

AIDSVax Inc.  
Department of Vaccine Research  
1979 Cure Drive  
Rochester, NY 14620

Dear Research Technician,

We are excited to collaborate with your organization in developing an AIDS vaccine. I am sending you 4 possible DNA vaccines that you could use in your vaccine trials. I'm not sure which combinations of the three HIV genes are in each one. You will have to check this.

Along with the HIV DNA, I have included some data collected from preliminary animal studies. We injected mice with each of the three HIV genes alone: the *gag* gene codes for HIV structural proteins; the *env* gene codes for the virus envelope proteins; and the *pol* gene codes for viral enzymes.

Sincerely,

A handwritten signature in cursive script that reads "Clarke Kent". The signature is written in dark ink and is positioned below the word "Sincerely,".

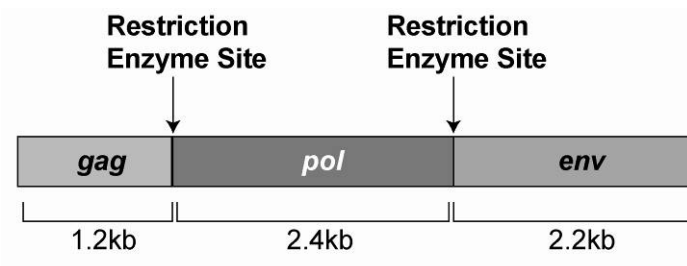
Clarke Kent, Ph.D  
Director  
Center for Vaccine Research  
University of Rochester

## Part 1: Gel Electrophoresis

You have been given 4 potential AIDS vaccines (V1, V2, V3, V4). These are DNA vaccines that contain different parts of the HIV genome, which is shown below.

The DNA vaccines have already been cut with a restriction enzyme. The restriction enzyme cut sites are shown below. The entire HIV genome is cut into three pieces (*gag*, *pol* and *env*).

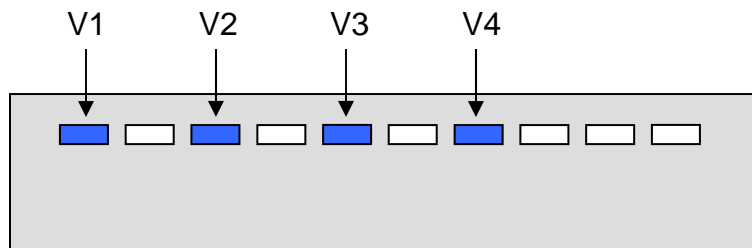
### HIV Genome:



You will need to identify which of the 4 DNA vaccines have parts of the HIV genome that you might be interested in using in an AIDS vaccine clinical trial. For example, you will not want to use a vaccine that contains all three genes (why not?)

You will use agarose gel electrophoresis to determine which parts of the HIV genome are in each of the 4 DNA vaccines.

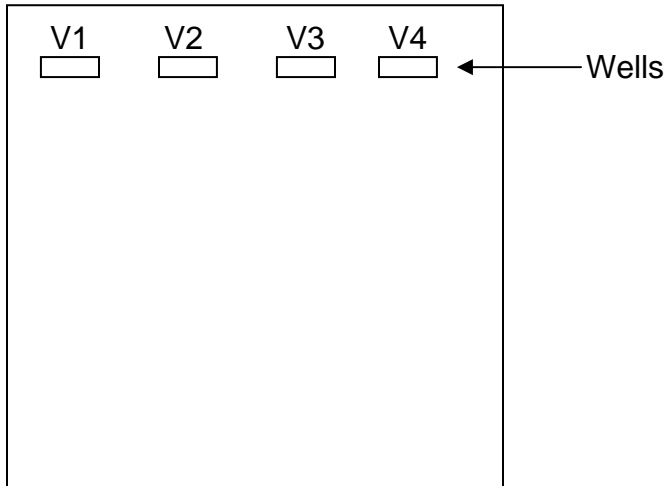
Load 10 $\mu$ L of each of the pre-cut DNA vaccines into the wells of an agarose gel and run the gel for about 15 minutes at 120 Volts



