Phase 1 Final report

Project code: NEF 153-1

Enhanced Multiplication, propagation, and establishment technologies combined with new varietal introductions for vegetatively propagated energy crops.

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References in the document are presented in this format [1] with the reference in Appendix 1.

The Biomass Production Challenge.

The Climate Change Committee's strategy projects that 708,000 hectares [1] of energy crops should be established in the UK by 2050 with a target planting of 30,000 ha per year by 2035 [1]. The first commercial crops of *Miscanthus* were planted in England in 1998 and since then the planted area in the UK has only expanded to about 10,000 ha. This underlines the extent of the challenge and raises the question 'why have so few commercial crops been planted?' New Energy Farms EU Ltd (NEF) believes that the primary constraints limiting the expansion of the production areas of high biomass energy crops over the past 20 years have been,

- the absence of new genera, species or variety introductions has not presented any opportunities for yield improvement.
- the method of planting new crops, vegetatively propagated rhizomes, has not changed and is expensive and more difficult than planting annual farm crops.

Phase 1 of this Project addressed these challenges by exploring four innovative approaches.

- **Section 1,** introducing into the UK a wider range of energy grass genera, species, and varieties.
- **Section 2,** delivering new methods of multiplication and establishment of vegetatively propagated energy grass crops, which are CEEDS (Crop Expansion, Encapsulation and Drilling System) and REEDS (Rhizome, Expansion, Enhancement and Delivery system).
- **Section 3,** integrating 'microbials' into CEEDS and REEDS planting propagules, to increase yield, reduce nutrient input requirement and support growth on different land types.
- **Section 4**, extending biomass production, using these new planting technologies and suitable species, on to poor quality, degraded and wetland soils.

1.0 Introduce a wider range of high biomass energy grass genera, species, and varieties to the UK market.

1.1. Variety access.

There is effectively only one variety of *Miscanthus* grown commercially in the whole of the UK, *Miscanthus x giganteus*. Energy crop breeding programmes around the world are virtually untapped by UK and European growers and industries. The UK biomass sector cannot ignore the opportunities which already exist to evaluate novel genera, species and existing (but new to the UK) varieties to potentially improve yields, expand production areas and introduce a wider genetic base into the supply chain. High biomass energy grass crops take up to 15 years to breed so the focus in this project has been on importing existing varieties from public and private breeding programmes globally, the key genera being non-invasive, vegetative, sterile, varieties of *Miscanthus* and other cold tolerant sugarcane types. New variety introductions, with higher yield potential, could be available in three years at a fraction of the cost of further breeding investment. NEF has reviewed energy crop breeding programmes worldwide that could feasibly be producing material suitable for the UK. Programmes outside the UK have already been through more than 10 years of breeding to create 'field ready' varieties to test.

Having only one variety of *Miscanthus* available in the UK for the last 25 years has denied energy crop growers the important yield improvements which are presented each year to arable growers when they consider their annual planting programmes. An analysis of UK *Miscanthus* yield data from UK published sources, (Appendix 2, Fig 1), indicates the extent of the yield 'stagnation'. Defra and Rothamsted long term experiments indicate a plateauing in replicated yield trials of *Miscanthus* at around 15t/ha. The UK *Miscanthus* breeding programme at IBERS, which has not produced any commercial varieties in the last 15 years, has demonstrated a control yield (from *Miscanthus x giganteus*) of 14.97t/ha, but hybrid genotypes in trials have averaged only 10.07t/ha. [redacted]

[redacted] It is well established that replicated plot yields will always exceed field production yields by around 25%, and these three separate analyses indicate an approximate 5t/ha difference from commercial field performance to genetic potential.

This variety initiative, approaching breeders worldwide for access to their varieties, has been very successful. Material Access Agreements (MTA's) are now in place with six breeding companies (four University programmes and two private programmes) in the USA, Canada, and Europe. The agreements cover varieties of *Miscanthus,* Miscane (a *Miscanthus* x sugarcane cross), Energy cane (a cold tolerant sugarcane), *Pennisetum* (often referred to as Napier grass) and [redacted] As a result of NEF's interaction with breeders during Phase 1 the total number of varieties available to the programme in spring 2022 will be 40, if all the material is successfully imported into the UK, via tissue cultured plantlets. Further details of the varieties are in (Appendix 2, Fig 2).

The ability for new genera and species to survive in different countries and climates is an important consideration. Cold tolerance is probably the most important consideration and plant breeders rely on 'hardiness zone' information to guide their interpretation of variety suitability to different geographic locations. Details of Hardiness Zones for the USA and Europe are presented in (Appendix 2, Fig 3). NEF is confident that the *Miscanthus* material in trial, all derived from Hardiness zones in the US, Canada and Europe which are similar or colder than the UK hardiness zone rating, will be acceptable. One of the objectives is to identify material that may be more suited for the north of England and Scotland than the current *Miscanthus x giganteus.* The [redacted] has been bred in the USA in a hardiness zone similar to that of the UK. The Miscane and Energy cane have been selected for their cold tolerance, from within the range of material within the respective breeding programmes. The initial geographic range of the Specimen Trial plots (Section 1.4.1) is designed specifically to address this cold tolerance characteristic. The four varieties of *Pennisetum* will be very interesting. The genus is widely grown in the UK as an ornamental but previously not considered in the UK for biomass production, outside the UK it is used as a biofuel feedstock for anaerobic digestion plants.

The breeding focus in Japan, China and other Asian countries has been more on fundamental genetic exploration of their natural grass species rather than breeding programmes for improvement. However, NEF is confident that it will be offered further varieties during 2022. The number of varieties offered for evaluation during the Phase 1 interaction with Breeders has exceeded expectations. NEF believes this is the result of two factors,

- breeding companies have identified that the BEIS Biomass Feedstocks Innovation Programme signifies an important strategic initiative to develop the UK biomass market and that variety evaluation will take place in the target market.
- there will be access to new planting technologies (CEEDS and REEDS) at an early stage for breeders who collaborate with the Project that will provide them market penetration.

In the UK, *Miscanthus x giganteus*, a sterile hybrid, occupies 95% of the market. Investment in *Miscanthus* breeding over the last 15 years in the US, EU and UK has mostly focussed not on yield, but developing a new type of establishment, using seeded varieties drilled like cereals. However, it has now been concluded that seeded hybrids of *Miscanthus* cannot be direct drilled but must be grown in a greenhouse before being planted as plug plants, resulting in costs equal to current rhizome production. The system for plug plant establishment often requires irrigation or plastic sheet protection, which are not aligned to low-cost establishment on lower grade land. In addition, seeded hybrids have not increased yields. Seeded hybrids are also fertile and could encounter barriers to larger scale planting due to invasive concerns. As a result, the USA's largest *Miscanthus* breeding programme (University of Illinois) has now discontinued its fertile breeding programme, focussing entirely on rhizomatous *Miscanthus*, defining its breeding targets as 'sterile genotypes to avoid potential invasiveness problems'. Breeding programs are now looking for partners to assist them commercialising sterile vegetative varieties they have been developing.

1.2. Plant importation.

Importing plant material into the UK was highlighted by NEF as a potential area of risk. Phytosanitary regulations do not allow the importation into the UK of rhizomes or stems of *Poaceae,* the botanical Family for high biomass energy grass varieties, without a quarantine period, which is usually more than 12 months. This very impractical and expensive route can be replaced by using Tissue Cultured (TC) material as the imported source. Each breeder in the USA had to commission tissue culturing of their varietal material using a TC lab of their choice. NEF commissioned the University of Guelph to tissue culture the plant varieties being made available from the Canadian company. The European varieties were tissue cultured in a UK laboratory where NEF already has an arrangement to generate plant material for multiplication of plantlets for propagation studies. All imported material is directed to this TC laboratory on importation to be 'nurtured' and recover from transportation and is sent to NEF when suitable to be prepared for potting on and field planting. One importation of 6 Varieties from the USA has already totally justified this cautious approach. The overnight flight from the US was followed by 6 days of 'lost in transit' in the UK and it was only the excellent skills at the TC laboratory in the UK that saved the situation. 36 of the 40 energy crop varieties that have been sourced for Phase 2 are now in the UK and being prepared for planting or in the process of import.

A 'Pairing system' exists between NEF and an international freight carrier to ensure that all importations can be speedily processed on arrival at a UK airport. If necessary, this system will be extended to other carriers if the overseas breeding companies chose an alternative carrier to transport their material to the UK. The incident described above was with the 'paired' carrier!

NEF has contacts within the Animal and Plant Health Agency (APHA) and Defra to ensure correct compliance with all Phytosanitary regulations. It should be noted that NEF has raised, on several occasions over the past 5 years with the above two contacts, an apparent inconsistency in importation regulations relating to many high biomass energy grass species. For example, *Miscanthus* plants, declared as ornamental, and destined for the horticultural trade nationwide can be imported as pot grown with no quarantine periods required. However, if *Miscanthus* is declared as ultimately a field grown crop it must be placed in quarantine for over 12 months or imported as a tissue cultured plantlet. This difference in regulation has delayed the large-scale yield evaluation of new varieties within the project by one year.

