



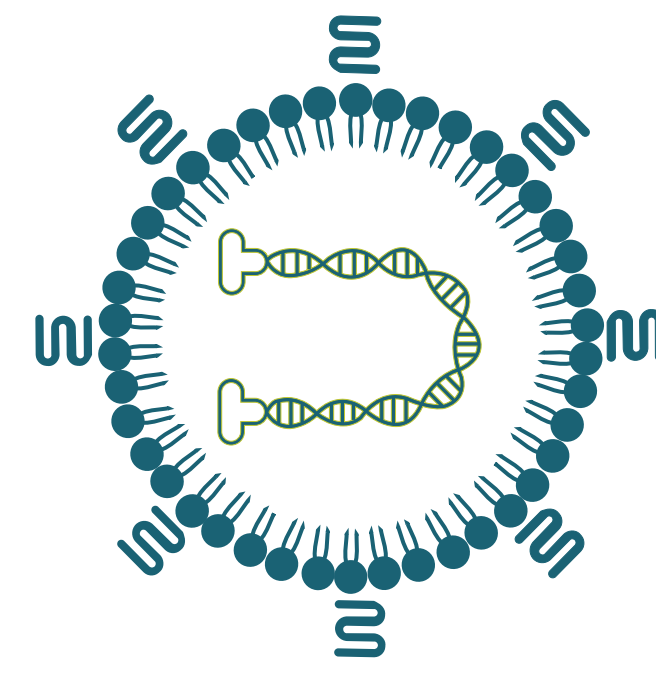
iqDNA is an engineered DNA cargo that avoids innate immune activation while retaining durable transgene expression

Russell Monds, Kelly Miller, Andrew O'Hagan, Nicholas Parsonnet, Joris Cipi, Nick Hanovice, Christian Slubowski, Luis Agosto, Anastasia Lymar, Phillip Samayoa, & Matthew Stanton
Generation Bio, Cambridge, MA 02142, U.S.A.

Generation Bio non-viral DNA therapy platform

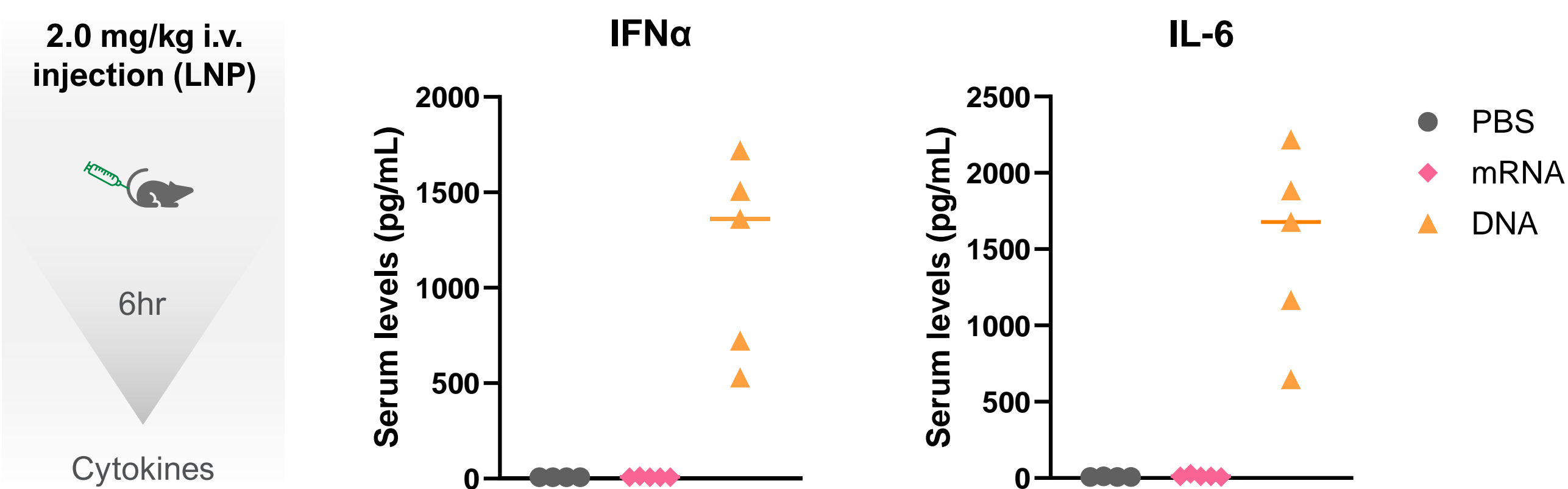
Cell targeted lipid nanoparticle (LNP) delivery of ceDNA cargo

