

**The Directed Evolution of *Escherichia Coli* in the Reduction of Methane Emissions in  
Livestock Production**  
(Technical Project)

**Investigation into the Social, Physical, and Political Factors that Influence the Success of  
Renewable Forms of Energy**  
(STS Project)

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On my honor as a University student, I have neither given nor received unauthorized aid  
on this assignment as defined by the Honor Guidelines for Thesis-Related Assignments.

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## Introduction

Modern infrastructure, in its myriad forms, has yielded and continues to yield profound environmental strain on a global scale (Mgbemene et al., 2016). With applications ranging from food production to energy generation, the industrialization of Western society, and consequently much of the contemporary world, is responsible for establishing the large-scale production systems we see and rely upon today. Unfortunately, these complex systems have been identified as major drivers of climate change, primarily due to their extensive facilitation of greenhouse gas (GHG) emissions.

This paper will investigate the technical and socio-technical intricacies, respectively, of two instantiations of industrialized infrastructure. The first will be food production, particularly within the animal production industries. Enteric methane emissions – methane released via animal eructation – are currently the single largest direct source of GHG emissions in beef and dairy supply systems (Vjin et al., 2020), and are a major contributor to global anthropogenic-related methane emissions. According to aggregated data provided by the EPA, methane emissions account for approximately 20 percent of total GHG emissions and, while second only to carbon dioxide (CO<sub>2</sub>) in abundance, has a warming potential 27-30 times greater than that of CO<sub>2</sub> over 100 years (EPA, 2022). Moreover, global hunger is expected to rise by 70 percent as early as 2050 (U.N., 2021), signaling a continued demand for wide-scale livestock production. As demand for livestock production continues to rise with growing population needs, resultant methane emissions are expected to increase dramatically. Therefore, research and development into technology that can mitigate methane production from livestock with immediacy is of paramount importance and, thus, will be explored in detail within this paper.

The second form of infrastructure to be discussed will be the implementation of renewable, decentralized energy sources. Renewable energy sources have emerged as robust alternatives to heavily carbonized energy production sources (i.e. fossil fuels, natural gas, oil, etc.) (Tierney and Bird, 2020), several of which have offered decentralized modes of use. Installation of these technologies has varied internationally, with disparities seen starkly between the United States, whose renewables comprise 12 percent of its total energy production (U.S. EIA, 2021), and other Western nations such as Switzerland, whose renewable energy sources supply almost the entirety of its energy needs (Zuttel et al., 2022). In addition to political actors, Science, Technology, and Society theorists have speculated that the success of these renewables is also dependent upon the politics of the communities in which they are located (Winner, 1980). Scholar Langdon Winner summarizes these arguments that more democratized forms of government are inherently more conducive to decentralized forms of energy, insofar that the latter mirrors the basic structure of the former and vice versa, engendering congenial integration. Similar to the lack of a singular authority in a democracy, the absence of an outright demographic majority in diverse communities may make them favorable to democratized energy forms. This paper will, therefore, examine the extent to which demographic composition within a community influences the implementation of decentralized energy systems, and the implications of such results on the future of the energy sector.

While existing public policy and current consumer demands will inevitably influence the global transition to revolutionary forms of infrastructure and the rate thereof, the amenability of the world to the supplantation of industrial systems with those of novel renewability will undeniably depend on the composition of geographic populations and the scientific prowess of developing technology.

## **Genetic Engineering of *Escherichia Coli***

Methane, beyond its potency as a GHG, presents serious environmental risks due to its photochemical reactivity in the atmosphere to produce tropospheric ozone. Tropospheric ozone has been proven to be harmful to human health through direct air pollution, a hindrance to crop production, and responsible for reducing biospheres' carbon storage capabilities (Mar et al., 2022). Agricultural production is the leading source of methane emissions, with “livestock emissions – from manure and gastroenteric releases – accounting for roughly 32 percent of human-caused methane emissions.” (UN, 2021). In recent studies, methanogens – methane-producing archaea – were discovered in livestock rumen and identified as the central source of livestock methane emissions (Difford et al., 2018). Technological advances designed to reduce emissions have, unfortunately, been limited to attempted reductions at livestock production altogether, instead focusing on plant-based production, alterations in livestock diet, mainly in the forms of feed additives, and anti-methanogen vaccines (Kaufmann et al., 2022). These strategies, however, have either had variable results, been deemed impractical when weighed against countervailing demand, or displayed disruption to livestock microbiota, making them largely unviable (Callaway et al., 2019).

