An Roinn Talmhaíochta,
Bia agus Mara
Department of Agriculture,
Food and the Marine

14SF847 - The comparative public and animal health risks associated with spreading anaerobic digestate, animal manure and slurry on land: science, policy and practice

Final Report

This project was funded under the Department of Agriculture, Food and the Marine Competitive Funding Programme.

SUMMARY

The overall objective of this project was to compare the public and animal health risks associated with land spreading of AD digestate (derived from manure/slurry and food waste and combinations thereof) (with & without pasteurisation) versus slurry. The target pathogens included Salmonella enterica serovars Senftenberg and Newport, Listeria monocytogenes and Escherichia coli O157 (representing Gram-negative bacteria), Enterococcus faecalis (representing Gram-positive bacteria), Clostridium spp. (representing sporeforming bacteria), Mycobacterium spp. (representing acid-fast bacteria), non-enveloped enteric viruses (representing viruses) and Cryptosporidium parvum (representing highly resilient parasites). Other indicator bacteria were used when appropriate.

The main findings may be summarised as follows; [1] AD feedstock material may be contaminated with pathogens; [2] based on laboratory experiments, pathogenic microorganisms may survive the AD process, depending on the initial concentration of the organism, feedstock formulation, AD conditions and retention time. However, in the commercial AD processes this seems unlikely considering the dilution effect. Moreover any pathogens in the digestate may die off during storage before spreading; [3] the fermentation conditions that maximise pathogen destruction can be predicted using mathematical models developed in this project; [4] pasteurisation of digestate at 70 °C for 1 hour will eliminate most pathogens except sporeformers; [5] pathogens survive in digestate applied to land but present no greater a risk than slurry in terms of cross-contamination of soil, and [6] pathogens in digestate applied to land pose less of a risk to water contamination as compared to pathogens in slurry.

This research addresses some of the concerns regarding AD and the land-spreading of digestate as a fertiliser and the findings support the development of the AD sector in Ireland.

KEYWORDS

digestate, food safety

ACRONYM	AD-Policy
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Section 1 - Research Approach & Results

Start Date

01 February 2015

End Date

31 October 2019

Select Research Programme

Food Institutional Research Measure

TRL Scale

TRL 1: Basic Principles Observed

NRPE Priority area

Sustainable Food Production and Processing

Total DAFM Award

€815,050.20

Total Project Expenditure

€779,965.68

Rationale for undertaking the Research

Anaerobic digestion (AD) is a process in which biodegradable materials are decomposed in airtight vessels under anaerobic conditions. This process produces biogas which can be used to generate heat and/or electricity, or which can be upgraded to methane and injected into the gas grid or used as vehicle fuel. The residue from the digesters is called 'digestate' and can be used as an organic fertiliser or soil improver. Materials that fall under the scope of the EU Animal By-product (ABP) Regulations (EU Regulation 1069/2009 and EU Regulation 142/2011) are subject to rules aimed at protecting public and animal health. These Regulations require AD raw materials or digestate to be heated to 70 °C for a minimum of 60 minutes with a particle size of 12 mm, before heat being applied. The legislation also allows for a derogation from the requirement for a pasteurisation treatment in AD plants transforming animal waste, provided the competent authority does not consider it to present a risk for the spread of any serious transmissible diseases. However, animal waste presents a significant risk with regard to the transmission of serious transmissible diseases to both humans and animals, including E. coli O157, Salmonella spp., Mycobacterium tuberculosis, Cryptosporidium spp., Clostridium botulinum, Mycobacterium avium subsp. paratuberculosis, and bovine viral diarrhoea virus (BVDV). AD digestate may also represent a greater risk than slurry as the raw materials used in AD bioreactors are typically derived from and spread on many farms.

Methodology

A range of AD feed materials as well as raw and dried digestate, were tested for 5 key bacterial pathogens; Salmonella spp., Listeria monocytogenes, Escherichia coli O157, Enterococcus faecalis and Clostridium spp. using ISO or equivalent methods. Raw material, pasteurised feed material, unpasteurised and pasteurised digestate samples were collected from 5 commercial AD plants and tested for the presence of Cryptosporidium spp, Mycobacterium avium subspecies paratuberculosis (MAP), noro-, rota- and astrovirus and a range of antimicrobial resistance genes (ARGs) using conventional PCR.

