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## Identification and Characterization of PD-L1 in Bovine Placentas

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# Identification and Characterization of PD-L1 in Bovine Placentas

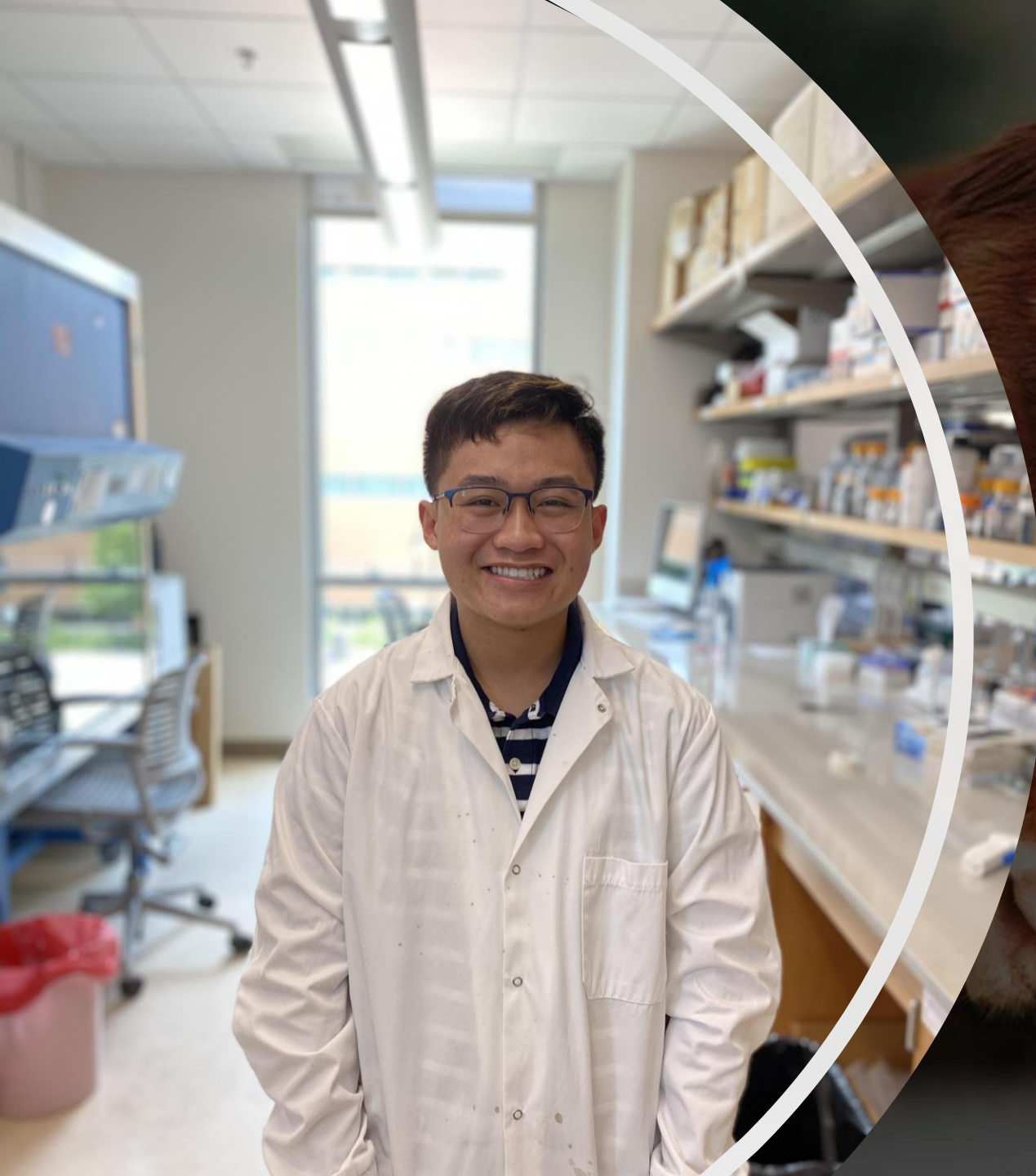
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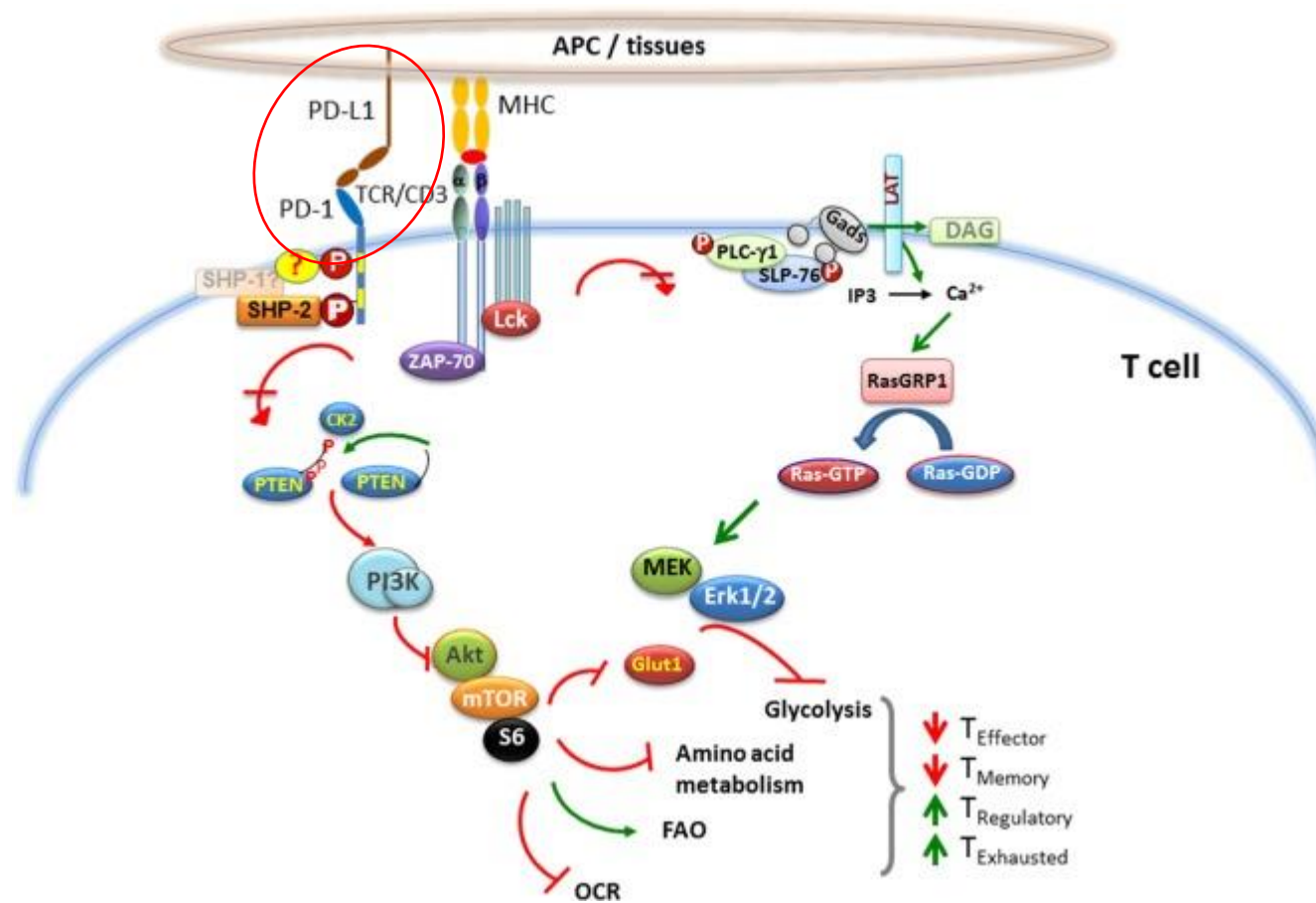
ADVS SRS 2020