Variety importation is envisaged as an ongoing arrangement, extending beyond the duration of the project. Regular contact with all breeding companies has been maintained throughout Phase 1 and will continue through Phase 2. The requirement to import material through the tissue culture route does have implications for the planning of variety evaluation trials, a point that is discussed in 1.4 Variety Evaluation section below

1.3. New Variety potential.

NEF has requested performance data from the breeders of *Miscanthus* and the other energy grass crops. Yields of over 34 t/ha for *Miscanthus* (Appendix 2, Fig 4) and 60 t/ha for cold tolerant sugarcane, *Saccharum* (Appendix 2, Fig 5) from hardiness zones potentially suitable to the UK, have been provided. NEF had already collected seven varieties of *Miscanthus* in the UK and were able to evaluate them in Phase 1, in a small trial at Taunton, Somerset which was testing the proposed Specimen Trial protocol prior to any multi-site 2022 plantings. Results of the yield evaluation were that 6 out of the 7 varieties yielded more than the current UK standard (*Miscanthus x giganteus*) which had a yield of 5.78t/ha, very typical for a first year *Miscanthus* crop in the UK when planted from rhizomes (Appendix 2, Fig 6a). One variety, [redacted] programme, imported by NEF in September 2020 as Tissue Cultured (TC) plantlets, produced a Specimen Plot yield of 24.29t/ha, (Appendix 2, Fig 7). Yield increases were correlated to increased stem numbers (Appendix 2, Fig 6b). All the plants in this trial were planted at the same time, having been grown on through the winter in a polytunnel, in 1 litre pots. The only difference was that the [redacted] originated from a tissue cultured plantlet and the other varieties were grown on from rhizomes (Appendix 2, Fig 8). Plants grown from tissue culture can exhibit increased growth when first exposed to normal growing conditions. More rapid growth, more leaves, more stems are all features recorded in the first few months of growth. Comparisons made between plants with different origins (TC, vegetatively propagated, seed grown) must identify the different origins and assess performance accordingly. NEF is removing this 'tissue culture' influence by ensuring that all varieties are grown on for one season, under similar conditions, before entering the main replicated yield trials.

NEF is not proposing to create another 15-year energy grass breeding project in Phase 2. It is demonstrating that the UK biomass feedstock program can harness the existing output from global breeding projects (potentially up to 10 programs) that already have 10 to 15 years of breeding work and field evaluation of new varieties completed. The initial results from Phase 1 support this approach.

The varieties that NEF has obtained have replicated yield data between 20 - 35 t/ha (*Miscanthus*) and other genera with significantly higher yield potential. The NEF initiative, importing and evaluating yield over 3 years, reduces the time and cost for new varieties to be commercially available in the UK from 15 to 3 years. Phase 2 plans to deliver 5 new varieties within 3 years, at a cost of less than £200,000 per variety compared to £3.75 million per variety from the current UK breeding programme, (Appendix 2, Fig 9). This will increase the yield potential of energy grass crops in 2025, the target being between 20 to 25 t/ha. If this project did not proceed, the importation and 3-year evaluation programme would not occur, and the projected yield increases would occur on the original timeline of 2050 as projected by the Committee for Climate Change report [2]. The deliverable metric for this component of the project would be 5 - 10 varieties that have a yield of between 20 to 25 t/ha. These are very exciting yield increases but until the material is tested in the UK, we have no confirmation that these superior levels of performance will be repeated

1.4 Variety evaluation.

Literature reviews conducted during Phase 1 revealed numerous yield trial protocols used around the world to evaluate high biomass energy grass varieties. NEF contacted all the breeders' supplying varieties to the project, outlining the range of options available but also suggesting an agreed way forward. The NEF proposals for Specimen Trials (replicated single plant protocols which are accepted for initial yield evaluation) plus replicated Yield plots with a minimum harvested area of $6m^2$, after edges discarded, have been accepted by all participants.

Four varieties of *Miscanthus*, *Miscanthus x giganteus*, [redacted] and the first US imported variety are currently available to NEF in the UK as rhizomes, the normal way to plant *Miscanthus*. However, all the other varieties being imported into the UK will arrive as tissue cultured plantlets in spring 2022 but some (four) may be delayed until early summer. As described above, the scientific literature clearly indicates that variety evaluations must commence by planting identical material, to ensure that propagation and planting technologies do not influence variety performance. The variety evaluation trials, which are focussed on yield performance, must therefore have uniform planting propagules. The seven *Miscanthus* varieties that NEF already has access to are therefore being tissue cultured so that they enter these comparative trials in the same 'physiological condition' as the candidate varieties. This is an important point and does create a small-time lag in variety evaluation. All varietal material available in 2022 will be planted in Specimen trials, primarily for observation. Further Specimen trials may be necessary each following spring to accommodate new incoming varieties and controls. Any future importations must follow the same tissue culture and potting on procedures so it will also be necessary to repeat 'control' varieties through this route if required.

During 2022 all the tissue cultured material, both UK and imported, is being grown on at the NEF Taunton facility, to prepare small pot grown plants for formal yield trials in spring 2023. This will involve almost 9,000 pot plants being propagated.

1.4.1 Specimen Trials.

Specimen trials is a methodology commonly used by plant breeders to evaluate new varieties. Single plants of each variety are planted in three replicated plots, on a 2m x 2m square plant spacing to allow access for observations and assessments The decision was taken in spring 2021 to plant Specimen trial plots at Taunton, Somerset to test the proposed evaluation protocol prior to any potential 2022 plantings.

The high number of varieties received for evaluation has led to a revision of proposed testing locations. It was proposed in Phase 1 that Specimen Trials would be conducted at two locations, but this has now been increased to five. All varietal material obtained throughout the project will be planted at Cornwall (Wadebridge), Isle of Wight (Sandown), Somerset (Taunton), Lincolnshire (Horncastle) and County Durham (Darlington). The project target is to have the 40 varieties planted by late May 2022. Assessments will consist of

- Plant height to top of stem, number of stems per plant, heading date
- Moisture content (from drying oven sample), corrected dry matter of whole plant (kg)
- Plant tissue analysis - Total Nitrogen and Sulphur with N:S ratio, Phosphorus, Potassium, Calcium, Magnesium, Manganese, Copper, Boron, Iron, Zinc, Molybdenum and Sodium.
- SPAD meter tests - non-destructive measuring for the chlorophyll content of leaves.
- IRGA meter analysis of photosynthesis rates of leaves, which is a proven method to determine adaptability and productivity of Miscanthus [2].

All trials will be managed by NEF staff and located on land rented from collaborating farmers. The geographic spread is designed to ensure good quality information on cold tolerance which, along with yield performance, will be an important screening parameter.

In year 1, due to the comments above about tissue culture influences on initial growth, any biomass measurements which are taken, must be viewed with caution. However, Specimen trial formats are very valuable to gain initial impressions and information on varieties. These Specimen trials will remain in place for the duration of the project (and beyond) to give a very comprehensive overview of variety performance. As all the varieties, including controls, will originate from tissue cultured material it will be possible to conduct a limited comparable yield evaluation from these replicated single plant plots, an evaluation which will be more realistic after each annual harvest. Any further varieties which are received after the 2022 planting window will be planted in Specimen trials the following spring along with selected control varieties.

1.4.2 Replicated Variety Yield Trials.

These planned trials will commence in spring 2023. This will allow one year of data collection from the tissue cultured Specimen plots but more importantly all the planting material will have been grown on as individual plants for one season beyond the time that they were tissue cultured, removing any influence that propagation method may have on performance.
These trials will be at two locations, Somerset (Taunton) and Lincolnshire (Horncastle) and will consist of three replicated plots of each variety, each plot containing 32 plants. At this stage it is planned to include all the varietal material planted in Specimen plots in the 2022 trial series in the spring 2023 trials so there will be 120 yield plots at each location. If any data collected in 2022 clearly indicates that some material is quite unsuitable for UK climatic conditions, then the opportunity is available to discontinue some varieties.

All varieties will be assessed in the same way as in 1.4.1 Specimen Trials. The first harvest from the Variety Yield Trials will be in March/April 2024, the second being before 31st March 2025 as project financing ends on that date. The trials planned for 2022 can be conducted using equipment currently owned by NEF. The two large scale yield locations, each of which could be 120 variety yield plots, plus a considerable number of planting propagule (CEEDS and REEDS) experiments, will require specialised trialling equipment. NEF has reviewed the options and is proposing a combination of purchase and hire for trials equipment for 2023 onwards.

NEF has interacted with two of the Multi-site Demonstrator applicants to understand their specific requirements for demonstration work both in terms of variety access and availability of novel planting propagules from the CEEDS/REEDS protocols. Further discussions will take place with the successful bidder to ensure yield plot evaluations are comparable between projects.

The forward project plan for the planned Phase 2 is to

- secure the import of up to 60 new energy grass varieties to the UK by Spring 2023. Using varieties with prior performance data maximises the chance of success in the UK and helps to develop a model to select the promising varieties for more detailed evaluation.
- establish a replicated, 3-year UK multi-site yield evaluation process for the new varieties to increase the number of non-invasive energy crop varieties available in the UK, to between 5 and 10 new varieties within 3 years, with verified UK yield and quality data.
- target an increase in the genetic yield potential per ha in the UK from 12 – 15t/ha to 20 - 25 t/ ha.

2.0 Delivering new methods of multiplication and establishment of vegetatively propagated high biomass energy grass crops.