- Durable
- Redosable
- Large cargo capacity



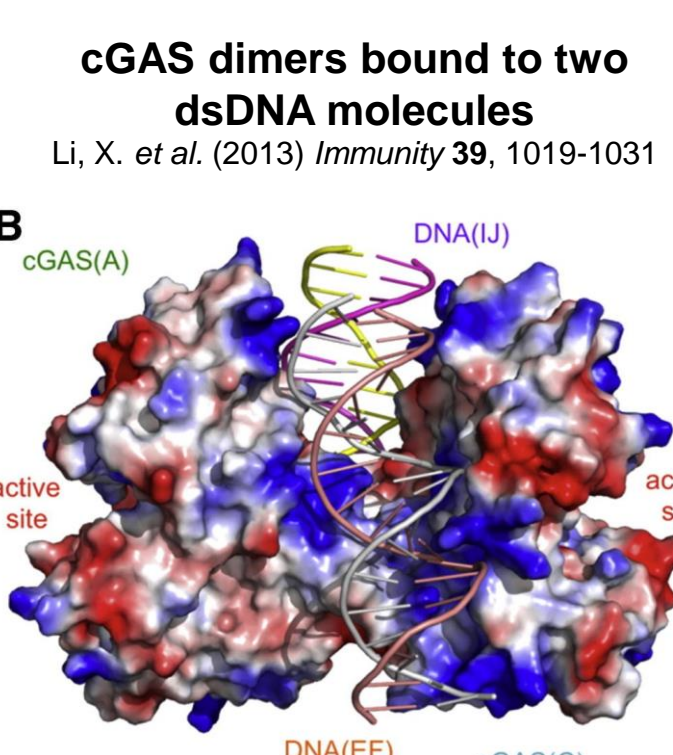
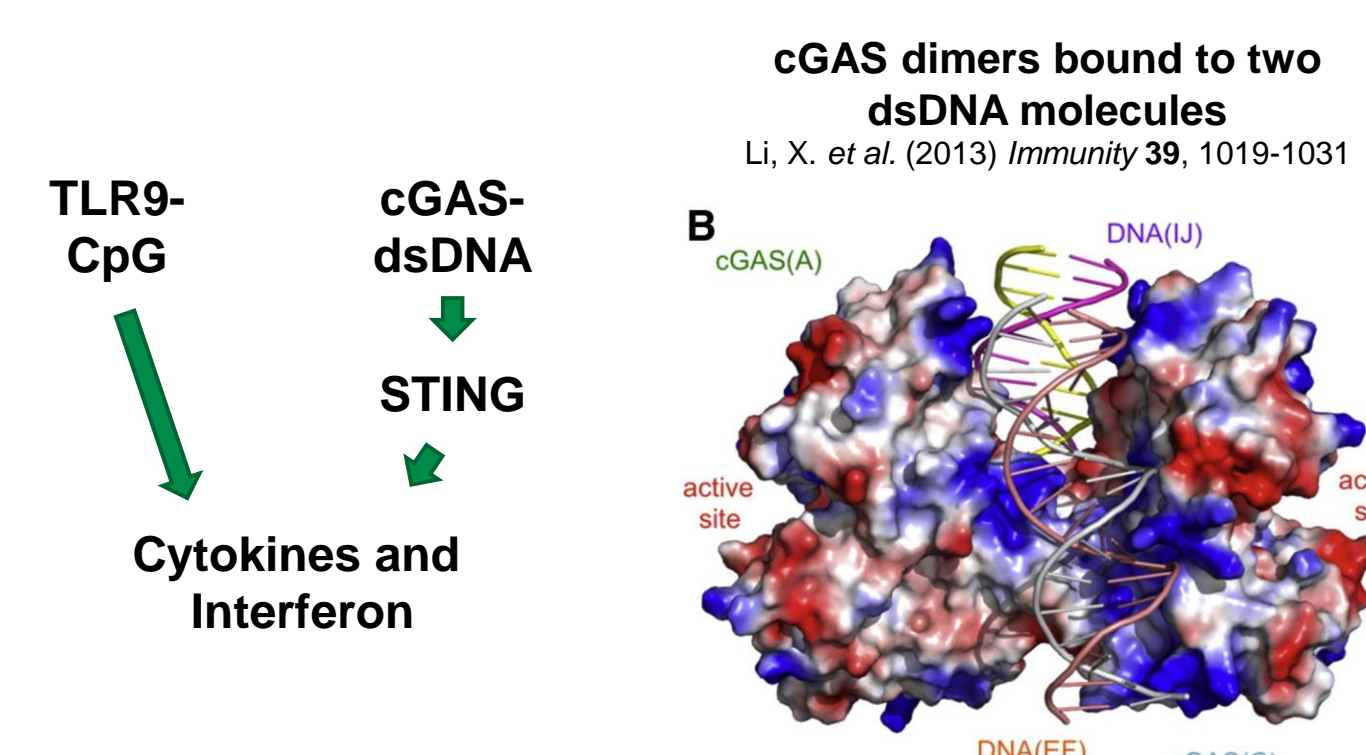
Immunogenicity limits therapeutic index of LNP-delivered DNA based genetic medicines *in vivo*

