIV vaccines containing constant amounts of H1N1 and H3N2. All adjuvants stimulated protective HI titers to both H1N1 and H3N2 in all pigs by Day 42 post vaccination (protection indicated by HI titers  $\geq 1:40$ )<sup>2</sup>. Additionally, all adjuvants stimulated a significant increase in HI antibody responses when compared with the negative control group at the  $p < 0.05$  level. **Pigs vaccinated with experimental vaccines formulated with either EMULSIGEN<sup>®</sup>-D or EMULSIGEN<sup>®</sup>-BCL gave enhanced HI responses when compared to all other adjuvants in this study (see Figures 1 and 2).** As noted in Figures 1 and 2, EMULSIGEN<sup>®</sup>-D was the most effective adjuvant as it produced significantly higher HI titers against both H1N1 and H3N2 on Days 21 and 42 than the Adjuvant Control ( $p < 0.05$  for all values). EMULSIGEN<sup>®</sup>-BCL induced the second highest antibody response, as it produced Day 42 HI titers that were significantly higher than those produced by the Adjuvant Control ( $p < 0.05$ ). The results of this study suggest that EMULSIGEN<sup>®</sup>-D and EMULSIGEN<sup>®</sup>-BCL are the preferred adjuvants for immunoenhancement of SIV vaccines.

## References

1. Larsen, D.L. et al. (2000) Vet. Microbio. 74:117-131.
2. Janke, B.H. (2000) Swine Health and Production. 8:79-83.