New plantings of 30,000 ha/yr of high biomass energy grasses cannot be delivered with the existing propagation stocks and systems in the UK. This project addresses this challenge by developing two technologies, to logistically support large scale planting, and to realise the full genetic potential of new varieties when planted at field scale, via optimised crop establishment methods. Propagation and establishment research on vegetatively propagated crops has been neglected over the last 15 years in the UK, in favour of investment in breeding fertile rather than non-invasive vegetative varieties of high biomass energy grasses. Globally, the vegetatively propagated grasses have higher yield potential. The vegetative propagation route for *Miscanthus*, and other energy grasses has been dismissed, incorrectly, as an option that is not suitable for large scale deployment. The 25 million ha of sugarcane grown globally, a crop that is vegetatively multiplied and replanted about every 3 to 7 years, is testament to the fact that vegetatively propagated crops can be economically multiplied and planted at a large scale. Vegetative propagation is not a barrier to success and large-scale vegetative propagation could be replicated in other perennial grass crops at scale.

NEF’s exploration of new methods of vegetative propagation and establishment began in 2010, focussing initially on *Miscanthus* and *Pennisetum,* as problems with multiplication and establishment hampered further development and uptake. The objective of NEF’s original studies was to create a resilient, reliable, vegetative propagule capable of being planted like a row crop. These propagules could either be small, vigorous rhizomes with several buds or miniature plants, encased in compost and enrobed in wax for easy handling.

2.1 CEEDS patented technology.

Sugarcane was included in the NEF studies in 2012 and was very responsive so the R&D focus moved from European, high biomass, energy grasses, to that crop. The result was small, wax coated vegetative planting propagules, produced by NEF’s patented CEEDS technology, being licenced for production and planting in the Brazilian sugarcane market.

In June 2012 a UK patent application was submitted, followed on 12 June 2013 by a Patent Cooperation Treaty (PCT) application, coded PCT/GB2013/051543. By January 2022, 39 countries have granted patents for CEEDS technology. Phase 1 studies in this BEIS programme have been

strongly influenced by the success of CEEDS technology in the Brazilian and other sugarcane markets. At pre commercial stage, with over 300 hectares planted on mill sites across Brazil, the technology is described as ‘disruptive’. CEEDS technology in Brazil’s 10 million ha sugarcane market, rates as TRL7. It is referred to as ‘disruptive’ technology because it can significantly influence several current activities in the sugarcane supply cane such as the land requirement to produce propagation material and the logistics of planting new sugarcane crops. Sugarcane CEEDS propagules are planted at around 400kg/ha whereas conventional cane stem planting requires between 12t and 20t/ha of stems to be planted to establish a new crop. Mechanised planting CEEDS is three times faster than conventional planting of sugarcane via billets (which are 30cm stem sections).
The cane required for new crop planting means that around 5% of the cropped area must be sacrificed to produce a new crop, allowing for a 5-year replanting schedule. This means that around 5% of the worlds’ sugar cane area is used as new planting material not sugar production. 1,000ha of conventional sugar cane planting material requires between 150 to 250 ha of standing crop to be sacrificed. CEEDS production is conducted in ‘Biofactories’ which are combinations of glasshouses and/or polytunnels and standing areas. As a comparison a sugarcane CEEDS Biofactory would require around 3.5 ha of land, with no existing crop sacrificed to plant 1,000 ha.

CEEDS application to *Miscanthus* and other UK energy crops will offer significant benefits. CEEDS contain multiple buds and roots, they are effectively miniature plants, that generate vigorous growth, images of CEEDS application on sugarcane in Brazil demonstrate this vigour, (Appendix 2, Fig 10). CEEDS has been developed to provide high quality uniform precision planting of perennial grasses, to obtain the maximum yield potential from a variety, and deliver a shorter timeline to peak yield. Green House Gas (GHG) emissions from CEEDS production are projected to be lower than current propagation options for *Miscanthus* (Appendix 2, Fig 11).

Sugar Research Australia (SRA), one of the leading sugarcane research establishments in the world, is currently developing CEEDS for the Australian market under a Development Licence from NEF, where it is at TRL5. It is also TRL5 in the USA where a Development Licence has been granted [redacted]

The sugarcane studies in Brazil, Australia and the USA gave NEF the confidence to revisit in Phase 1 the ‘original’ NEF vegetatively propagated study crops, *Miscanthus* and *Pennisetum,* from 2010 to 2013. Using relevant information from 7 years of NEF research on sugarcane from NEF’s CEEDS protocols, a series of protocols have been created for experimentation through 2022 to 2025 to assess other energy crops responses to a range of multiplication and propagation treatments (CEEDS technology).

Current *Miscanthus* field rhizome production has a multiplication ability between 10 - 20 fold under normal field conditions. One hectare of field multiplication can produce up to 0.5 million rhizomes per year, sufficient to plant 25 ha of new crop. The resulting material can be fully graded to produce new rhizomes for sale or for replanting on the same farm. Crucially, for *Miscanthus x giganteus*, it can be lifted on farm and replanted on the same farm without processing. This option can reduce the cost of establishment to 50% of the current commercial cost. The typical range of establishment costs is between £750 to £2,000 per ha, with a mid-point of around £1,500 per ha. This is replicating what happens in sugarcane globally, where a proportion of existing cropping is used for propagation. Any forward strategy in the UK will include some use of existing crops for rhizome propagation. NEF have evaluated three options for advancing current vegetative propagation of *Miscanthus* (and other energy grasses) for the UK. They are CEEDS, a version 2 of CEEDS and a new version of the current *Miscanthus* rhizome propagation approach, branded REEDS. By adapting the established NEF CEEDS (artificial seeds) technology and REEDS (a 2nd generation *Miscanthus* rhizome production system) for the UK, NEF will develop a platform, using a network of Biofactories, to multiply and plant a range of these high biomass energy crops. These two technologies will support the development of a greatly enhanced UK biomass supply capability.

2.2. CEEDS protocols.

Phase 1 presented the opportunity to undergo a very detailed appraisal of how the protocols developed for CEEDS technology on sugarcane could be modified for use on *Miscanthus* and other high biomass grasses. New NEF studies from July 2021 to January 2022, conducted in the facility at Taunton, have been added to this existing data base to produce a comprehensive series of protocols for implementation from 2022 to 2025, listed below. Initial horticultural work was also completed in Phase 1 to validate application of CEEDS to *Miscanthus* in the UK (Appendix 2, Fig 12). This included work to confirm that the multiplication potential using CEEDS in the UK on *Miscanthus*, could deliver a 50 fold increase in production per m^{-2} of growing space when moving from field production to the intensive horticultural approach of CEEDS (Appendix 2, Fig 13). The deliverable metrics for this component of the project would be CEEDS at the stage of commercial deployment for Biofactories with an output of between 1,000 to 5,000 ha of planting per year.

2.2.1 CEEDS - Intensive node multiplication protocols
2.2.2 CEEDS - Hardening phase
2.2.3 CEEDS - Field delivery
2.2.4 CEEDS - Mechanisation
2.2.5 CEEDS - Process integration
2.2.6 CEEDS - Paludiculture
2.2.7 CEEDS - Version 2 CEEDS process

2.2.1 to 2.2.3 – These are the protocols from the previous CEEDS work on sugarcane project, earlier NEF studies on other grasses and the recent Phase 1 studies which have all helped to develop the CEEDS process for UK *Miscanthus*. During Phase 1 over 100 protocols have been developed for UK energy crops.
2.2.4 – Covers machinery development to mechanise parts of the process specific to the UK crops. 75% of the mechanisation has already been completed by using the outcomes of the sugarcane CEEDS progress in Brazil, reducing the number of new areas required for UK crops. Commencing in 2022 these will be dependent upon the outputs from 2.2.1 and 2.2.2, which provide the specification of plant material that requires mechanical processing.
2.2.5 – Integrates the outputs from 2.2.1 to 2.2.4 into an operating solution, covering validated production protocols and machinery. This will be delivered in the final year of the project.
2.2.6 – Determines if the new CEEDS protocols can be applied to key Paludiculture crops *Phragmites australius, Typha latifolia, Phalaris arundinaceae* and *Molinia* through successful greenhouse and field trials conducted in spring 2023 and 2024.
2.2.7 – will explore whether the CEEDS process for *Miscanthus* can be adjusted more significantly than originally planned from sugarcane to the extent that [redacted]. There are characteristics of Miscanthus that NEF believe are suitable for this adjustment, as a result of a full literature review of germination factors that was undertaken in Phase 1. This would radically reduce the cost of the product and the production area requirement. Potentially the space requirement for CEEDS v2 would be 131 times lower than current rhizome production, and 6 times lower than *Miscanthus* seeded plug production (Appendix 2, Fig 17). Delivery will be in three stages, year 1 a literature review, year 2 protocol evaluation and year 3 a small-scale system to validate production. The Deliverable is to demonstrate to commercial licensees that their Biofactories could be enhanced even further to significantly improve performance with CEEDS v2.

2.3 REEDS technology focusing on rhizomatous high biomass energy grass crops, using spin off technology from CEEDS.

CEEDS technology, which ultimately produces a miniature plant, encased in growing medium and wax coated, is not the correct solution for all the species in this proposal. Some species may respond better to other methods of stimulation and priming during the vegetative propagation process. REEDS technology (Rhizome Expansion, Enhancement & Drilling System) explores other physiological stimulation and priming treatments to improve vegetative propagation techniques. It represents a 2nd generation *Miscanthus* rhizome propagation system, based on NEF's 25 years' experience of the crop. REEDS technology has not been submitted to patent authorities yet.