Consequences include elevated cytokines, poor tolerability, limited dose range and limited expression



cGAS-STING is the major innate immune pathway activated by LNP delivered ceDNA

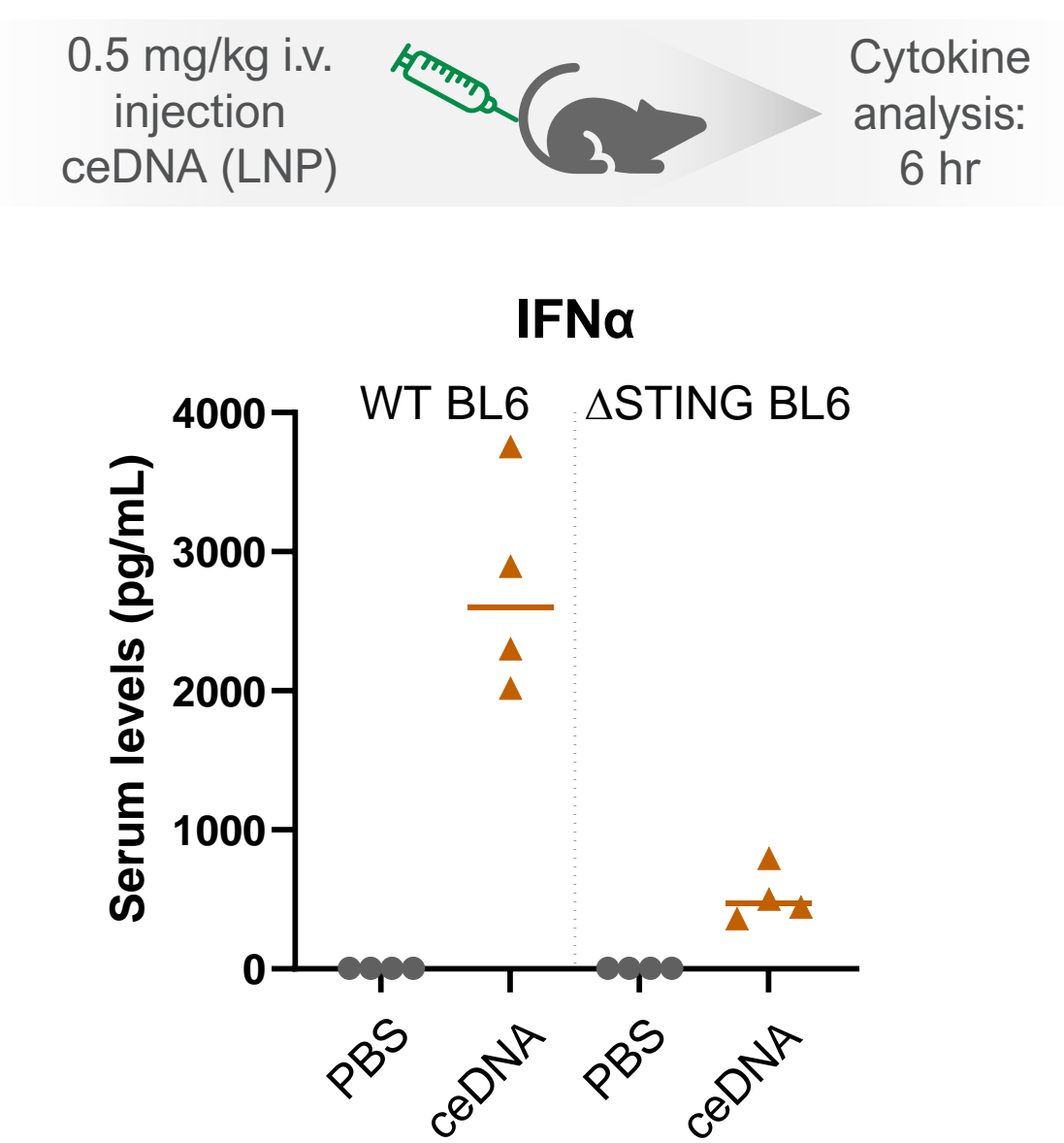
TLR9 and cGAS are innate immune sensors of DNA motifs



cGAS has higher binding affinity for dsDNA vs ssDNA
• dsDNA Kd ~87.6 nM
• ssDNA Kd ~1470 nM