## Part 1: Observations/Conclusions

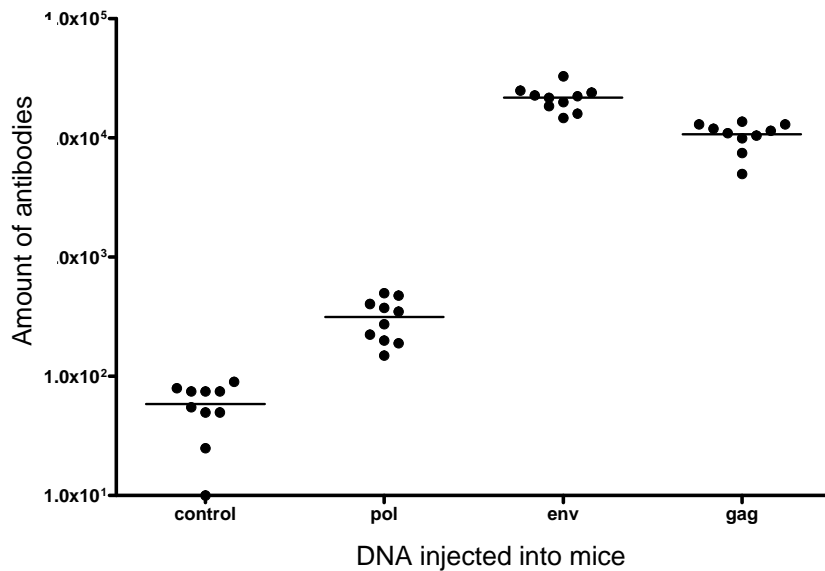
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Draw what you see on your gel:

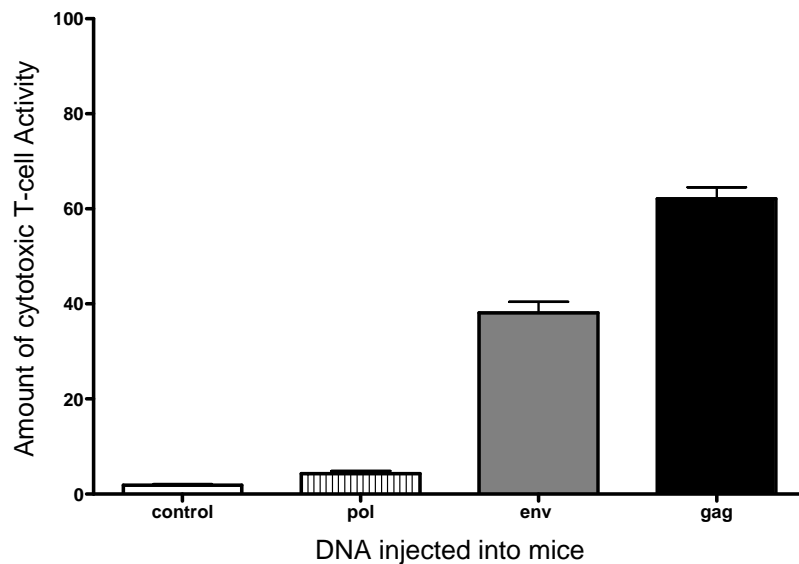


What does each band represent on the gel?

## Part 1: Studies in mouse models



**Figure 1.** Comparison of antibody titers collected from 10 mice injected with different DNA HIV vaccines encoding *pol*, *env*, *gag*, or control DNA. Serum from each animal was collected 8 weeks after injection. Each filled circle represents one mouse.



**Figure 2.** Cytotoxic T cell activity from 10 mice injected with different DNA HIV vaccines encoding *pol*, *env*, *gag*, or control DNA. Immune cells from each animal were collected 8 weeks after injection.

**Use the information from your gel and the animal studies to answer the following questions:**

Vaccine Name	What genes are in this vaccine?	Would you use this in a human trial?	If no, why not?
<b>V1</b>			
<b>V2</b>			
<b>V3</b>			
<b>V4</b>			

## Part 2: Testing for an Immune Response using ELISA (Enzyme Linked ImmunoSorbant Assay)

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Which vaccine results in the best immune response, V2 or V4?