Two alternative REEDS approaches have already been actively explored by NEF during the development of CEEDS technology, and more recently during Phase 1 and will be explored in more detail in Phase 2. [redacted]

[redacted]

NEF has conducted research protocols for REEDS technology from 2016 to 2021. A total of 54 trials were implemented on a range of *Miscanthus* rhizomes resourced from different parts of England and lifted from the ground at various stages through the 'lifting window' of rhizome processing. This technology is not changing the architecture of the rhizome or plant part (which is the target of CEEDS technology) it is stimulating bud emergence. The field emergence response of 'treated' rhizomes can be recorded as both a higher % emergence of the total number of rhizomes planted and also higher total number of stems, as recorded in a 2019 trial, (Appendix 2, Fig 16). In the trial (Appendix 2, Fig 16) rhizome germination was 32% higher, and 57% more stems were produced as a result of the REEDS treatment. Considerable response differences between rhizome stocks and years have been recorded and future work will seek to understand these different responses and convert to commercial protocols.

The timing of the Phase 1 programme funding did not coincide with the time when rhizome lifting takes place in conventional rhizome production (mid-February to early April), so no further practical studies were possible during Phase 1. However, a more detailed analysis of the data was undertaken during Phase 1 which has allowed more refined protocols to be designed for 2022 to 2025 proposals. REEDS technology, producing more rhizomes of higher quality could upgrade and integrate with existing UK *Miscanthus* rhizome production, processing and planting systems, ensuring that the existing investment in production can be harnessed to provide an early contribution to increase new plantings. The system can also be used by large and small rhizome producers, to improve *Miscanthus* rhizome establishment, which has previously been a barrier.

2.4 REEDS protocols.

[redacted]

[redacted] EF developed the CEEDS and REEDS systems from an understanding of field rhizome production, experience gained from the horticultural production of plug plants and an understanding of tissue culture technologies. The three vegetative propagation options in this project, CEEDS, CEEDS v2 and REEDS represent progressively higher starting TRL levels with less development to commercial scale, but higher rewards in terms of deliverables. The deliverable metrics for this component of the project would be commercial readiness for REEDS to be integrated into existing rhizome processing sites (and increase rhizome bud germination by at least 60%).

2.5. CEEDS and REEDS logistics, economics, and commercialisation

2.5.1 Market opportunity

NEF management have worked in commercial development of energy crops for 25 years in the UK, EU, North and South America. NEF was established (in 2010) to develop new systems for scaling up and establishing energy crops, which resulted in the research for CEEDS and REEDS systems. This experience has provided a solid understanding of the full value chain from breeding to end use conversion. The Climate Change Committee have indicated that energy grass crops planting should be expanded up to the required 30,000 ha per year by 2035. During Phase 1 of the project NEF have analysed the critical factors for creating this step change in energy crop planting. The result of this has been the focus on providing varieties to increase yield and grower income. Secondly, to develop technologies to rapidly scale up high quality plantings that will give growers the confidence to invest in perennial energy crops.

2.5.2 Commercial models for CEEDS and REEDS

During Phase 1 UK options for how best to implement the expansion of new planting technologies and new variety plantings were explored. As a result, NEF plans to develop a licensing model as used by NEF in the Brazilian sugarcane market with end users (mills) as licensees, where it has been successful. Licensing is a low capital model that supports fast scale up supported by licensees, putting them in control of producing their own planting material (CEEDS) and reducing their costs (with only a NEF royalty to pay, not a commercial margin to a distributor). In the UK the potential licensees could be seed companies or end users. [redacted]

[redacted]

The forward project plan is as follows,
-Develop the NEF REEDS system to the point of commercial use, with protocol, equipment, GHG savings, and a commercial model as a version 2.0 of the current *Miscanthus* rhizome propagation.
- Enhance the germination efficiency and vigour of *Miscanthus* rhizomes and reduce the production footprint required for rhizome propagation.
- Develop the NEF CEEDS system to the point of commercial use for energy grasses, with protocols, equipment, GHG savings and commercial model (for CEEDS Biofactories). This will be a step change delivery system for scaling energy grasses in the UK, that is a common platform for all energy grass crops including *Miscanthus*, up to 30,000 ha per year, to be ready for commercial implementation by 2025.
-Provide increases in efficiency, logistical capability, and reductions in the cost and GHG balance of establishing new energy crops (Appendix 2, Fig 11).
- Deliver these benefits through the commercial development of NEF CEEDS and REEDS technologies, allowing the maximum genetic yield potential of new varieties to be realised at field scale.

3.0 Integrating 'microbials' into CEEDS and REEDS planting propagules

3.1 The opportunity

Capturing synergies that exist between crops and endophytes, other beneficial fungi, and bacteria, is a rapidly developing technical area in world agriculture. It has been demonstrated that members of the Family *Poaceae* can be very responsive to bacterial and fungal stimulation. CEEDS and REEDS technologies also present a route to deliver microbials such as endophytes, and other beneficial fungi and bacteria, to planting propagules during the automated production processes.

Microbials are also referred to as 'biologicals' and are being increasingly used in many important crops worldwide. The market for Rhizobium and Mycorrhiza is already considered to be just under $1 billion. New Zealand (NZ) science organisations have developed significant expertise in working with endophytes, predominately through work on Epichloe type endophytes in perennial forage grasses. This expertise has been extended to work on a broad range of crops with a range of endophyte species including work on sugarcane and *Miscanthus*. The relationship between NEF and NZ researchers was developed in 2017 when NEF visited two endophyte, fungi & bacteria Research programmes.

In Phase 1 NEF commissioned two short Reviews, undertaken by Nick Pyke MSc, of current NZ/Australian activity in endophytes and other 'microbials'. Two primary research programmes are at AgResearch and Lincoln University.

AgResearch, led by Dr Stuart Card, undertook work for Sugar Research Australia (SRA) with a focus on entomopathogenic endophytes for sugarcane cane grub and soldier fly. (Note SRA is the sugarcane research organisation licenced with CEEDS technology, mentioned in 2.1)
Lincoln University, led by Professor Travis Glare (Sadly, Prof Steve Wratten, who previously led this research programme, died in 2021) has focused on isolates and mixtures of Trichoderma with *Miscanthus*. Their important conclusions are that,

- Endophytes generally need to be selected in the environment or circumstances where they will be ‘operating’ commercially.
- endophytes that have their whole life cycle in the host are more likely to be transferrable between locations, an important consideration for CEEDS/REEDS studies.
- mixtures of isolates appear to be the most beneficial approach.

The reviews highlighted a wide range of benefits from microbials including growth promotion, nitrogen fixation, disease control, insect control, stress (drought management) and tolerance of heavy metals. There is also clear evidence that some of the most productive microbial associations with plants are those already occurring in nature.
NEF is proposing a research programme specifically targeted at identifying microbials which currently have associations with *Miscanthus* crops in the field. It will source UK *Miscanthus* microbials, explore availability of suitable microbials for importation and develop laboratory, greenhouse, and field trials. The aim will be to develop a protocol to integrate microbials into CEEDS by 2025. It will be led by NEF’s research consultant, Nick Pyke who will coordinate the information from NZ researchers and the Research contract that NEF has agreed with Professor Bending (Warwick University).
[redacted]

3.2 identifying and isolating microbials.

[redacted]

The studies will be laboratory and polytunnel based exploring the interactions between the localised fungal populations and their interactions with other sources of field grown rhizomes which may have established other fungal relationships. NEF have agreed a Research Contract with Professor Gary Bending, School of Life Sciences, University of Warwick, an environmental scientist whose research focuses on the structure and ecosystem functions of microbial communities. The Warwick University team will determine if beneficial microbials can be isolated from existing *Miscanthus* crops and identified by DNA extraction and sequencing. They will also provide comparisons of fungal community diversity and composition, the presence of taxa of interest (pathogens, mycorrhizal fungi) and any fungi that are of interest from the NZ microbial work.

This area has the potential to reduce the GHG balance of production, by increasing biomass production with no additional fossil fuel fertilisers. This is TRL 5, as most of the microbials have already been demonstrated to have a positive impact at greenhouse trial level. NEF initially considered, in Phase 1, incorporating other types of plant stimulants in this review. However, many of these amino acids, seaweed extracts, bioactive peptides, carbohydrates, Humic and Fulvic Acids, although occupying a $2 billion market, have limited technical support, and it was considered that comparisons of this nature are not suited to the Phase 2 project.
The project plan is to source beneficial biological products from the UK and outside the UK, that have proven potential to benefit energy grass crop production by enhancing nutrition and pest protection and expanding soil type production areas. To then test the performance of biological

products under greenhouse and field conditions. Then finally to integrate effective products into the manufacture of either CEEDS or REEDS to provide a field delivery system.

4.0 Extend biomass production by using new planting technologies and suitable species, on contaminated and wetland soils

Historically in the UK biomass production has been focused on low grade arable land but excluded long term grassland. It is only recently that attention has focussed on the possibilities presented for biomass production on the considerable areas of wetlands and metal contaminated soils.