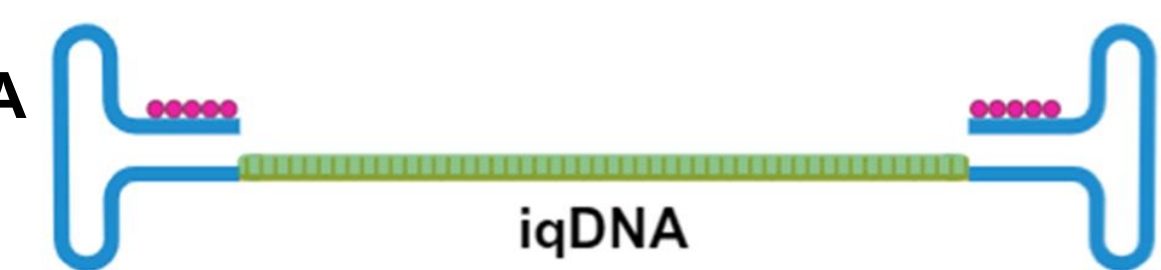
Kranzusch et al (2013), Cell Reports 3, 1362-1368

Decreased cytokine levels in STING knock-out mice

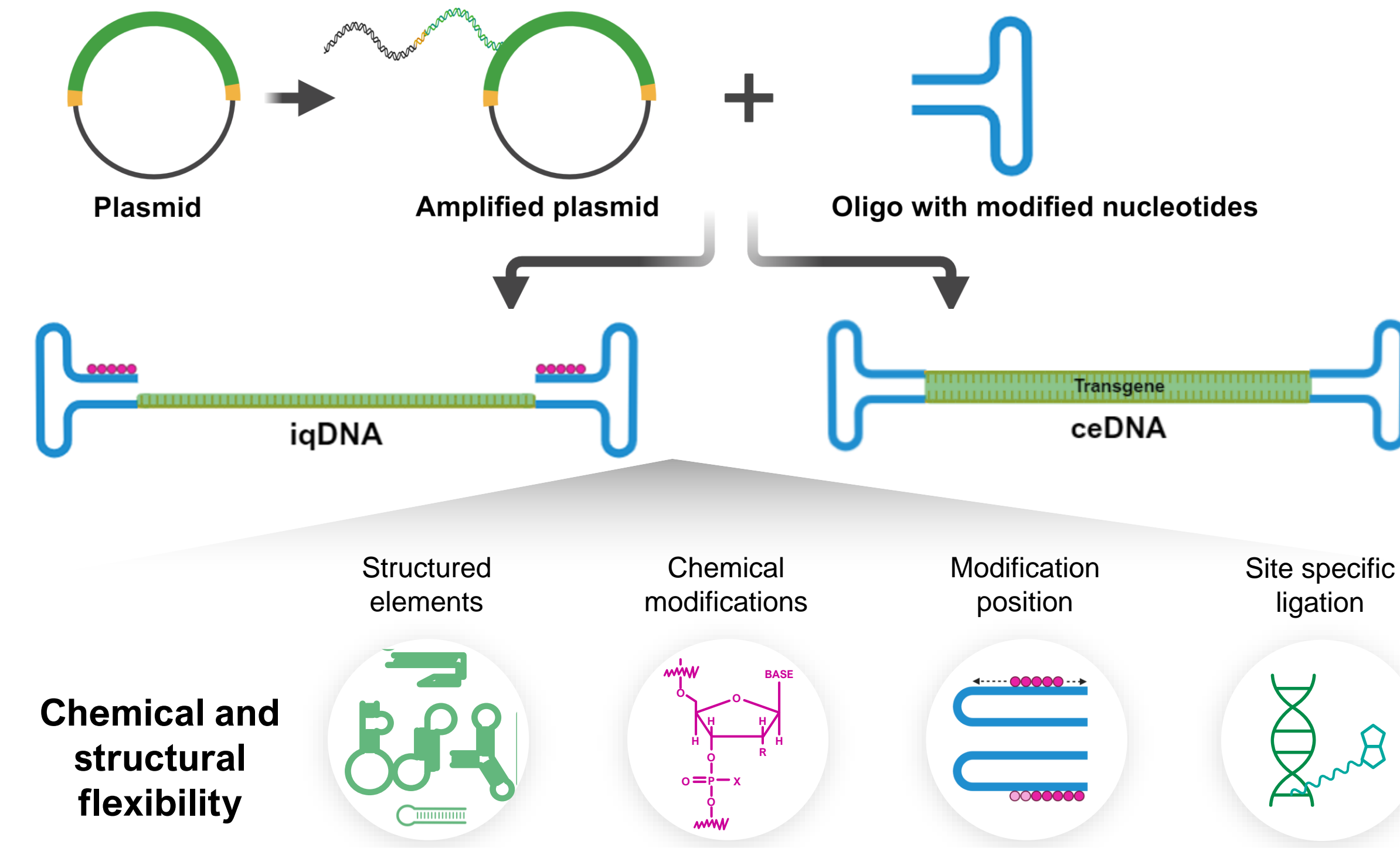


Innovation: Design and manufacture of a ssDNA