Survival studies were undertaken at mesophilic temperatures in triplicate 10 L continuously stirred tank bioreactors fed with slurry and food production waste to mimic Irish full-scale anaerobic digestion. This research examined the effect of feedstock mix and process configuration on the fate of the target pathogens.
Pre- and post-AD pasteurisation was investigated using a range of time-temperature combinations; 60 °C x 1 hour, 70 °C x 1 hour, 80 °C x 30 minutes, 80 °C x 1 hour, 90 °C x 30 minutes and 90 °C x 1 hour. These treatments were carried out in 7 matrices; 4 commercial feedstock recipes, 2 digestates and a brain heart infusion (BHI) broth control, inoculated with a known level of each target bacterium; Clostridium sporogenes spores, Listeria monocytogenes (marked strain), Salmonella Newport (marked strain), Enterococcus faecalis and E. coli O157.
Field trials were carried out in Teagasc's Johnstown Castle and Ashtown facilities to investigate bacterial survival and presence in run-off during rainfall when applied in pasteurised and unpasteurised digestate and in slurry.
Predictive modelling and risk assessment tools were developed and parameterised from data generated as part of controlled laboratory experiments. Input uncertainty and variability were captured in a Monte Carlo probabilistic modelling approach.

Project Results

Bacterial, viral, mycobacteria and/or parasitic pathogens were detected in the feedstock materials and in the digestate samples. It was concluded that AD feedstock material may be contaminated with pathogens and these either survive the AD process or there is postdigestion cross-contamination.
Pathogenic bacteria survived the AD fermentation process, but this depended on the initial concentrations and the retention time. The data generated was used to calculate the time required to kill 90% of a given bacterial population which were as follows; Salmonella spp.: 1 to 3 days; Listeria monocytogenes: 3 to 24 days; Escherichia coli O157: 2 to 3 days; Enterococcus faecalis: 2 to 7 days, and Clostridium spp.: 3 to 15 days. It was concluded that these bacteria would be eliminated during the fermentation process if the initial concentrations were sufficiently low and if the retention time was sufficiently long. The laboratory scale experiments suggested that parasites, viruses & mycobacteria probably do not survive anaerobic digestion.
Further studies on the impact of feedstock mixture and process configuration were undertaken using four commercially relevant feedstock mixtures in a small-scale model system. Mixtures were individually spiked with the 5 bacterial pathogens and their survival assessed. T90 (time required for bacterial population to decrease by 1 log unit/ or 90%) was calculated for each recipe/bacterial combination. The average T90 value per recipe was 3.64 – 6.73 days. Feedstock mixtures composed of slurry & grease-trap waste (fat, oil and grease, FOG) in a 2:1 ratio and slurry & food waste, (1:3 ratio) had average T90 values of 5.06 and 3.86 days, respectively. However, the latter had a T90 value for Listeria monocytogenes of 23.47 days, which could be an issue depending on retention time in the bioreactor. Pre- and post-AD pasteurisation was investigated using a range of time-temperature combinations in 7 matrices; 4 commercial feedstock recipes, 2 digestates and a brain heart infusion (BHI) broth control. These mixtures were spiked with a known level of each target bacterium; Clostridium sporogenes spores, Listeria monocytogenes (marked strain), Salmonella Newport (marked strain), Enterococcus faecalis and E. coli O157. As expected, C. sporogenes was not eliminated by any of the pasteurisation treatments. In most cases, post-AD pasteurisation showed a greater reduction regardless of treatment and was effective at 70 °C for 1 h. This study suggested that the current EU standard of 70 °C for one hour is sufficient to eliminate the majority of bacterial pathogens and does so most effectively post-AD. Field trials investigating bacterial survival and presence in run-off during rainfall when applied in pasteurised and unpasteurised digestate and in slurry, found that pathogens survive in digestate applied to land but present no greater a risk than slurry in terms of cross contamination of the soil. The quantitative risk assessment highlighted that there is very low risk due to Clostridium spp., norovirus and Salmonella across all the scenarios. However, Cryptosporidium, E. coli O157 and Mycobacterium may pose a higher risk for the application of raw manure or slurry.

Section 2 - Research Outputs

Summary of Project Findings

The main findings include:

1. AD feedstock material may be contaminated with pathogens;
2. Pathogenic microorganisms may survive the AD process although this is unlikely under commercial conditions;
3. Pasteurisation of digestate at 70 °C for 1 hour will eliminate most pathogens except sporeformers;
4. Pathogens survive in digestate applied to land but present no greater a risk than slurry in terms of cross-contamination of soil, and
5. Pathogens in digestate applied to land pose less of a risk to water contamination as compared to pathogens in slurry.