4.1. Wetlands.

The Government plans to restore 280,000 ha of peatland by 2050 based on the England Peat Action Plan [4] and the Net Zero Strategy [5]. Full restoration may not be possible on all of this lowland peat so management regimes including re wetting and paludiculture, which could involve biomass production, could be put in place on this land.
During Phase 1 NEF interacted with contacts in Natural England, DEFRA, The Broads Authority and The Lowland Agricultural Peat Task Force, to develop NEF's Phase 2 strategy. Reducing the high emission levels that are released from drained and eroded peat areas is a major target in many countries. NEF's innovative planting technologies, being used to plant *Miscanthus, Phragmites, Typha, Molinia* and *Phalaris* on regenerated peatlands could see currently unproductive areas of land being reclaimed for biomass production. During Phase 1, NEF agreed field trial's locations from 2022 onwards, using CEEDS and REEDS technologies, at Horsey Fen, Norfolk (Broads Authority) and the Greenhouse Gases Removal Demonstrator (GGRD) wetland site, Yorkshire.
Some wetland areas are also acting as 'sinks', storing phosphate from surrounding agricultural land at levels which are now unacceptable. It is suggested that high biomass plants could be used to extract phosphate from wetlands. This phytoremediation will be explored in a Research Contract (see 5.2) with Professor Sean Comber, University of Plymouth. This third site is on the Somerset levels, also in collaboration with Natural England. This work is compatible with 3.0 where beneficial microbes that solubilise phosphate could be a target mode of action. NEF have obtained stock plants of the 5 genera and have been exploring CEEDS and REEDS protocols to determine their suitability to these wetland planting opportunities. Initial studies on the detailed plant architecture and growth habits of these genera began in Phase 1. It was concluded in Phase 1 that NEF can develop CEEDS and REEDS protocols for these target paludiculture crops.

4.2. Metal contaminated soils.

Just under 2% of UK land area is classed as contaminated, with 60% being metal contamination. Spoil heaps, from mining activities and other degraded soils, cover considerable areas and currently there are few methods of large-scale remediation. CEEDS technology plus energy crops could be one useful phytoremediation tool.
In 2019 NEF conducted a small trial in collaboration with a company testing a remediation product, potentially suitable for counteracting the detrimental effects of metal contamination in soil from mining spoil heaps. The results, measured in terms of *Miscanthus* survival and growth, were encouraging. A Research Contract has been agreed with Professor Sean Comber, Professor of Environmental Chemistry, School of Geography, Earth and Environmental Sciences, University of Plymouth, to provide analytical and academic support to NEF to conduct experiments on these soils. The Research Contract with NEF involves three locations, two mining operations and one paludiculture site (5.1 Somerset Levels). The studies will involve sampling and analysis of soils at the two metal contaminated sites and the high phosphate wetland location, sampling, and analysis of up to 3 plant species for nutrient and metals uptake and collaboration with the design and soil/plant sampling from NEF pot trials. The Plymouth team will bring a great deal of academic and practical experience of dealing with contaminated soils to the project.

The performance of different genera, new planting technologies and remediation chemistry will be evaluated. These will begin in spring 2023 onwards, with control greenhouse plantings in 2022. Work in 3.0 may identify beneficial microbes which aid phytoremediation by energy crops of heavy metal contaminated soils.

NEF already have a contact in Devon where the performance of *Miscanthus* was tested on mine tailings from a Lead and Baryte (Barium) mine. This location has been revisited and arrangements are now in place to begin planting small areas of 'spoil' and on catchment areas associated with different water decontamination treatments over the next three seasons. This will commence on a small scale in spring 2022. [redacted]

Material for planting trials at these two mine locations will not be available in large amounts until spring 2023 but it is hoped to conduct exploratory planting of conventional material in 2022. The feasibility of growing biomass on regenerated wetland and metal contaminated soils is now a serious possibility and the concept of NEF planting technologies assisting this progress is attracting significant attention. The current rating for these opportunities is TRL 4 but progress in CEEDS and REEDS protocols could raise this to TRL 5 quite rapidly.

4.3 Commercial opportunities

CEEDS and REEDS technologies are planned as common propagation platforms for a range of energy crops. In addition, NEF plan to develop protocols for key genera involved in soil regeneration and wetland/Paludiculture situations. These planting environments are more challenging to automate but if they can be established with new crops they could make an essential contribution to UK domestic feedstocks, and carbon sequestration.

NEF plan to develop protocols for these crops, so existing Biofactories can add them as additional supply products or have dedicated Biofactories created just for these crops. NEF have completed initial evaluation work in Stage 1 of this project and established that key Paludiculture crops appear suited to CEEDS or REEDS technologies. The smaller planting propagules could allow systems to be developed to plant these more challenging sites either using lightweight planters, floating planters, robotic planters or even drones (as used for remote tree reseeding).

The project plan is to screen the diverse crops required for expansion on wetlands and rewetted lowland crop areas and contaminated soils and develop commercial propagation systems with CEEDS or REEDS as appropriate, to provide an option that does not currently exist to deliver planting material and use the propagation platforms developed in the project to develop a scalable system via CEEDS to support large scale planting of metal contaminated soils and wetland areas.

5.0 Overall commercial model.

NEF plan a commercial licensing model, providing customers two propagation options CEEDS and REEDS (via licensing), plus layering on top of this the supply of leading new varieties and biological products. This combined platform can then be applied to support the large-scale expansion of bioenergy feedstocks on arable land, contaminated land and paludiculture areas.

During Phase 1 NEF established that a significant resource exists of new energy crop varieties that can be imported to the UK, with the potential to increase yield by over 60%. During Phase 2 the project plans to generate yield and performance data so licensees have performance data to make commercial decisions to invest and scale up these varieties without delay. The NEF financial model is to supply these new varieties with a small royalty paid by the growers, based on a one-off payment at planting integrated into the CEEDS price (a common model for other crops [redacted] Any variety royalty would be modelled to ensure there is a Net Present Value (NPV) benefit to the grower, in terms of increased yield, drought or frost tolerance.

This royalty will fund ongoing *Miscanthus* breeding and is essential so that the development of new varieties going forward is funded by the market, like other crops. NEF have already scaled up new varieties of *Miscanthus* in the US and Canada via conventional rhizome propagation, and paid royalties to breeders, which we believe is a first. The CEEDS platform also provides a unique ability to effectively incorporate biological products into field delivery, as opposed to the more unreliable option of field application after planting. During Phase 1 NEF have established that there is an under exploited resource of biological products, to provide nutrition, protection, and tolerance (such as on contaminated land) for energy crops. During Phase 2 these will be imported or produced in the UK and tested to determine a protocol for integration into CEEDS, and establish which products provide economic benefits. Licensees would then have the ability to enhance the production of each variety, and/or widen the production area, such as supporting the growing of energy crops on contaminated land. Royalties for biologicals would be generated in the same way as for varieties but will be based on a cost per CEEDS unit for the licensee. Some biological products will also be able to be incorporated into REEDS products. Microbials are not suited to direct agricultural field application, through expense and lack of targeting. The CEEDS process presents horticultural opportunities to introduce microbes to the host plant during manufacture and become endophytic early in the plant development.

During Phase 2 NEF will develop a package of information for due diligence questions from prospective licensees. These will cover technical performance, site CAPEX, operating expenses, and licensee technology transfer. An abbreviated version could be used for licensee marketing. NEF has a deep understanding of potential partners in the UK, and in other export markets. In Brazil NEF have already commenced this process, so have experience to leverage into UK commercialisation. NEF have experience in Brazil, the US and Australia licencing CEEDS technology for sugarcane, using models that will be adapted for the UK market. Post licence agreement, NEF have experience in technology transfer, and will establish a full program of licensee training and ongoing support. Royalty revenue will fund ongoing CEEDS and REEDS development work.

[redacted]

[redacted]

6.0 The implications for biomass supply.

The CCC suggests at least a 30-fold increase in area of biomass feedstock crops in the UK, from the 10,000 ha currently planted to a step change of 30,000 new hectares each year. End use demand is outside the scope of this study but is planned to rise with Net Zero policy to encourage planting. After this, the speed of uptake is dependent on how many landowners make the decision to plant energy crops over other competitive land use options. NEF management have had direct interaction with landowners planting energy grasses for the last 22 years. This project uses that knowledge to provide new innovations to enable the supply to be scaled up as required.

The CCC strategy projects 708,000 ha is available for biomass feedstock in the UK, and 30,000 ha per year of new planting is required by 2035 at a modelled yield of 12t/ha for *Miscanthus* currently, increasing to 20t/ha by 2050 [1]. At 12t/ha, energy grass crops provide a reasonable return for landowners, but not one that leads to a rush of new planting. This is reflected in the accumulated level of UK plantings over the last 15 years, which does not exceed 10,000 ha for *Miscanthus*. This has been delivered using a non-improved, sterile vegetative *Miscanthus* cultivar. Increasing yield is the main agronomic factor that could increase the economic value to farmers, being more important than reducing the cost of establishment.

Investment in *Miscanthus* breeding over the last 15 years in the US and Europe has focussed on developing a different form of establishment, via direct drilled seed to reduce the establishment cost. Yield increases and field establishment cost reductions have not been delivered as projected. *Miscanthus* like other perennial crops takes up to 15 years to breed new cultivars. The UK has one internal programme, started in 2005, that has 7 cultivars at registration stage, but none released commercially. Average yield from trials of these new seeded cultivars is less than the 12t/ha policy baseline yield. In addition, seed planted *Miscanthus* is fertile, and will pose an invasive risk, if these new seeded cultivars are planted commercially in the future. Increasing the yield of *Miscanthus* and other energy grasses to 20t/ha or higher, before 2050 is a highly significant CCC requirement in delivering the required level of feedstocks.