cargo that, following LNP delivery:

1. Evades cGAS-STING
2. Becomes transcriptionally active inside host cell



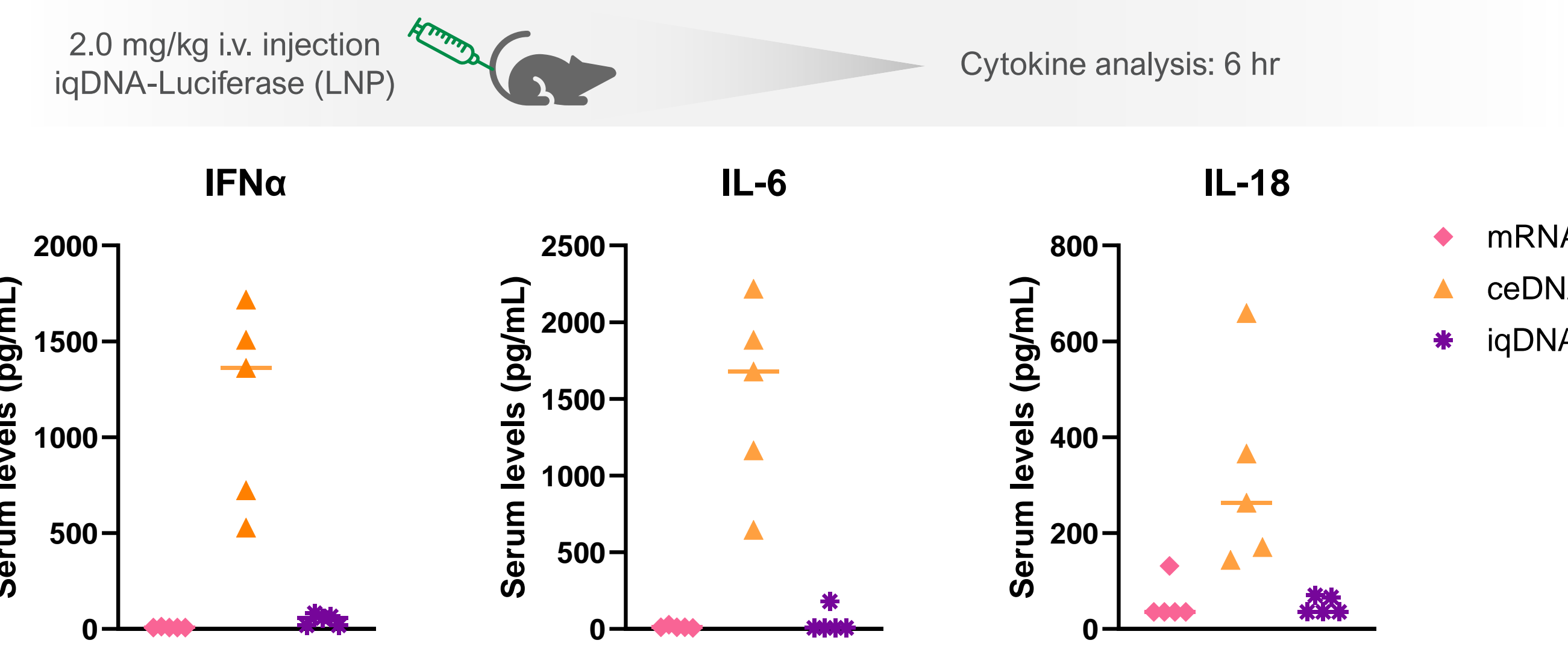
Rapid enzymatic synthesis (RES) enables generation of immune quiet DNA