A modelling tool was developed to predict pathogen survival/destruction based on key fermentation parameters including temperature and pH. This deterministic model was developed within Microsoft Excel to consider the behaviour of 10 pathogens (Clostridium spp., coliform bacteria (other than E. coli), Cryptosporidium parvum, Enterococcus spp. (E. faecalis), E. coli O157, Escherichia coli, feline calicivirus FCV (a surrogate for norovirus), Listeria monocytogenes, Mycobacterium thermoresistibile and Salmonella Newport) during the AD process. The model is a user-friendly tool allowing the user to select from a suite of inputs to model a given scenario.
A quantitative assessment of the risk of spreading a transmissible disease through the use of unpasteurised digestate from an AD plant was undertaken. The simulated mean daily human exposure (HE) and the annual probability of illness can be predicted using this model.
A quantitative assessment of the comparative risks associated with applying digestate, manure and slurry to land was also undertaken. This model predicted that Cryptosporidium, VTEC E. coli O157 and Mycobacterium may pose a high risk for the application of raw manure or slurry and the use of AD reduces this risk significantly. This model also found that there was a higher annual probability of risk associated with spreading unpasteurised mesophilic AD compared with pasteurised digestate.

Summary of Staff Outputs

Research Output	Male	Female	Total Number
PhD Students	2	2	4
Post Doctorates	1	0	1

Summary of Academic Outputs

Research Outputs	Total Number	Details
Publications in Peer Reviewed Scientific Journals	12	1. Agathe Auer, Vande Burgt, Nathan; Abram, Florence; Barry, Gerald; Fenton, Owen; Markey, Bryan; Nolan, Stephen; Richards, Karl; Bolton, Declan; De Waal, Theo; Gordon, Stephen; O'Flaherty, Vincent; Whyte, Paul; Zintl, Annetta (2016). Agricultural anaerobic digestion power plants in Ireland and Germany: policy & practice. Journal of the Science of Food and Agriculture, 97, 3, 719-723.

2. Stephen Nolan, Nicholas Walters, Fiona Brennan, Agathe Auer, Owen Fenton, Karl Richards, Declan Bolton, Leighton Prichard, Vincent O'Flaherty, Florence Abram (2018). Towards assessing farm-based anaerobic digestate public health risks: comparative investigation with slurry, effect of pasteurisation treatments and use of miniature bioreactors as proxies for pathogen spiking trials. Frontiers in Sustainable Food Systems, 2:41. Doi: 10.3389/fsufs.2018.00041 [Open Access].
3. Stephen Nolan, CE Thorn, SM Ashekuzzaman, I Kavanagh, R Nag, D Bolton, E Cummins, V O'Flaherty, F Abram, K Richards, O Fenton (2020). Land-spreading with co-digested cattle slurry, with or without pasteurisation, as a mitigation strategy against pathogen, nutrient and metal contamination associated with untreated slurry. Science of the Total Environment, 744, 140841, available at https://www.sciencedirect.com/science/article/pii/S0048969720343655
4. Lauren Russell, Paul Whyte, Annetta Zintl, Steve Gordan, Bryan Markey, Theo de Waal, Stephen Nolan, Vincent O'Flaherty, Florence Abram, Karl Richards, Owen Fenton and Declan Bolton (2020). A small study of bacterial contamination of anaerobic digestion materials and survival in different feed stocks. Bioengineering, 7(3), 116, https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7552645/.
5. Lauren Russell, Whyte Paul, Zintl Annetta, Gordon Stephen V., Markey Bryan, de Waal Theo, Nolan Stephen, O'Flaherty Vincent, Abram Florence, Richards Karl, Fenton Owen, Bolton Declan (2022)The Survival of Salmonella Senftenberg, Escherichia coli O157:H7, Listeria monocytogenes, Enterococcus faecalis and Clostridium sporogenes in Sandy and Clay Loam Textured soils when applied in bovine slurry or unpasteurised digestate and the run-off rate for a test bacterium, Listeria innocua, when applied to grass in slurry and digestate. Frontiers in Sustainable Food Systems, 6, URL=https://www.frontiersin.org/article/10.3389/fsufs.2022.806920.
6. Rajat Nag, Agathe Auer, Bryan Markey, Paul Whyte, Stephen Nolan, Vincent O'Flaherty, Lauren Russell, Declan Bolton, Owen Fenton, Karl Richards and Enda Cummins (2019). Anaerobic digestion of agricultural manure and biomass critical indicators of risk and knowledge gaps. Science of the Total Environment, 690 (10), 460-475.
7. Rajat Nag, Agathe Auer, Stephen Nolan, Lauren Russell, Bryan K. Markey, Paul Whyte, Vincent O'Flaherty, Declan Bolton, Owen Fenton, Karl G. Richards, Enda Cummins (2021). Evaluation of pathogen concentration in anaerobic digestate using a predictive modelling approach (ADRISK). Science of the Total Environment, 800 (2021), 149574.
8. Rajat Nag, Russell L, Nolan S, Auer A, Markey BK, Whyte P, O'Flaherty V, Bolton D, Fenton O, Richards KG, Cummins E. (2022).