During the Phase 1 project NEF investigated how to harness the potential of global breeding programmes of energy grasses, to remove the reliance on one UK *Miscanthus* seeded breeding programme. This study has now made available 40 cultivars of *Miscanthus* and other energy grasses, which have high yield potential and are from comparable hardiness zones to the UK, Yield performance data accompanying these new varieties from their source countries is encouraging. The NEF Phase 2 application project aims to deliver 5 new cultivars within 3 years, at a cost of £200,000 per cultivar, compared to £3.75 million from the current UK breeding programme, (Appendix 2, Fig 9). This should bring forward the yield increase potential of energy grass crops to 2025, onwards, with target yields of between 20 to 25 t/ha. If this project did not proceed to Phase 2, the importation and 3-year evaluation program would not occur at anything like the scale in this project and the projected yield increases would occur on the original timeline.

In Phase 1 NEF explored the economic impact of improved yield performance compared to other potential improvements such as establishment costs being reduced. At current biomass prices (£75/t ex farm 15% moisture content), increasing the yield from 12 to 20t/ha increases farmer NPV/ha from £4331 to £11,174 (via conventional rhizome establishment cost at £1580/ha). Yield improvement raises NPV more than that achieved by lowering establishment costs. Seeded plugs with 30% lower establishment cost but 15 t/ha yield have an NPV of £8,252 per ha (Appendix 2, Fig 21). Phase 2 of this project will target significant establishment cost reductions via CEEDS and REEDS of over 30% and potentially to 50%.

After yield, the next priority are systems to scale up UK energy crop planting, and make it as easy as planting a conventional arable crop. This project plans to develop two propagation systems to deliver that scale up. REEDS will integrate into existing and new rhizome supply sites to enhance rhizome quality, reduce the production footprint and cost, to an expected supply capability of 6,000 ha per year. REEDS is important to integrate with the existing Miscanthus supply capability that already exists in the UK. The more advanced CEEDS system plans to develop an additional system that can implement a network of CEEDS biofactories, small intensive horticultural units, each planned to generate a minimum of 2,500 ha of planting capacity per year. 10 CEEDS biofactories are planned to be examined in Phase 2, potentially co-located at AD plants, to deliver 25,000 ha of energy crop planting, which would reduce the land area required by 17-fold compared to conventional rhizome propagation. CEEDS technology whilst reducing establishment costs, also provides benefits of automatic drills, minimum tillage, and vigorous planting material (Appendix 2, Fig 10). This network of sites will bulk up and supply new varieties and integrate beneficial biologicals into the REEDS and CEEDS product, with the combined aim of delivering a planting system at scale that gets the maximum genetic potential from a variety, for establishment on arable, degraded and even contaminated land. The network will also have the additional capability to propagate new crops for paludiculture and wetlands, where no large-scale capability currently exists.

Yield increases make energy crop growing more financially attractive to growers and should increase uptake. Planting 30,000 ha/yr at 20t/ha (compared to 12t/ha) produces 67% (240,000 tonnes) more biomass per year or requires 40% less land for the current production level. This project plans to deliver higher yielding material much sooner than predicted, 2025 rather than 2050. It will increase planting uptake, and increase the land use efficiency in the UK, which is a precious resource.

This also has a positive impact on later stages of the biomass supply chain. Production of liquid biofuels for the Renewable Transport Fuel Obligation (RTFO) or Sustainable Aviation Fuel (SAF) market has not previously been considered in the UK as the sites require large volumes of feedstock, typically (around 250,000 t/yr) 5 times the size of a current UK electricity biomass plant intake. The higher yield deliverables from this proposed project would reduce the footprint of a 250,000 tonne project by 52% from over 20,800 ha to 10,000ha.

In 2.5.2 reference is made to 579 AD plants in the UK. Maize, an important feedstock for AD plants, is grown on good quality arable land. In June 2020 the area of maize grown for AD in England was 75,000ha. This is an increase of 12% compared to 2019 and equates to 34% of the total maize area in 2020 and 1.3% of the total arable area [3]. It is totally feasible for some high biomass energy grasses to be grown on low grade land for AD end use thereby releasing the higher quality land for food production.

7.0 Project team skills

Responsibilities listed below, are aligned to the workplans listed in section 8.

7.1 NEF EU Ltd

Dr Paul Carver and Dr Mike Carver have been involved in the biomass industry, in the UK and overseas, since 1995 and 2001 respectively and both bring research and commercial experience to the project.

Dr Paul Carver, a joint owner and CEO of New Energy Farms EU Ltd (NEF), invented the patented CEEDS technology and has seven years' experience of developing the technology on sugarcane in Brazil, Australia, and the USA. Involvement in numerous large scale Miscanthus projects in the USA, Canada, Poland, and Russia has added a level of commercial experience which is probably greater than any other scientist in the UK biomass industry. His doctorate addressed the Physiology of Miscanthus. He will be the LEAD for Work plan 2 – Develop CEEDS and REEDS propagation systems and Work plan 5 Commercial model.

Dr Mike Carver has spent his whole career in crop variety and agronomic field trialling and providing technology transfer to the arable industry. As joint owner of New Energy Farms EU Ltd and NEF Technical Manager since 2014 he has considerable experience in the biomass sector being Chairman of a biomass company and three regional Biomass Producer Groups, for 4 years. He was the Industrial Mentor for TSEC Biosys 'A whole system approach to Bioenergy Supply and Demand' which ran from 2005 to 2010. (NERC funded). His doctorate addressed seed physiology. He will be the Lead for Work plan 1

Reg Embleton BSc is a NEF Technical Officer who qualified with an Honours degree in Geology but has sought a career in a more environmentally focussed industry. Reg joined NEF at the start of the Phase 1 project and has developed Miscanthus experience to build on his background in soils and geology. He will be the lead for Work plan 4 – Extend biomass production to wetlands and contaminated soils

Nick Pyke MSc, Member of the New Zealand Order of Merit (MNZM), Director of Ag Innovate, New Zealand. Nick has over 40 years' experience leading annual and perennial crop research programmes, research organisations and research funding allocation. CEO of the Foundation for Arable Research (FAR), the equivalent of AHDB in the UK, from 1997 to 2018. In 2017 Nick was awarded the NZ Order of Merit for services to the arable industry, in the Queen's Birthday Honours List. Nick has developed the strategy during Phase 1 for sourcing and integrating biologicals. He will lead Workplan 3 Improve energy grass performance with microbials.

Tina Jansen (project management) has been responsible for coordinating activities in several large US and Canadian Government projects on Miscanthus since 2011 and she will bring this valuable experience to this project. Her primary roles will be ensuring data collection and reporting schedules are correctly managed.

Jane Fernanda Lopes (Researcher) joined NEF in 2021, in Phase 1, to assist with the transfer of the CEEDS Brazil sugarcane project data into research protocols for other vegetatively propagated crops and to coordinate UK data collection. She is UK based and will provide research support for chemicals, technical products, equipment, and knowledge transfer from sugarcane.

NEF will recruit six technical Officers to work in the areas of field trials management and CEEDS/REEDS development programmes. These will be ideally graduates in these areas, with agricultural or horticultural backgrounds.

7.2 Subcontractors

Miscanthus Nurseries Ltd (MNL). This company is a Grower Cooperative, it is the largest producer of Miscanthus rhizomes in the UK and also holds many end use contracts for Miscanthus cane which it manages for its members. Based at Taunton, it provides NEF with an office, laboratory, and access to trial areas on the farm. MNL also supplies labour to NEF, either sharing senior staff time or the provision of casual labour from its recruitment contacts. **Mike Cooper,** the Managing Director of MNL, is considered by many in the industry to be the leading authority on the commercial aspects of Miscanthus production. Mike has worked in the Miscanthus industry since 1998 and prior to that he was involved in the wider farm supply industry. MNL will dedicate resources and staff to ensure full support to NEF using Phase 2.

Ed Whittles FdSc is an MNL employee seconded to NEF. Ed has an NDA (Triple Distinction) from Kingston Maurward College, Dorchester and a FdSc in Agriculture from Harper Adams University College, Shropshire. Ed worked for three and a half years for Eurofins Agroscience Services as a Research and Development Agronomist responsible for conducting field studies on agricultural crops so he brings valuable experience to the project. Ed will be involved in the Specimen and Variety trial programmes.

Richard Fenwick, Managing Director and Owner of the company that specialises in micropropagation research and development. Lecturer at Askham Bryan College and Ampleforth College and created Plantlet Culture to manufacture micropropagation kits within the UK for Schools, Colleges & Universities. Richard is responsible for all NEF tissue culture work in the UK, and liaison with overseas breeders providing TC samples to NEF. He will also be the main subcontractor to assist NEF in developing the CEEDS v2 project for Work Package 2. This program requires tissue culture skills to reduce the footprint of CEEDS to an even more intensive system. Richard has committed significant time to dedicate work to this project.