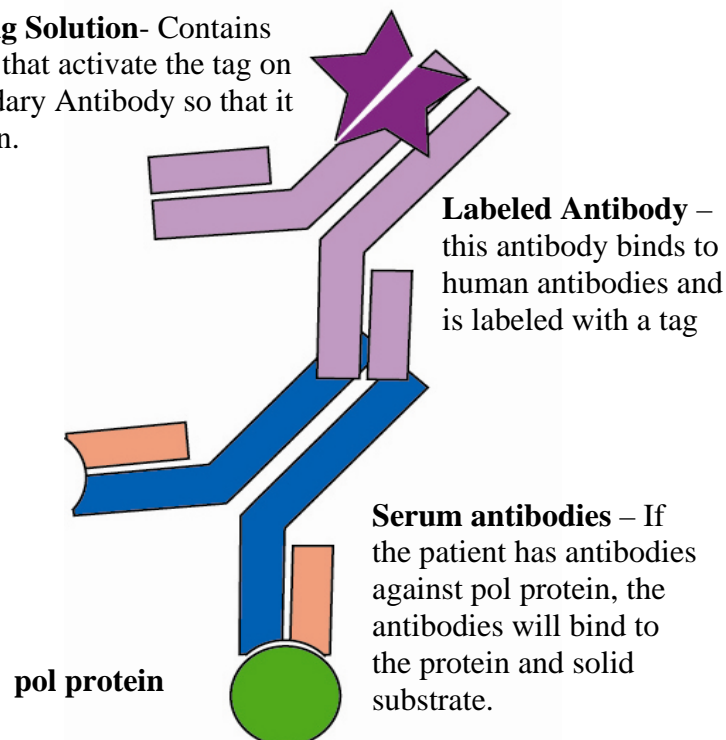
You have tested V2 and V4 vaccines by injecting volunteer patients with V2 or V4 vaccine, or with a placebo.

14 people were injected with V2  
14 people were injected with V4  
6 people were injected with placebo

Several months later, you collect serum from these patients. You will test their serum samples to see if these patients have made antibodies against the proteins that are coded for by the DNA vaccine. This is done using an Enzyme Linked ImmunoSorbant Assay, or ELISA.

### ELISA:

**Developing Solution-** Contains molecules that activate the tag on the Secondary Antibody so that it can be seen.



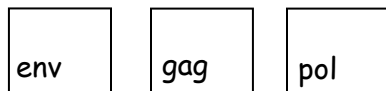
**Solid substrate** – Made of plastic or specially treated paper, this holds the protein in place.

## Part 2: Clinical Trial: Individual ELISA Data

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### PROCEDURE

- 1) Label three **absorbent squares** using a pencil. One for each HIV protein (As shown below)



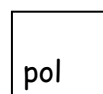
- 2) Place one labeled absorbent square in each of the three small plastic trays



- 3) Use a transfer pipette to:
  - a. drop 1 drop of **env** proteins onto the square labeled “env”
  - b. drop 1 drop of **gag** proteins onto the square labeled “gag”
  - c. drop 1 drop of **pol** proteins onto the square labeled “pol”
- 4) Let the squares dry for 1 minute
- 5) Use a transfer pipette to put 0.5mL of your **patient serum** onto each square
- 6) Let soak for 5 seconds
- 7) Pour off the excess serum
- 8) Transfer 0.5mL of **labeled antibodies** on the square
- 9) Let soak for 5 seconds
- 10) Transfer 0.5mL of **water** onto each square to quickly rinse the card
- 11) Pour off the water
- 12) Drop 1 drop of **Developing Solution** onto each square
- 13) Record your results

Patient # \_\_\_\_\_

Patient was given vaccine \_\_\_\_\_



## Part 2: Clinical Trial Group Data

Patient #	Vaccine given	pol antibodies?	env antibodies?	gag antibodies?
1	V2			
2	V2			
3	V2			
4	V2			
5	V2			
6	V2			
7	V2			
8	V2			
9	V2			
10	V2			
11	V2			
12	V2			
13	V2			
14	V2			
15	V4			
16	V4			
17	V4			
18	V4			
19	V4			
20	V4			
21	V4			
22	V4			
23	V4			
24	V4			
25	V4			
26	V4			
27	V4			
28	V4			
29	Placebo			
30	Placebo			
31	Placebo			
32	Placebo			
33	Placebo			
34	Placebo			

- What antibodies did you expect a person injected with V2 would have?
- What antibodies did you expect a person injected with V4 would have?
- What antibodies did you expect a person injected with Placebo would have?
- Which vaccine do you think works the best? Why?