		Quantitative microbial risk assessment associated with ready-to-eat salads following the application of farmyard manure and slurry or anaerobic digestate to arable lands. Science of the Total Environment. Feb 1;806(Pt 3):151227. Doi: 10.1016/j.scitotenv.2021.151227. Epub 2021 Oct 27. PMID: 34715220. 9. Rajat Nag, Stephen Nolan, Vincent O'Flaherty, Owen Fenton, Karl G Richards, Bryan K Markey, Paul Whyte, Declan Bolton, Enda Cummins (2021). Quantitative microbial human exposure model for faecal indicator bacteria and risk assessment of pathogenic Escherichia coli in surface runoff following application of dairy cattle slurry and co-digestate to grassland. Journal of Environmental Management,299, 113627. Doi: 10.1016/j.jenvman.2021.113627 10. Rajat Nag, Bryan K Markey, Paul Whyte, Vincent O'Flaherty, Declan Bolton, Owen Fenton, Karl G Richards, Enda Cummins (2021) A Bayesian inference approach to quantify average pathogen loads in farmyard manure and slurry using opensource Irish datasets. Science of The Total Environment 786, 147474. Doi: 10.1016/j.scitotenv.2021.147474 11. Rajat Nag, Paul Whyte, Bryan K Markey, Vincent O'Flaherty, Declan Bolton, Owen Fenton, Karl G Richards, Enda Cummins (2020). Ranking hazards pertaining to human health concerns from land application of anaerobic digestate. Science of the Total Environment, 710, 136297. Doi: 10.1016/j.scitotenv.2019.136297 12. Vande Burgt NH, Auer A, Zintl A. 2018. Comparison of in vitro viability methods for Cryptosporidium oocysts. Experimental Parasitology, 187, 30-36.
Peer reviewed conference papers	7	1. Lauren Russell, Karl Richards, Owen Fenton, Annetta Zinc, Bryan Markey, Theo De Waal, Stephen Gordon, Paul Whyte, Florence Abram, Vincent O'Flaherty and Declan Bolton (2019). Bacterial Pathogen Prevalence and Survival During Anaerobic Digestion. Poster presentation at the One Health EJP ASM, Teagasc (Ashtown) Dublin, Ireland, 22nd to 24th May 2019, Abstract Book, page117. 2. Lauren Russell, Paul Whyte and Declan Bolton (2019). Presence and survival of bacterial pathogens during anaerobic digestion and in digestate on farmland. Oral presentation (and second place) at Teagasc Walsh Fellowship Seminar/Regional Competition 2019, held at Teagasc Moorepark on Friday 22 November 2019. Abstract Book, page 28 available athttps://www.teagasc.ie/media/website/publications/2019/Teagasc-WalshFellowship-Seminar-2019.pdf 3. Rajat Nag, BK Markey, P Whyte, V O'Flaherty, D Bolton, O Fenton, KG Richards, E Cummins (2020). A quantitative risk assessment of E. coli O157: H7 on ready to eat foods following the application of biomaterials on land. 11th International Conference on Simulation and Modelling in the Food and Bio-Industry 2020 (FOODSIM'2020). EUROSIS-ETI Ghent, Belgium, 141-144.