Professor Gary Bending, School of Life Sciences, University of Warwick, is an environmental scientist whose research focuses on the structure and ecosystem functions of microbial communities. He is a member of the BBSRC Pool of Experts and NERC Peer Review College and is also Associate Editor of Frontiers in Environmental Science. In the Research Contract with Warwick University NEF will benefit from his knowledge and the facilities provided by his team. NEF have secured access, through MNL, to 20 commercial crops of Miscanthus including some of the very first fields of commercial Miscanthus production in the UK dating back to 2001. This provides the Warwick research team with the invaluable access to developing an understanding of how microbial populations (endophytes, rhizobium etc) may build up over time on Miscanthus root systems. They will extract DNA, perform sequencing, and provide comparisons of fungal community diversity and composition, and the presence of taxa of interest (pathogens, mycorrhizal fungi and any fungi that are of interest from the NZ microbial work)

Professor Sean Comber Professor of Environmental Chemistry, School of Geography, Earth, and Environmental Sciences (Faculty of Science and Engineering), University of Plymouth. Professor Comber is responsible for the Research Contract with NEF on two mining locations and one paludiculture site (Somerset Levels) which involve sampling and analysis of soils at two metal contaminated sites and the high phosphate wetland location, sampling, and analysis of up to 3 plant species for nutrient and metals uptake and collaboration with the design and soil/plant sampling from NEF pot trials. The Plymouth team will bring a great deal of academic and practical experience of dealing with contaminated soils to the project.

Mechanical Botanical Limited, Mike Berry Managing Director. This company is probably the largest in the UK providing practical, ergonomic solutions for production systems on Nurseries and other horticultural enterprises. The company has already provided design and mechanical development inputs for the NEF sugarcane project in Brazil. They will be supporting NEF during Phase 2 design, equipment sourcing and development planning of the CEEDS/REEDS production lines. It is planned to access up to three types of engineering skills Design, Electrical and Mechanical during the duration of the project.

[redacted] has been involved in regenerating the spoil heaps from this mine for many years mainly from his own private resources. Small University projects and some development work by British Coal have taken place on the site and [redacted] will be providing his experience and land access for planting trials in the Plymouth University Research contract.

WH Loxton Ltd. Hugh Loxton is a Miscanthus farmer and agricultural engineer. He designed the first automatic Miscanthus planters, 20 years ago that are still used today. Hugh has been involved in designing new planter systems for Miscanthus, for which he is considered an expert. He will be designing and building a UK CEEDS planter for the project.

7.3 Job creation. Phase 2 funding will support NEF creating 6 new fulltime positions, of Trials Officers based at Taunton (currently 2 part time), plus additional part time support for the two Northern variety trial locations. All roles will be in rural areas where job opportunities are scarcer. There will also be 10 part time positions created during Phase 2 for manual operations during peak trial work each spring, planting and harvesting activities. The project will also support existing sub-contractors staff job retention and create some additional employment opportunities. Regional and rural economies will be supported by these employment opportunities created during the project. On project completion there are several clear opportunities for further job creation. The establishment of multiple CEEDS and REEDS processing biofactories are one example. [redacted]

The project is also supporting two UK universities. Phytoremediation and biomass production on contaminated soils is a significant green technology initiative being addressed in the Plymouth University Research Contract. The field work will be conducted by NEF staff with analytical support and academic guidance provided by the University There is a budget proposal for additional local part time staff at key activity points. This arrangement also applies to the wetland regeneration study in the Plymouth University contract. University of Warwick are providing analytical services for DNA analysis and sequencing of the microbial groups plus academic support. NEF will provide the staff for the field work and Polytunnel studies within the Technical Officer team. Both Plymouth and Warwick have indicated they will require additional inhouse support to be recruited for these contracts.

8.0 Project management and reporting

The planned Phase 2 project has five Work Plans

Work plan 1 – Import and test new energy grasses.

Work plan 2 – Develop CEEDS and REEDS propagation systems.
Work plan 3 – Improve energy grass performance using microbials.
Work plan 4 – Extend biomass production to wetlands and contaminated soils.
Work plan 5 - Commercial model.

The enclosed project Gantt chart (Appendix 2, Fig 22) outlines the planned overall project delivery plan and the 48 milestones proposed for Phase 2 with work durations and completion dates. The Gantt chart also covers the plan for producing reports for the planned monthly update, quarterly report and 6 monthly stage gate reviews. All NEF data is managed through the Microsoft Teams platform. All work package documentation, managed as a sub project in Teams, with be stored with Cloud backup. NEF will establish a reporting area, accessible by BEIS project management, with all reports supplied via this format in addition to email supply. The project management team have extensive experience reporting on large scale multiyear projects. Risks for the project have been identified and a plan enclosed for how these will be managed (Appendix 2, Fig 23). The project will be managed by a Board consisting of Dr Paul Carver (Project lead, and responsibility for WP 2, and 5), Dr Mike Carver (WP 1), Nick Pyke (WP 3) and Reg Embleton (WP 4) and Tina Janzen. They will be responsible for delivery of the work packages, internal staff, and subcontractors in these respective areas. The project will be financially operated with management accounts provided by current NEF accounting staff to ensure financial management and oversight of the project. The progress of the project against milestones will be reported at NEF monthly internal meetings, in addition to project monthly meetings (Appendix 2, Fig 22).

9.0 Appendices

Appendix 1 - References

[1] Policies for the sixth Carbon Budget and Net Zero. Climate Change Committee, December 2020.

[2] Does greater leaf-level photosynthesis explain the larger solar energy conversion efficiency of Miscanthus relative to switchgrass? F. G. Dohleman, E. A. Heaton,A. D. B. Leakey,S. P. Long. Plant Cell and Environment. Volume 32, Issue1 1, November 2009. Pages 1525-1537.

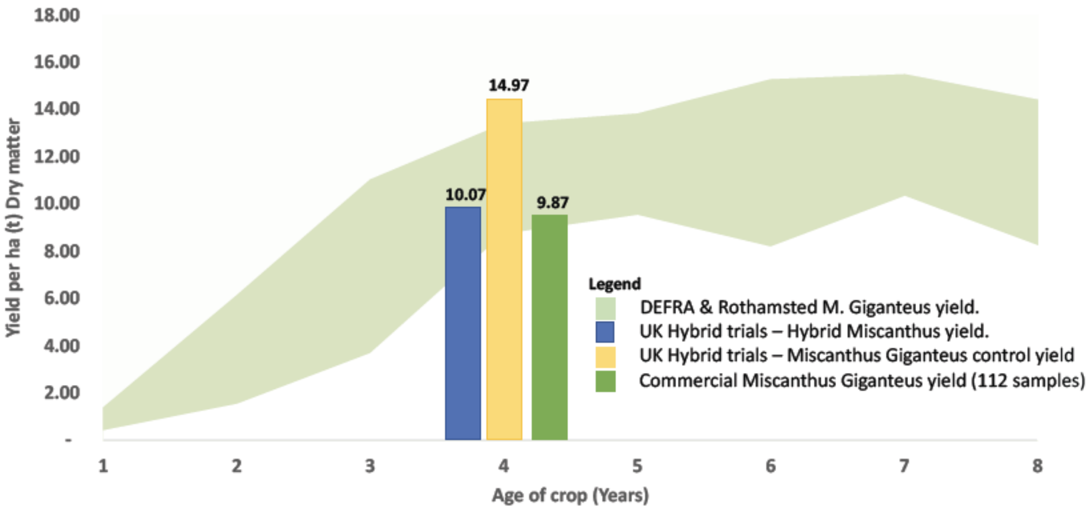
[3] https://www.gov.uk/government/statistics/area-of-crops-grown-for-bioenergy-in-england-and-the-uk-2008-2020/summary

[4] https://www.gov.uk/government/publications/england-peat-action-plan

[5] https://www.gov.uk/government/publications/net-zero-strategy

Appendix 2 – Figures

Fig 1. Yield performance of *Miscanthus x giganteus* in UK funded trials, compared to performance of Hybrid *Miscanthus*, and *Miscanthus x giganteus* controls in the Hybrid trials[3,4,5,6,7]. With yield comparison to current UK *Miscanthus* crops over 112 harvest events in the UK, all on marginal land, and UK Government trials of *Miscanthus x giganteus*[1,2].



References from Fig 1.

1 Continued assessment of agronomy and yield potential of Miscanthus for industrial cropping in the UK. DEFRA project code. NF040.
2 Christian D.G et al. Growth Yield and mineral content of Miscanthus x giganteus grown as a biofuel for 14 successive harvests, Industrial Crops and Products (2008)
3 - Optimizing seed-based *Miscanthus* plug plant production with supplemental heat and light, compost type and volume. Global Change Biology, 12920. Wu *et. al.* 2021.
4 "Two-steps" seed-derived plugs as an effective propagation method for the establishment of *Miscanthus* in saline–alkaline soil. Global Change Biology, 12820. Zheng *et. al.*. 2021.
5 Extending Miscanthus Cultivation with Novel Germplasm at Six Contrasting Sites.Front. Plant Sci. 8:563. Kalinina O *et. al.* 2017.
6 A new carbohydrate retaining variety of Miscanthus increases biogas methane yields compared to M x giganteus and narrows the yield advantage of maize. Food Energy Security. 2020; 9: e224. Kam *et. al.* 2020.
7 Breeding strategies to improve Miscanthus as a sustainable source of biomass for bioenergy and biorenewable products. Agronomy, 9(11), 1-17. Clifton Brown, J. et. al. 2019.

Fig 2 – Sources, genera and number of varieties sourced so far by NEF during Phase 1.

Country	New Material	Number of varieties sourced
USA	Miscanthus.	4
USA	[redacted]	2
USA	Miscanthus.	6
USA	Energy Cane	3
USA	Miscane	3
USA	Pennisetum.	4
Germany/USA	Miscanthus.	3
Canada	Miscanthus.	11
Control varieties	Miscanthus	4
	Total	**40**

Fig 3. The rational for importing is based on the overlap in hardiness zones between key areas of germplasm importation from North America. The two maps show hardiness zone 8 overlap for the US, light yellow (8a) and mid yellow (8b) colours and EU hardiness zone 8 which is in red on the EU map. NEF is importing material mainly from zone 8 with some from 9 (warmer) and 7 (colder).