Key Points: PD-L1 expression differs between trimesters and is a potential non-invasive method in inducing a receptive maternal immune system.



# Introduction and Significance

- Decline of reproductive efficiency in dairy cattle
- Pregnancy requires regulation of T lymphocytes at the fetal-maternal interface
- PD-L1 may be mechanism involved in immune tolerance to fetal antigens



# Methods

## *E. Coli* (DH5-alpha) PD-L1 PET28

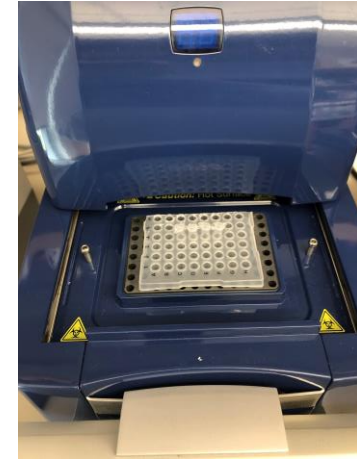


Culture and RNA  
Extraction



Qubit mRNA  
Quantification and  
Analysis

cDNA Synthesis

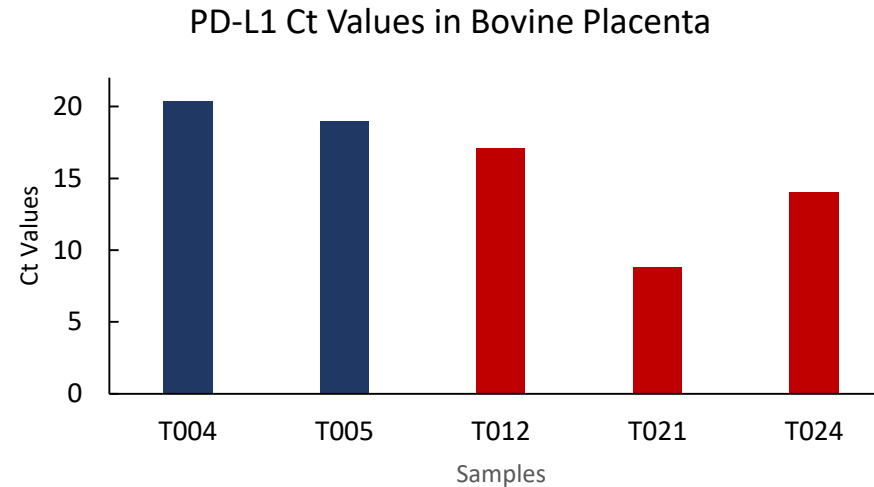


Quantitative Real-  
Time PCR

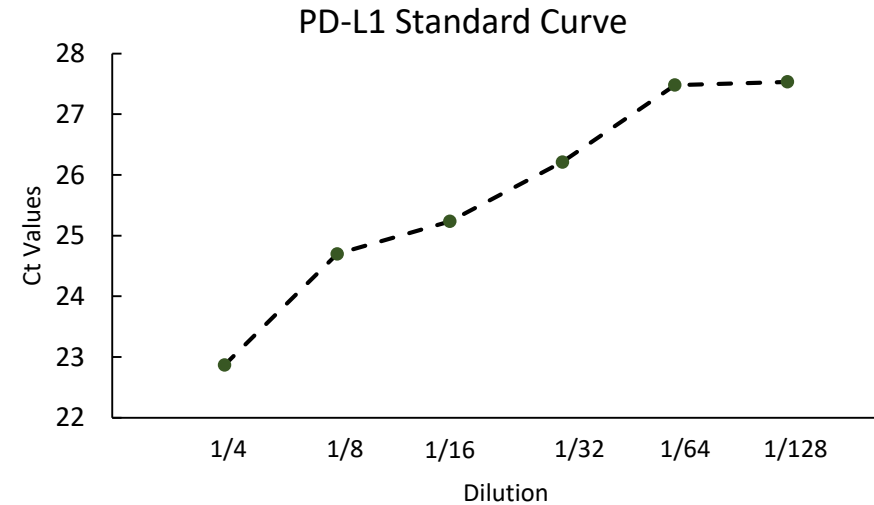


# Results and Discussion

- Amplification of PD-L1 cDNA in bovine placenta (Figure 1)
- Plasmid amplification used for standard curve
- PD-L1 RNA is expressed in bovine placenta



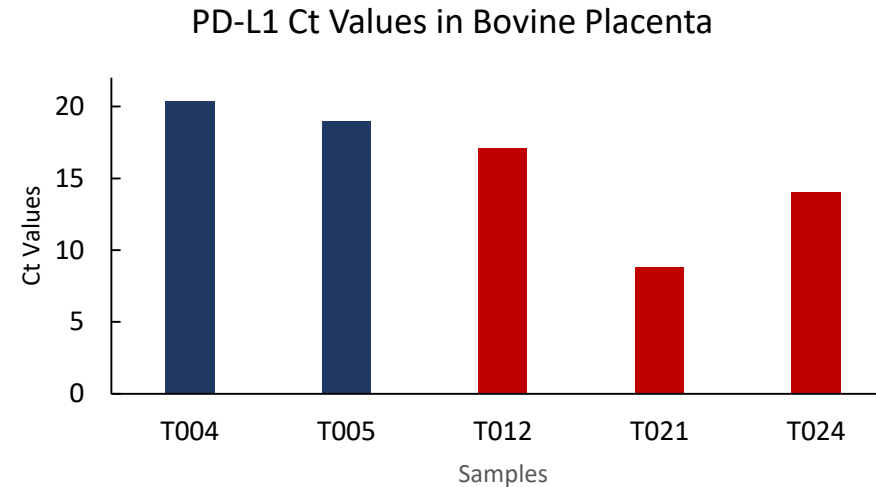
**Figure 1.** quantitative RT-PCR optimization, Red is samples in third trimester Blue is samples in second semester