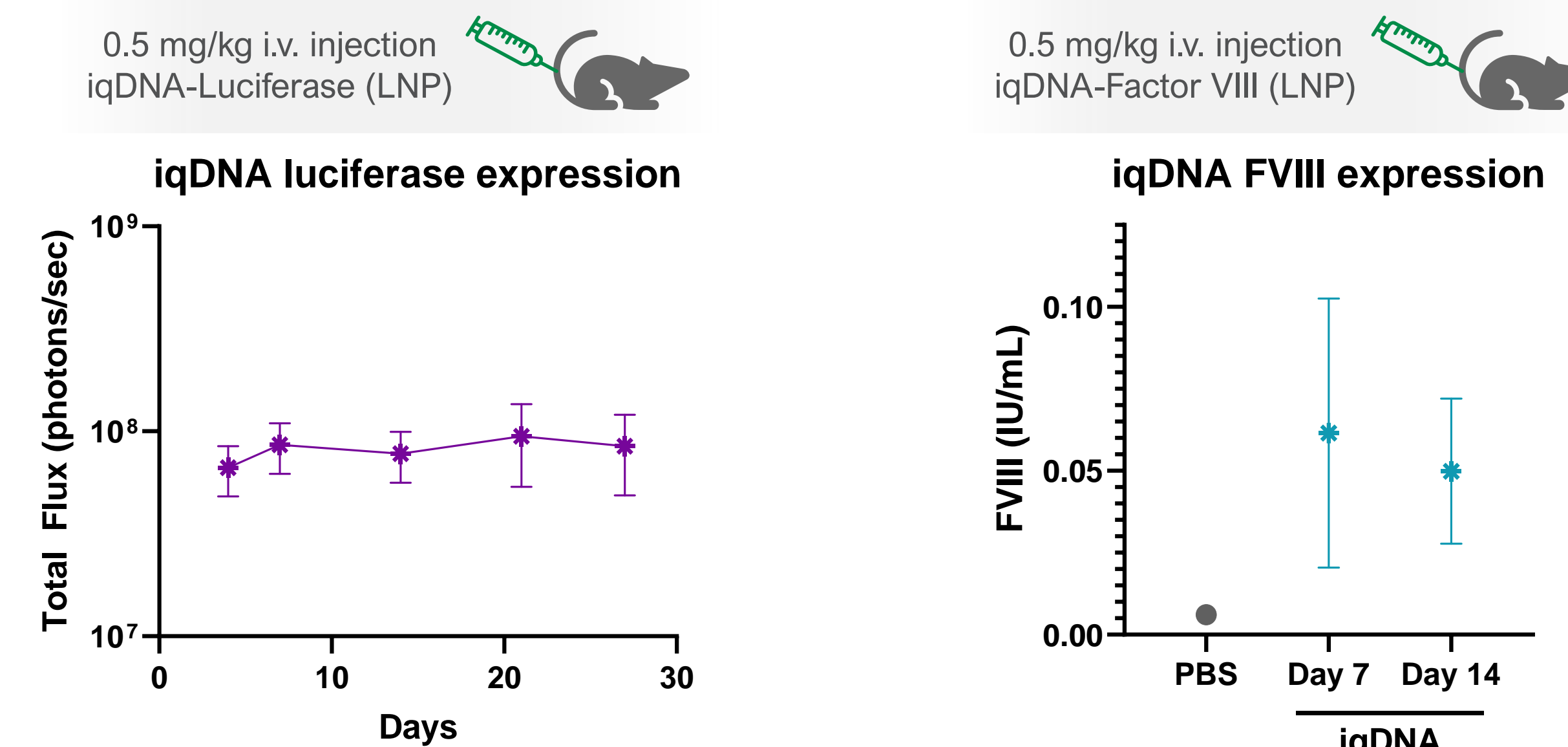
Learn more about RES: Thursday 5/9, Daniel Blackstock, poster 1232 "Rapid Enzymatic Synthesis and Scaling of DNA Gene Therapy Molecules",

iqDNA avoids innate immune activation and supports robust, durable expression (Gen1)

