

DISEASES FOUND IN MANURE: A SOLUTION PROPOSAL FOR REDUCTION

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Background

- Efficient use of animal manure can significantly reduce greenhouse gas emissions [1].
- Animal manure contains a lot of harmful pathogens, including Providencia spp. [2], E. Coli [3], and Salmonella spp.
- We aim to reduce some of these pathogens by genetically modifying [4] animals for increased pathogenic resistance.



Results

Genes	sgRNA (highest CRISPRscan score)	Locus (location)
SCA2	GTGAGCCGTGGCGTCCGGCGGGG	5:37431924-37431947 (+)
IRG1	TGGACGGCGGCGATCCAGGGGGG	5:490869-490892 (+)
CD79B	GAAAGGAGTGATGTCCTCGTAGG	27:1274554-1274577 (+)
PTPN3	CGTCGTGGCGGTGGCCGCAACGG	2:84350067-84350090 (-)
SMOC0	GAAGGCAGGAGCCGACCCAAAGG	5:27234138-27234161 (-)
Genes	sgRNA	Locus (location)
CCL19	GCAGCAGTCTTCAGCGTCGTTGG	8:76054763-76054786 (+)
CXCL12	GTGAGTGCGCCCAGCGACCGGGG	28:45022553-45022576 (+)
IL16	AGCTTTGGGACCGCCGCGGTGG	21:27086808-27086831 (-)
IL2RA	GGCCGTCATGCGCTGCGTGGGGG	13:17485934-17485957 (+)
IL1A	GTCATTATTTGGACCGGTATAGG	11:46493661-46493684 (-)

Conclusion

- SgRNA sequences can be edited using CRISPR-Cas9 technology (or in a lab) to genetically modify an organism
- More genes with immunological properties (such as cytokines) should be sequenced
- CRISPR scan is not 100% accurate [9]
- No lab, limited time and technology
- These edits need to be tested to ensure accuracy of the genetic modification and to confirm there aren't any side effects

Methods



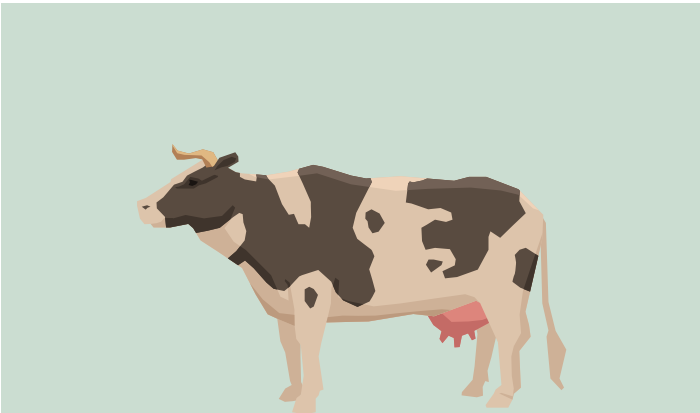
We identified 5 genes in the chicken genome [5] and cow genome [6] with potential immunological properties. We used CRISPR scan [7]---a program designed by Yale University---to identify sgRNA sequences [8], which can be used for gene editing. We used the sequence with the highest CRISPRscan score which determines the sgRNA sequence with the highest efficiency.



▲1	CRISPRscan score ▼2	Locus	Target sequence
59		11:46493661-46493684 (-)	GTCATTATTTGGACCGGTATAGG
30		11:46488009-46488032 (+)	CGTCGTTTCAGGATGCATTCTGG
29		11:46487975-46487998 (-)	AGTATAATTCGAGATATGCAGG
26		11:46486086-46486109 (-)	AGACGAACCCGCTTGCTAAAGG
20		11:46488048-46488071 (+)	AGTTGTATTTACGTTACTCTGG
82		11:46482972-46482995 (-)	TGGGGTGCCATTGCCTTCTCCGG
80		11:46489679-46489702 (-)	GTGGTGATGGTGGCAGCCAGTGG
79		11:46482972-46482994 (-)	GGGGTGCCATTGCCTTCTCCGG
66		11:46483176-46483199 (+)	GCCTGGTGGGCTGCCGTCTATGG
61		11:46489678-46489701 (-)	TGGTGATGGTGGCAGCCAGTGGG

Acknowledgements

- Our amazing mentor, Laura Sofie Vinter!



Wrap-Up

The components of animal manure: nutrients, organic matter, solids, energy, and fiber can be utilized for products such as fertilizer and biofuels [1]. Harmful pathogens get in the way of this, but those harmful pathogens may be mitigated with genetic modification.



SOURCES

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