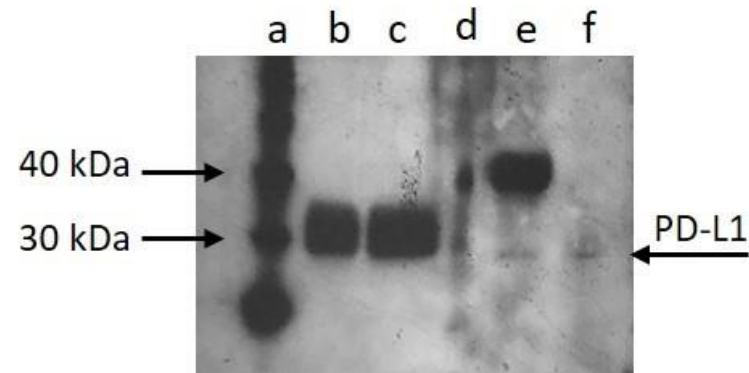
**Figure 2.** PD-L1 Plasmid Standard Curve

# Results and Discussion

- Amplification of PD-L1 cDNA in bovine placenta (Figure 1)
- Plasmid amplification used for standard curve
- PD-L1 RNA is expressed in bovine placenta
- PD-L1 protein could be identified in bovine placenta through preliminary Western blotting (Figure 4)



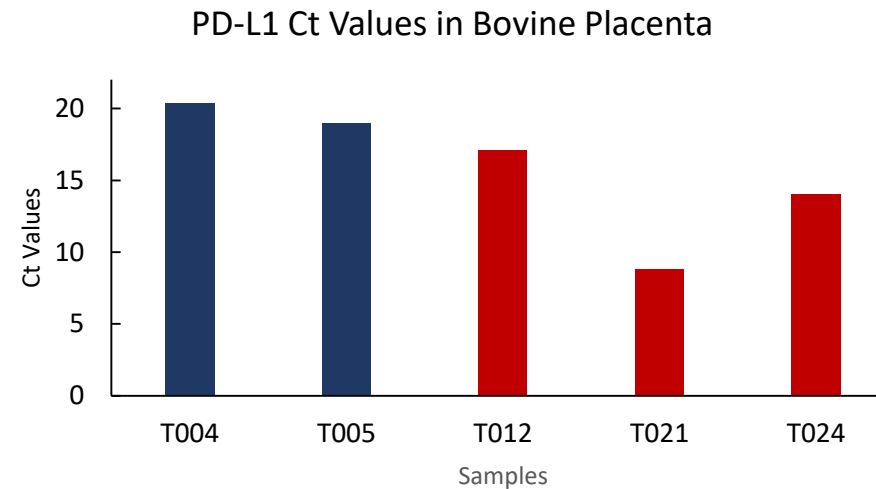
**Figure 1.** quantitative RT-PCR optimization, Red is samples in third trimester  
Blue is samples in second semester



**Figure 4.** Picture of western blotting of bovine samples against PD-L1. a) Protein standard; b) and c) PD-L1; d) skeletal muscle tissue; e) placenta tissue; f) trophoblastic extracellular vesicles.

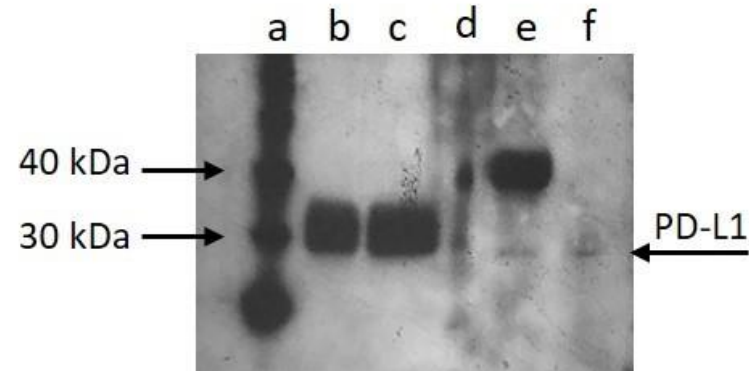
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**Figure 1.** quantitative RT-PCR optimization, Red is samples in third trimester  
Blue is samples in second semester

Under optimization!



**Figure 4.** Picture of western blotting of bovine samples against PD-L1. a) Protein standard; b) and c) PD-L1; d) skeletal muscle tissue; e) placenta tissue; f) trophoblastic extracellular vesicles.

# Conclusion

- Program death-Ligand 1 (PD-L1) differs between trimesters
- PD-L1 RNA exist in bovine placentas
- Western Blotting will be used in future research to identify PD-L1 protein
- Future directions:
  - Determine how PD-L1 modulates immune cell expression



# References



- Bardhan, Kankana et al. “The PD1:PD-L1/2 Pathway from Discovery to Clinical Implementation.” *Frontiers in immunology* vol. 7 550. 12 Dec. 2016, doi:10.3389/fimmu.2016.00550
- Silva, Ana. “Role of Placental Programmed Death Ligand1 Protein on Immune Tolerance During Bovine Pregnancy.”2019.

# Acknowledgments

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