		4. Auer A, Vande Burgt N, Abram F, Barry G, Fenton O, Markey B, Nolan S, Richards K, Bolton D, De Waal T, Gordon S, O'Flaherty V, Whyte P, Zintl A. Cryptosporidium in water- No thanks! Joint BSPP, ISP, BAVP and EVPC meeting. Brussels, May 2018. 5. Vande Burgt N, Auer A, Nolan S, Zintl A. Comparison of Cryptosporidium viability assays. Irish Society for Parasitology, Trinity College Dublin, Dublin, June 2016. 6. Auer A, Vande Burgt N, Abram F, Barry G, Fenton O, Markey B, Nolan S, Richards K, Bolton D, De Waal T, Gordon S, O'Flaherty V, Whyte P, Zintl A. Comparison of Nucleic Acid Extraction Methods for Materials Processed in Irish Anaerobic Digestion (AD) Plants. 6th annual DAPI symposium, 15th Jan 2016, Dept Microbiology, Trinity College Dublin. 7. Auer A, Vande Burgt N, Abram F, Barry G, Fenton O, Markey B, Nolan S, Richards K, Bolton D, De Waal T, Gordon S, O'Flaherty V, Whyte P, Zintl A. Survival of Mycobacterium spp. during mesophilic anaerobic digestion. AVTRW, Irish Branch Annual Meeting, Backweston, Kildare 2018.
PhD Theses	4	1. The comparative public and animal health risks associated with spreading anaerobic digestate and slurry on land (Ph.D. Lauren Russell, UCD, 24th March 2022). 2. Potential and optimisation of agriculture-based anaerobic digestion for environmental mitigation of agriculture-associated pollution (Ph.D. Stephen Nolan, NUIG, December 2020). 3. Risk assessment of hazards through aggregate environmental pathways following the application of anaerobic digestate and animal waste to agricultural land (Ph.D. Rajat Nag, UCD, 30 November 2020). 4. Survival of infectious agents during commercial anaerobic digestion processes (Ph.D. Agathe Auer, UCD 2019)
Training courses	1	A Workshop was held in Teagasc Food Research Centre (Ashtown) on Monday 2nd December 2019 (agenda attached) and included presentations by Dr Annetta Zintl (UCD), Dr Stephen Nolan (NUIG), Ms. Lauren Russell (Teagasc), Dr Rajat Nag (UCD) and Dr Declan Bolton (Teagasc) describing the experiments undertaken and the results obtained. The attendees included; Julian Beatty (Nova Q Ltd), Luke Moran (Nova Q Ltd), Noel Gavigan (Irish Bioenergy Association), Morgan Bourke (Steam Bioenergy Limited), Fintan Conway (Executive Secretary, Irish Farmers Association), Percy Foster (Executive, Cré (Composting & Anaerobic Digestion Association of Ireland), Colm Staunton (Director, Halston Environmental & Planning Ltd), Paul Carson (Strategic Power), Jason Hannon (Gas Networks Ireland), Tom Knitter (Veolia Water Ireland), Stephen Hayes (Triskel Green Gas Ltd), James McGreer (NOVA UCD and RGFI), Oisin Doherty (Glenmore Generation), Pauric Tague (Glenomre Estate), Pat McCormack (Ormonde Organics), Robbie (Green Generation Ltd), Tim Gleeson (Teagasc), Karl McDonald (FSAI), Gerard McCutcheon, Regional Pig Advisor (Teagasc), Maeve Henchion (Teagasc), Karl Richards

	(Teagasc), Owen Fenton (Teagasc), Lauren Russell (Teagasc), Barry Caslin (Teagasc), Declan Bolton (Teagasc), Melanie Farrer (DAFM), Justin Byrne (DAFM), Noeleen McDonald (DAFM), Florence Abram (NUIG), Stephen Nolan (NUIG), Annetta Zintl (UCD), Paul White (UCD), Bryan Markey (UCD), Enda Cummins (UCD), Rajat Nag (UCD), Sharon O'Rourke (UCD) & Ajay Menon (UCD).

Intellectual Property

Not applicable.

Summary of other Project Outputs

Project Outputs	Details	Total No.
New Industry Collaborations Developed	Teagasc are now working with Bord Bia and AD bioreactor operators to help develop this green energy sector without incurring any risks for public and animal health.	1
New Technology	The predictive models developed allow AD operators to design their processes to ensure optimal pathogen reduction.	1

Potential Impact related to Policy, Practice and Other Impacts

Impact	Details
Environmental Sustainability	The data and information generated in this project suggests that digestate from animal wastes presents a lower risk than spreading raw slurry on farmland. Moreover, any risk to human and animal health associated with spreading digestate on agricultural land is negligible to low, if the digestate is pasteurised and other precautions such as those set out in relevant legislation, good farming practices, etc. are diligently followed. There is a legal requirement that digestate be pasteurised before land application and two pasteurization processes are available in Ireland including 70°C for 60 minutes, European Commission (Regulation No. 142/2011) and 60°C for 48 hours to be undertake at least twice, as provided by the Irish Department of Agriculture, Food and the Marine (DAFM). Our date and the predictive modelling exercises validates this approach I terms of the requirement to pasteurise and the time-temperature combinations to be used.

