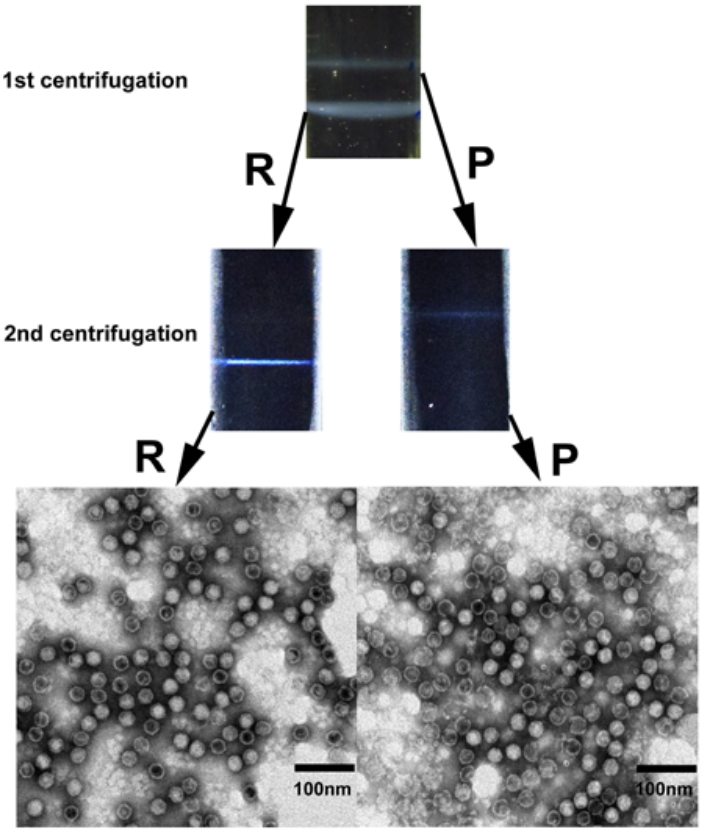
a




b

| Name | Contents (μg/ml) |
|---|---|
| P fractions | 232.45±5.89 |
| R fractions | 117.47±3.07 |

c

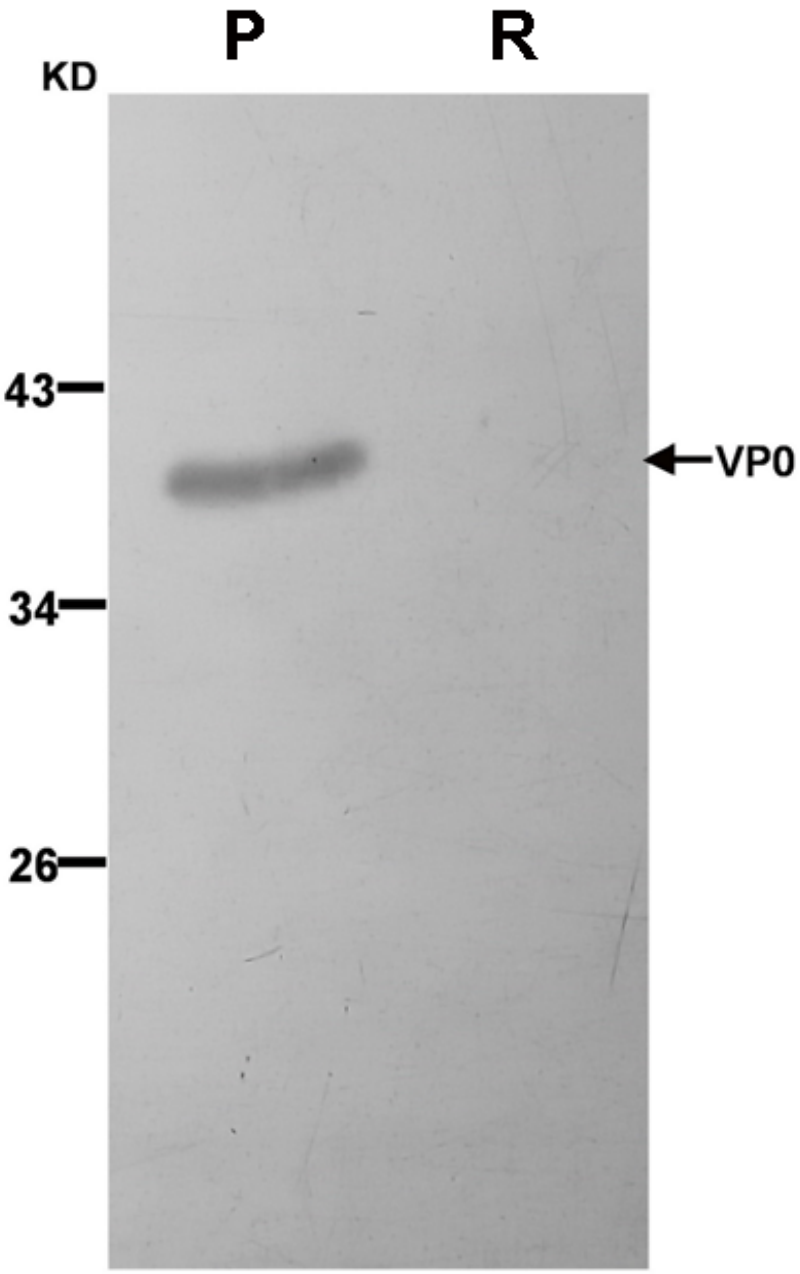
| Name | Antigenicity (U/ml) | Viral titer ($lgCCID_{50}$/ml) |
|---|---|---|
| P fractions | 16000±349 | 4.25±1.50 |
| R fractions | 8000±225 | 5.75±1.25 |

**Supplemental Fig. 1 Characterization of the two virion fractions.**

a: The virion fractions purified by iodixanol gradient centrifugation. The upper layer was located on ~fraction33, and the lower layer was on ~fraction38. The crude CVA16 concentrate was loaded onto a 10-60% continuous iodixanol gradient and centrifuged at 38,000 rpm for 5 hours using a SW41Ti rotor. The fractions were shown by a beam of light in a darkroom. After the first iodixanol centrifugation, the fractions of each visible band were collected, and each fraction was loaded onto a new 10-60% continuous iodixanol gradient for a second round under the same conditions. Each fraction is observed by electron microscopic observations (EM). The bar represents 100 nm.

b: The protein from the P and R fractions was determined using a BCA protein assay kit. The values (Mean± SD) are the means of three independent experiments.

c: The assay of antigenicity and viral titer. The antigenicity and viral titers of the two virions were detected setting the proteins of the P and R fractions at some level (56 μg/ml). The viral antigen titers were determined by an ELISA assay with anti-CA16 polyclonal antiserum. The viral titration was determined by a microscale assay. The values (Mean± SD) are the average of three independent experiments.




**Supplemental Fig. 2 Western blots of the two virion fractions.**

The CA16 VP0 protein of the P virion fraction (P) and the R virion fraction (R) were detected by western blot using rabbit anti-VP4 serum which was found to cross-react with VP0.