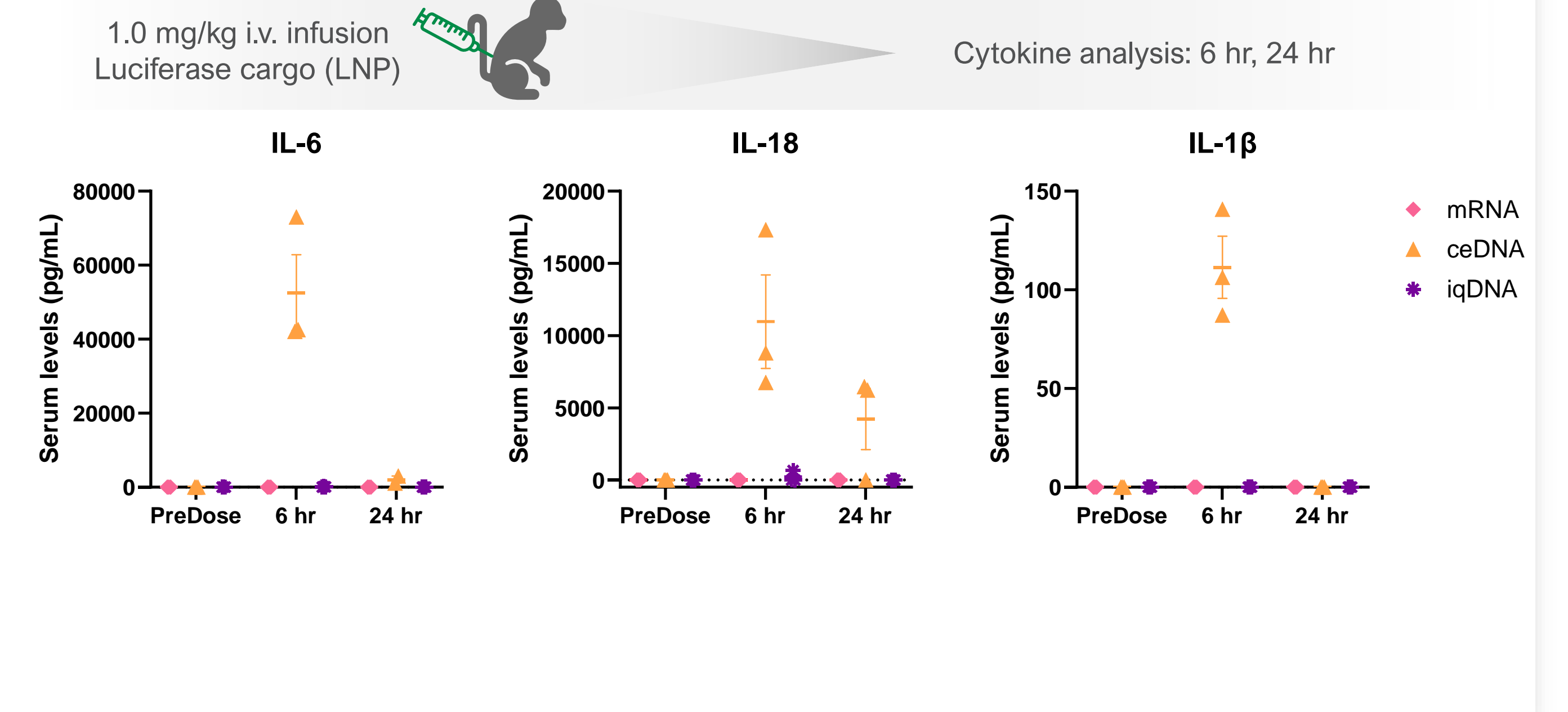
iqDNA immune profile is consistent with avoidance of cGAS-STING, TLR9 and inflammasome pathways



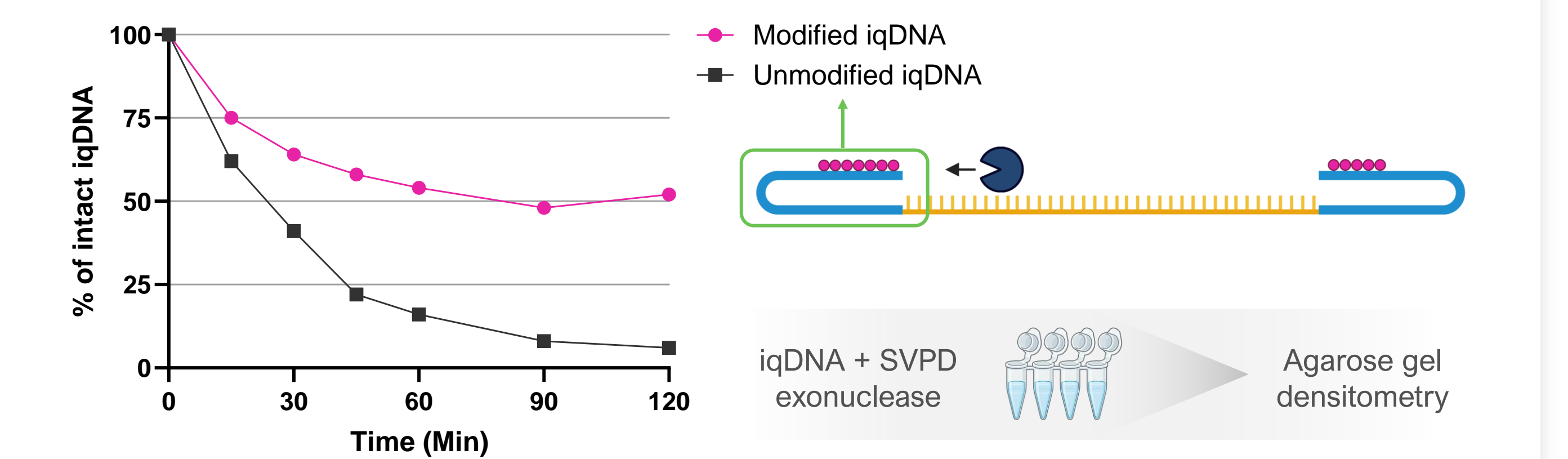
iqDNA demonstrates robust and durable expression