Dissemination Activities

Activity	Details
Workshops at which results were presented	A Workshop was held in Teagasc Food Research Centre (Ashtown) on Monday 2nd December 2019 (agenda attached) and included presentations by Dr Annetta Zintl (UCD), Dr Stephen Nolan (NUIG), Ms. Lauren Russell (Teagasc), Dr Rajat Nag (UCD) and Dr Declan Bolton (Teagasc) describing the experiments undertaken and the results obtained. Additional details provided above.
Seminars at which results were presented	The knowledge and data generated in this project were presented at several different events including, the One Health EJP ASM, Teagasc (Ashtown) Dublin, Ireland, 22nd to 24th May 2019, the 11th International Conference on Simulation and Modelling in the Food and Bio-Industry 2020 (FOODSIM'2020). EUROSIS-ETI Ghent, Belgium, the Joint BSPP, ISP, BAVP and EVPC meeting. Brussels, May 2018. The Irish Society for Parasitology, Trinity College Dublin, Dublin, June 2016, the 6th annual DAPI symposium, 15th Jan 2016, Dept Microbiology, Trinity College Dublin and at the AVTRW, Irish Branch Annual Meeting, Backweston, Kildare 2018. More details are provided above.
Other	Information leaflets were disseminated to stakeholders at the outset (introducing the project) and at the end of the project (summarising the key findings).

Knowledge Transfer Activities

Identify knowledge outputs generated during this project.	Knowledge for industry: This project generated knowledge for the Green Energy sector in terms of process optimisation to ensure the elimination of pathogens in the bioreactor. Moreover, predictive models were developed as a tool to manipulate the anaerobic digestion conditions to achieve the same. Knowledge for regulatory: The knowledge on pathogen decreases in the bioreactor and the effectiveness of current pasteurisation validates current policy in terms of land-spreading digestate and the time-temperature conditions required for pasteurisation. Academic knowledge: New insights and data were generated on the survival of bacterial, viral and parasitic pathogens under different fermentation conditions and in different matrices, which add to our current knowledge of these pathogens that are relevant in public health, animal health and environmental protection.
Identify any knowledge transfer activities executed within the project.	The knowledge generated above was transferred directly to our industry and regulatory stakeholders through ongoing communications, a final report and the project workshop. Knowledge was also shared with academia and food safety regulatory personnel through the peer reviewed papers and participation in the seminars, conferences and other events mentioned above. The knowledge presented in posters was discussed with interested parties at dedicated sessions while oral presentations affording an opportunity for discussion and feedback.
List any impacts resulting from the knowledge transferred during the project.	Anaerobic digestion (AD) is a process in which biodegradable materials are decomposed producing biogas, which is used to generate heat, electricity or upgraded to methane for injection into the gas grid. If AD is adopted in a sizable way over the next five years, it will help Ireland reach legally binding greenhouse gas (GHG) emission targets. The provision of knowledge, collated from international publications and generated in the project significantly contributed to allaying concerns with respect to the prevalence and survival of pathogenic bacteria, viruses and parasites via land spreading of AD residues. Results from a Teagasc (Ashtown & Johnstown Castle), NUIG and UCD four-year project have shown that land application of AD residues present less of a risk than the application of cattle slurry. A national framework was delivered for control interventions to aid in the safe application of AD residue to land. Thus, our data provides a green light for AD green energy in Ireland.

Section 3 – Leveraging, Future Strategies & Reference

Leveraging Metrics

Type of Funding	Funding €	Summary
Exchequer National Funding	€1,239,374.43	'Assuring the Chemical and Microbial Safety of Organic Waste Spread on Land in Ireland' [2021R453] funded by DAFM.

Future Strategies

The data generated in this project contributed a report provided by Dr Declan Bolton for Bord Bia on 'The biological safety of digestate from animal wastes and wastewater treatment sludge'. The expertise and knowledge generated in the project has and will continue to provide support for the AD sector as the ambitious plans for several hundred AD facilities are realised over the coming 5-10 years. It is also intended to use the experience and expertise gained to participate in European funded projects, if and when suitable opportunities arise. To date this has included participation in 2 Horizon Europe proposals including: ' the 'A multi-actor knowledge hub and computational infrastructure for fully probabilistic risk assessment and multicriteria decision making to predict and mitigate existing and emerging food safety issues' submitted to call HORIZON-CL6-2021-FARM2FORK-01 in 2021 and 'Food Loss: What is it, why is it there and how can it be measured, reduced and utilized in a sustainable way? [LessFoodLoss]' submitted to call HORIZON-CL6-2024-FARM2FORK: Preventing and reducing food loss and waste to help reach 2030 climate targets.