One area of research that shows promise in curtailing carbon emissions is the use of directed evolution of microorganisms to impart carbon-digestive abilities. Directed evolution has emerged as a promising method in gene editing and protein engineering in which organisms are subjected to iterative rounds of mutagenesis until a desired response is achieved. The technique has markedly evolved with the incorporation of complementary technologies such as machine learning and viral vector modalities, where genes can be more precisely targeted. Gleizer et al. successfully demonstrated an experiment in which *Escherichia coli* (*E. Coli*) were genetically

modified via directed evolution to express carbon-fixing autotrophy, despite naturally occurring heterotrophy; the bacteria were consequently able to consume CO<sub>2</sub> and utilize the compound for all necessary biomass (Gleizer et al., 2019). In the technical portion of this paper, a similar methodology will be employed in which *E. Coli* will be genetically modified by means of directed evolution to consume methane as its primary energy source. Genetic targets involved in *E. Coli* metabolism will be identified through literature analysis and machine learning algorithms, and random mutagenesis of discerned targets will be accomplished by way of viral vector insertion (Nayerossadat et al., 2012). Genetic mutations will be confirmed using polymerase chain reaction (PCR) and relative methane production will be measured with gas chromatography (GC). This proposed method of genomic sequencing and directed evolution of *E. coli* species to consume methane will provide a viable means of altering methane production in ruminating species while circumventing digestive complications in livestock. Additionally, it will leverage well understood techniques with demonstrable success and apt applicability.

### **Establishment of Democratized Forms of Energy**

The discussion around democratized energy has been one of burgeoning interest (Sorman et al., 2020). There are three key and distinct reasons why it has gained so much attention, each accessing a specific realm of socio-technical interpretation.

The first is that energy, in and of itself, is a necessary good (T. Energy, 2021). To exist in modern society, or at the very least, a commonly understood version of daily life, one must have access to power. This creates a fundamental reliance upon the centralized energy source.

The second is the organization of this basic need. Most countries and societies rely on enormous power grids – networks of interconnected and often centralized sources of power,

distributed across the country's geographical boundaries (Munro et al., 2020). In this structure, energy is produced in a consolidated manner and distributed according to individual need.

Although each member of society has access to the grid and can modulate individual usage, there is no user input on the behind-the-scenes production. The generation of this treasured commodity is held closely within a socio-technical elite (i.e. energy companies) that have near unchallenged control over the energy supply of a given community.

Third, technology must meet consumer demands while responding to the pressing issues of the era. In the present case, countervailing interests are manifested in climate change, depletion of resources, and harm to local environments. A new energy form, especially if intended to eventually supplant or reduce the dependency on existing sources, must match energetic needs of the modern citizen, with reliability and efficiency.

My working definition of democratized energy is the idea that individuals participate in energy production and are either able to become fully or partially self-reliant or receive financial incentives to contribute back to the grid. With regards to the first requirement, in which every citizen must access power, it engages the individual need for energy by contributing to energetic production. Admittedly, not a difficult standard to meet. As it pertains to the second category, it revolutionizes the very politics of energy production. As Winner (1980) discusses, technology has politics. The top-down system described above lends itself to a consolidated form of government, with no individual input: not very democratic. It relies on a social sense of conformity and reliance upon strict governmental authority, albeit perhaps to a monopolistic private business. But, if an energy system could capitalize on political attitudes, not just with the idea of its benefits, but with the acceptance of its essential structure, that could mean an entirely different conceptualization in the way in which we approach energy reform. Finally, regarding

how it might meet current pressing issues, democratized energy has become synonymous with renewable energy. This association is primarily due to the fact that innovation in certain energy forms, namely solar photovoltaics and biomass sources (Vezzoli et al., 2018), coupled with the inability of individual production of fossil fuels or oil or coal, has made them inseparable entities.

Extending this understanding to other facets of society is not unintuitive. In the same way Winner espouses a relationship between the politics of a society and its energy systems, it's not difficult to imagine the possibility that other factors, such as diversity in communities, play significant roles as well. When endeavoring to redesign the very nature of these longstanding systems, it's essential that the implementation of the new is done with proper care and consideration, so as not to prematurely preclude their success, and be left dealing with an even greater existential behemoth.