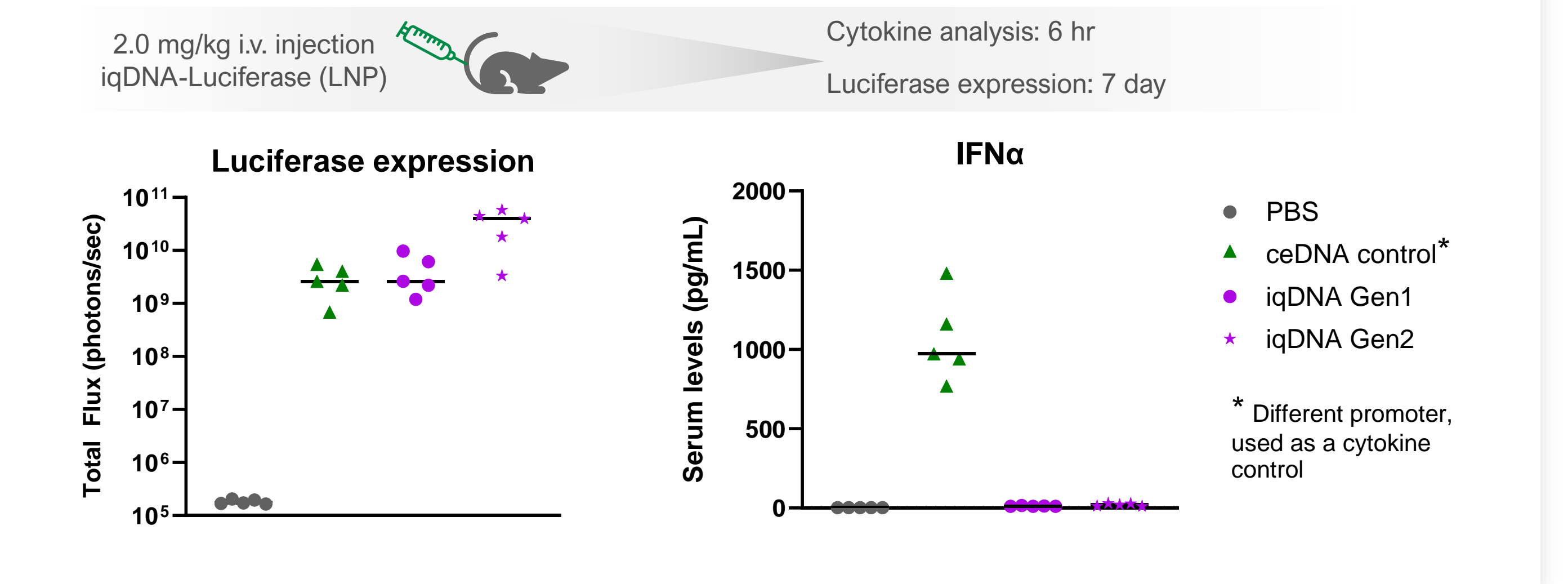
iqDNA immune quiet profile translates to non-human primates



RES iqDNA synthesis process enables end stabilization against exonucleases



iqDNA optimization increases potency while maintaining immune quiet profile (Gen2)



Next steps

- Translation of Gen2 optimization to Factor VIII iqDNA and *in vivo* assessment
- Optimization of iqDNA LNP formulations to enhance delivery of this novel immune quiet cargo