Project Publications

1. Agathe Auer, Vande Burgt, Nathan; Abram, Florence; Barry, Gerald; Fenton, Owen; Markey, Bryan; Nolan, Stephen; Richards, Karl; Bolton, Declan; De Waal, Theo; Gordon, Stephen; O'Flaherty, Vincent; Whyte, Paul; Zintl, Annetta (2016). "Agricultural anaerobic digestion power plants in Ireland and Germany: policy & practice. Journal of the Science of Food and Agriculture, 97, 3, 719-723.
2. Stephen Nolan, Nicholas Walters, Fiona Brennan, Agathe Auer, Owen Fenton, Karl Richards, Declan Bolton, Leighton Prichard, Vincent O'Flaherty, Florence Abram (2018). Towards assessing farm-based anaerobic digestate public health risks: comparative investigation with slurry, effect of pasteurisation treatments and use of miniature bioreactors as proxies for pathogen spiking trials. Frontiers in Sustainable Food Systems, 2:41. doi: 10.3389/fsufs.2018.00041 [Open Access].
3. Stephen Nolan, CE Thorn, SM Ashekuzzaman, I Kavanagh, R Nag, D Bolton, E Cummins, V O'Flaherty, F Abram, K Richards, O Fenton (2020). Land-spreading with co-digested cattle slurry, with or without pasteurisation, as a mitigation strategy against pathogen, nutrient and metal contamination associated with untreated slurry. Science of the Total Environment, 744, 140841, available at https://www.sciencedirect.com/science/article/pii/S0048969720343655.
4. Lauren Russell, Paul Whyte, Annetta Zintl, Steve Gordan, Bryan Markey, Theo de Waal, Stephen Nolan, Vincent O'Flaherty, Florence Abram, Karl Richards, Owen Fenton and Declan Bolton (2020). A small study of bacterial contamination of anaerobic digestion materials and survival in different feed stocks. Bioengineering, 7(3), 116, https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7552645/.
5. Lauren Russell, Whyte Paul, Zintl Annetta, Gordon Stephen V., Markey Bryan, de Waal Theo, Nolan Stephen, O'Flaherty Vincent, Abram Florence, Richards Karl, Fenton Owen, Bolton Declan (2022)The Survival of Salmonella Senftenberg, Escherichia coli O157:H7, Listeria monocytogenes, Enterococcus faecalis and Clostridium sporogenes in Sandy and Clay Loam Textured soils when applied in bovine slurry or unpasteurised digestate and the run-off rate for a test bacterium, Listeria innocua, when applied to grass in

slurry and digestate. Frontiers in Sustainable Food Systems, 6, URL=https://www.frontiersin.org/article/10.3389/fsufs.2022.806920.