### **Methods and Research Question**

The research question I aim to explore is how have decentralized forms of energy production (i.e. solar energy) been more successful in diverse communities? Alternatives to unsustainable forms of energy production must be realized if the ongoing damage to the environment is to be stopped. Therefore, researching the factors that influence these alternatives' success, both technically and societally, will be of great importance.

To examine the relationship between community diversity and receptiveness to decentralized energy, I will select a single, distinct metric of diversity: ethnic composition. In selecting 'communities of interest', I will select the countries Switzerland and Germany with the intention of designing a specified, comparative case study. These two countries share a multitude

of characteristics that will hopefully avoid confounding factors. For example, Switzerland and Germany are geographically adjacent to one another, they are fairly similar in size, and exhibit policy initiatives aimed at expanding renewable energy forms. The key area of differentiation will be their relative ethnic homogeneity, that is, the percentage of total population comprised of an ethnic majority and subsidiary ethnic minorities. Further analysis will be conducted in order to investigate which specific ethnic minorities exist and if that is also a critical point of distinction.

The second variable under consideration will be the success of decentralized energy forms within these localities. This will be assessed by comparing the fractional contribution of these forms to each country's total energy production. Additionally, the comparative incorporation of decentralized energy forms into the civil infrastructure of Swiss and German localities, both of which have extensive energy cooperatives in place (Schmid et al., 2020). Determining if these communities implemented these technologies and further embellished upon their installment or organized around their establishment, as in the case of studies in Italy (Formolli et al., 2022). Another example of such phenomena is the energy sharing programs implemented in France and other countries, where communities' local economic and interrelational ecosystems adapted to new energy systems (Fontaine and Labussière, 2019). Another method of potential analysis is the resultant change in pertinent public policy following these decentralized energy systems' instantiation.

Both categories of data will be extracted from governmental entities, as well accredited scientific literature. Exploration into these metrics will provide crucial insight into the broader question of what makes for preferential success of a given energy form. Technological



advancement indubitably leads the charge in commodifying novel, decentralized energy media, but while the calculus begins there, where it ends, or even continues, remains to be seen.

### **Conclusion**

The present state of established infrastructure systems imparts overwhelming harm onto the environment, both in the context of global pollution and the climate crisis. Specifically, the realms of agricultural and energy production have been classified as areas with notable GHG emission output quotients and ever-increasing demand, necessitating intense remediation.

The success of the proposed technology will largely reshape the livestock industry and its environmental impact. With anthropogenic-related emissions reduced by almost a third of its current state, modern production methods, even without systemic overhaul, will likely be able to supply the growing global food demands while avoiding deleterious environmental consequences.

The results concerning the relationship between ethnic diversity and decentralized energy success are expected to corroborate a direct relationship. Decentralized energy systems leverage an appreciation of systemic variety and independence that is believed to be more prevalent in ethnically diverse communities. Moreover, the decentralized energy forms rely on social structures in which there is no discernible societal hierarchy or innate class system that could compel a top-down system of energy or accessibility thereof. For these reasons, the findings of this research is expected to establish the aforementioned relationship and enhance our understanding of energy systems to regard them as impetuses for social change.

## References

- Callaway, E. (2019). E. coli bacteria engineered to eat carbon dioxide. *Nature*, 576(7785), 19–20.  
<https://doi.org/10.1038/d41586-019-03679-x>
- Energy, T. (2021, January 14). *What Is Energy? A Guide to Understanding Energy*. Tara Energy.  
<https://taraenergy.com/blog/what-is-energy-a-guide-to-understanding-energy/>
- Fontaine, A., & Labussière, O. (2019, November 1). Community-based solar projects: sun-sharing politics and collective resource construction trials. *Local Environment*, 24(11), 1015 - 1034.
- Formolli, M., Croce, S., Vettorato, D., Paparella, R., Scognamiglio, A., Mainini, A. G., & Lobaccaro, G. (2022). Solar Energy in Urban Planning: Lesson Learned and Recommendations from Six Italian Case Studies. *Applied Sciences*, 12(6), Art. 6. <https://doi.org/10.3390/app12062950>
- G. F. Difford *et al.*, “Host genetics and the rumen microbiome jointly associate with methane emissions in dairy cows,” *PLOS Genetics*, vol. 14, no. 10, p. e1007580, Oct. 2018, doi: 10.1371/journal.pgen.1007580.
- Gleizer, S., Ben-Nissan, R., Bar-On, Y. M., Antonovsky, N., Noor, E., Zohar, Y., Jona, G., Krieger, E., Shamshoum, M., Bar-Even, A., & Milo, R. (2019). Conversion of Escherichia coli to Generate All Biomass Carbon from CO<sub>2</sub>. *Cell*, 179(6), 1255-1263.e12. <https://doi.org/10.1016/j.cell.2019.11.009>
- Kauffman, J.B., Beschta, R.L., Lacy, P.M. *et al.* Livestock Use on Public Lands in the Western USA Exacerbates Climate Change: Implications for Climate Change Mitigation and Adaptation. *Environmental Management* 69, 1137–1152 (2022). <https://doi.org/10.1007/s00267-022-01633-8>
- Mar, K. A., Unger, C., Walderdorff, L., & Butler, T. (2022). Beyond CO<sub>2</sub> equivalence: The impacts of methane on climate, ecosystems, and health. *Environmental Science & Policy*, 134, 127–136.  
<https://doi.org/10.1016/j.envsci.2022.03.027>

