# Proposition; PCR does not tell everything !!

- PCR is a very sensitive diagnostic test; one single viral genome molecule may be detected in a sample.
- Cycles needed to generate a detectable PCR fragment (Ct value) is an inverse measure for the concentration (copies of genome segments present in the sample)
- Cut-of ; Ct >35 means negative ?? >> Not necessary; dependent on reagents and hardware (machine) used for the test (sequencing of PCR product can be used to confirm)
- Ct >35 cut-of is arbitrary; often used to exclude detection of contaminations with traces of viral genomes circulating in the lab and of false-positives (especially in high-throughput labs).
- European labs for notifiable viral diseases use ring-trials to synchronize PCR tests (e.g. CSFV, FMDV)
- A positive result in the PCR test only tells you that PEDV genome segments are present in the sample. (genome segments packed in a protein-capsid/envelop, as well as non-packed "free" genome segments).

  *Non-packed genomes are released from disrupted infected cells in affected animals and by breakdown of the intact virus capsid/envelop by proteases in the animal (blood and feces contains much protease activity).*

- A positive result in the PCR test does not tell you whether viable/infection-competent virus is present in the sample. A low Ct value (high conc.) does not always correlate with the presence of viable virus in the sample, and vice versa a sample with a high Ct (low conc.) can contain viable virus.

  *The virus surface can be damaged and fail to bind to a specific receptor on the surface of susceptible cells for cell-entry, or the virus surface is altered by mutations (virus variants).*

- What tells you if there is viable virus present in your sample ? > **infection experiments**

  *Virus isolation using cell-cultures (like Vero cells) and/or infection in-vivo (e.g. injection into, or challenge of susceptible animals*)