6. Rajat Nag, Agathe Auer, Bryan Markey, Paul Whyte, Stephen Nolan, Vincent O'Flaherty, Lauren Russell, Declan Bolton, Owen Fenton, Karl Richards and Enda Cummins (2019). Anaerobic digestion of agricultural manure and biomass - critical indicators of risk and knowledge gaps. Science of the Total Environment, 690 (10), 460-475.
7. Rajat Nag, Agathe Auer, Stephen Nolan, Lauren Russell, Bryan K. Markey, Paul Whyte, Vincent O'Flaherty, Declan Bolton, Owen Fenton, Karl G. Richards, Enda Cummins (2021). Evaluation of pathogen concentration in anaerobic digestate using a predictive modelling approach (ADRISK). Science of the Total Environment, 800 (2021), 149574.
8. Rajat Nag, Russell L, Nolan S, Auer A, Markey BK, Whyte P, O'Flaherty V, Bolton D, Fenton O, Richards KG, Cummins E. (2022). Quantitative microbial risk assessment associated with ready-to-eat salads following the application of farmyard manure and slurr y or anaerobic digestate to arable lands. Science of the Total Environment. Feb 1;806(Pt 3):151227. doi: 10.1016/j.scitotenv.2021.151227. Epub 2021 Oct 27. PMID: 34715220.
9. Rajat Nag, Stephen Nolan, Vincent O'Flaherty, Owen Fenton, Karl G Richards, Bryan K Markey, Paul Whyte, Declan Bolton, Enda Cummins (2021). Quantitative microbial human exposure model for faecal indicator bacteria and risk assessment of pathogenic Escherichia coli in surface runoff following application of dairy cattle slurry and co-digestate to grassland. Journal of Environmental Management,299, 113627. Doi: 10.1016/j.jenvman.2021.113627.
10. Rajat Nag, Bryan K Markey, Paul Whyte, Vincent O'Flaherty, Declan Bolton, Owen Fenton, Karl G Richards, Enda Cummins. (2021). A Bayesian inference approach to quantify average pathogen loads in farmyard manure and slurry using open-source Irish datasets. Science of The Total Environment 786, 147474. Doi: 10.1016/j.scitotenv.2021.147474.
11. Rajat Nag, Paul Whyte, Bryan K Markey, Vincent O'Flaherty, Declan Bolton, Owen Fenton, Karl G Richards, Enda Cummins (2020). Ranking hazards pertaining to human health concerns from land application of anaerobic digestate. Science of the Total Environment, 710, 136297. Doi: 10.1016/j.scitotenv.2019.136297
12. Vande Burgt NH, Auer A, Zintl A. 2018. Comparison of in vitro viability methods for Cryptosporidium oocysts. Experimental Parasitology, 187, 30-36.
13. Lauren Russell, Karl Richards, Owen Fenton, Annetta Zinc, Bryan Markey, Theo De Waal, Stephen Gordon, Paul Whyte, Florence Abram, Vincent O'Flaherty and Declan Bolton (2019). Bacterial Pathogen Prevalence and Survival During Anaerobic Digestion. Poster presentation at the One Health EJP ASM, Teagasc (Ashtown) Dublin, Ireland, 22nd to 24th May 2019, Abstract Book, page117.
14. Lauren Russell, Paul Whyte and Declan Bolton (2019). Presence and survival of bacterial pathogens during anaerobic digestion and indigestate on farmland. Oral presentation (and second place) at Teagasc Walsh Fellowship Seminar/Regional Competition 2019, held at Teagasc-Moorepark on Friday 22nd November 2019, Abstract Book, page 28 available at https://www.teagasc.ie/media/website/publications/2019/Teagasc-Walsh-Fellowship-Seminar-2019.pdf.
15. Rajat Nag, BK Markey, P Whyte, V O'Flaherty, D Bolton, O Fenton, KG Richards, E Cummins (2020). A quantitative risk assessment of E.coli O157: H7 on ready to eat foods following the application of biomaterials on land. 11th International Conference on Simulation and Modelling in the Food and Bio-Industry 2020 (FOODSIM'2020). EUROSIS-ETI Ghent, Belgium, 141-144.
16. Auer A, Vande Burgt N, Abram F, Barry G, Fenton O, Markey B, Nolan S, Richards K, Bolton D, De Waal T, Gordon S, O'Flaherty V, Whyte P, Zintl A. Cryptosporidium in water- No thanks! Joint BSPP, ISP, BAVP and EVPC meeting. Brussels, May 2018.
17. Vande Burgt N, Auer A, Nolan S, Zintl A. Comparison of Cryptosporidium viability assays. Irish Society for Parasitology, Trinity College Dublin, Dublin, June 2016
18. Auer A, Vande Burgt N, Abram F, Barry G, Fenton O, Markey B, Nolan S, Richards K, Bolton D, De Waal T, Gordon S, O'Flaherty V, Whyte P, Zintl A. Comparison of Nucleic Acid Extraction Methods for Materials

Processed in Irish Anaerobic Digestion (AD) Plants. 6th annual DAPI symposium, 15th Jan 2016, Dept Microbiology, Trinity College Dublin.
19. Auer A, Vande Burgt N, Abram F, Barry G, Fenton O, Markey B, Nolan S, Richards K, Bolton D, De Waal T, Gordon S, O'Flaherty V, WhyteP, Zintl A. Survival of Mycobacterium spp. during mesophilic anaerobic digestion. AVTRW, Irish Branch Annual Meeting, Backweston, Kildare 2018.
20. Bolton et al. (2019) The comparative public and animal health risks associated with spreading Anaerobic Digestate and animal slurry onland: Science, Policy and Practice. Final report published November 2019.