- Mgbemene, C. A., Nnaji, C. C., & Nwozor, C. (2016). Industrialization and its Backlash: Focus on Climate Change and its Consequences. *Journal of Environmental Science and Technology*, 9(4), 301–316.  
<https://doi.org/10.3923/jest.2016.301.316>
- Nayerossadat, N., Maedeh, T., & Ali, P. A. (2012). Viral and nonviral delivery systems for gene delivery. *Advanced biomedical research*, 1, 27. <https://doi.org/10.4103/2277-9175.98152>
- Robertson Munro, F., & Cairney, P. (2020). A systematic review of energy systems: The role of policymaking in sustainable transitions. *Renewable and Sustainable Energy Reviews*, 119, 109598.  
<https://doi.org/10.1016/j.rser.2019.109598>
- Schmid, B., Meister, T., Klagge, B., & Seidl, I. (2020). Energy Cooperatives and Municipalities in Local Energy Governance Arrangements in Switzerland and Germany. *The Journal of Environment & Development*, 29(1), 123–146. <https://doi.org/10.1177/1070496519886013>
- Sorman, A. H., Turhan, E., & Rosas-Casals, M. (2020). Democratizing Energy, Energizing Democracy: Central Dimensions Surfacing in the Debate. *Frontiers in Energy Research*, 8.  
<https://www.frontiersin.org/articles/10.3389/fenrg.2020.499888>
- U.S. energy facts explained—Consumption and production—U.S. Energy Information Administration (EIA)*. (n.d.). Retrieved October 21, 2022, from <https://www.eia.gov/energyexplained/us-energy-facts/>
- US EPA, O. (2016, January 12). *Understanding Global Warming Potentials* [Overviews and Factsheets].  
<https://www.epa.gov/ghgemissions/understanding-global-warming-potentials>
- Tierney, S., & Bird, L. (2020). *Setting the Record Straight About Renewable Energy*.  
<https://www.wri.org/insights/setting-record-straight-about-renewable-energy>
- Vezzoli, C., Ceschin, F., Osanjo, L., M’Rithaa, M. K., Moalosi, R., Nakazibwe, V., & Diehl, J. C. (2018). Distributed/Decentralised Renewable Energy Systems. In C. Vezzoli, F. Ceschin, L. Osanjo, M. K. M’Rithaa, R. Moalosi, V. Nakazibwe, & J. C. Diehl (Eds.), *Designing Sustainable Energy for All:*

*Sustainable Product-Service System Design Applied to Distributed Renewable Energy* (pp. 23–39).

Springer International Publishing. [https://doi.org/10.1007/978-3-319-70223-0\\_2](https://doi.org/10.1007/978-3-319-70223-0_2)

Vijn, S., Compart, D. P., Dutta, N., Foukis, A., Hess, M., Hristov, A. N., Kalscheur, K. F., Kebreab, E., Nuzhdin, S. V., Price, N. N., Sun, Y., Tricarico, J. M., Turzillo, A., Weisbjerg, M. R., Yarish, C., & Kurt, T. D. (2020). Key Considerations for the Use of Seaweed to Reduce Enteric Methane Emissions From Cattle. *Frontiers in Veterinary Science*, 7.

<https://www.frontiersin.org/articles/10.3389/fvets.2020.597430>

Winner, L. (1980). Do Artifacts Have Politics? *Daedalus*, 109(1), 121–136.

<http://www.jstor.org/stable/20024652>