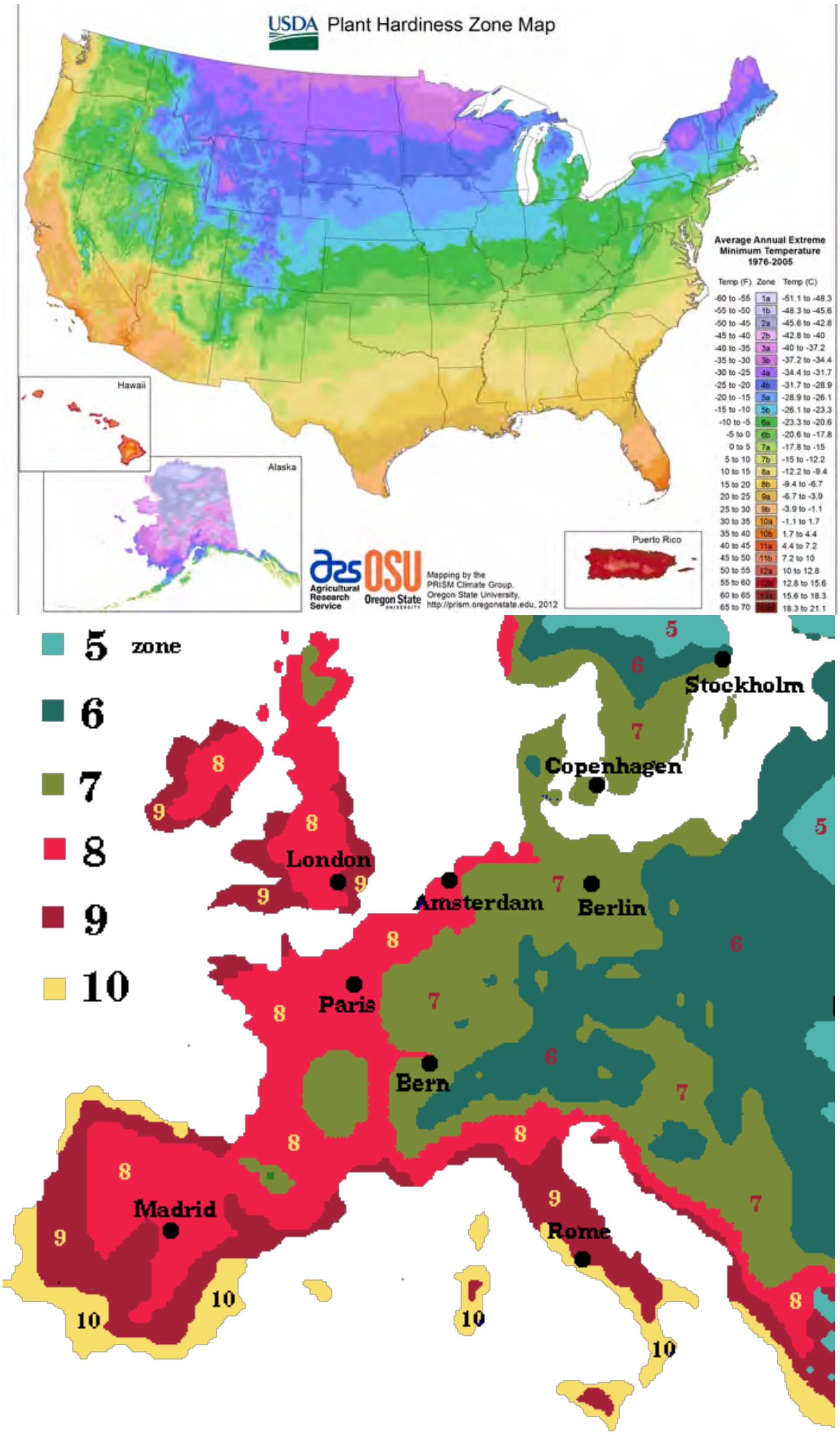
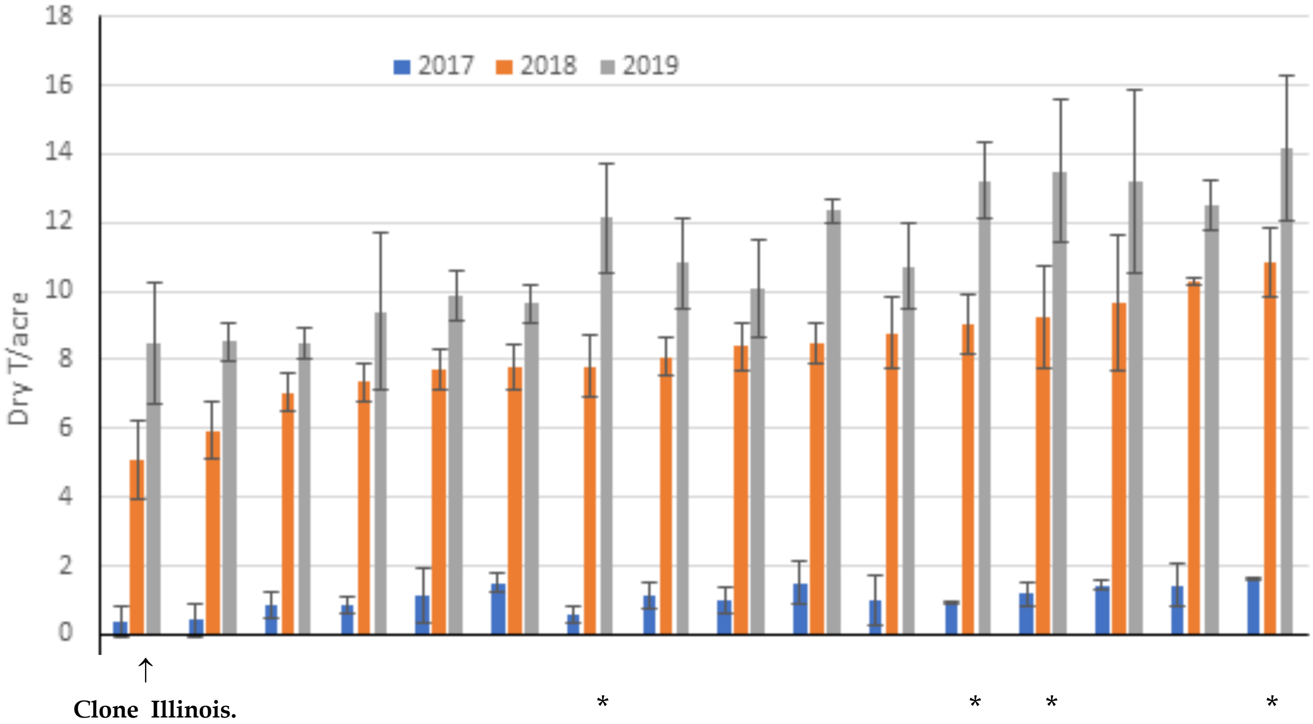
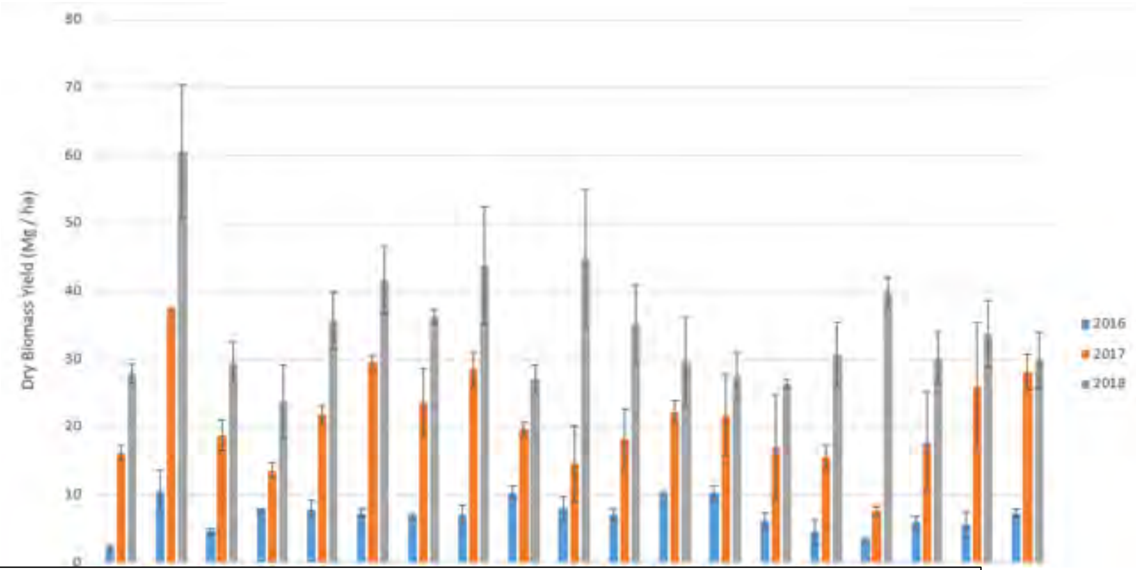


Fig 4. Variety yields (over 3 years) from a US Miscanthus breeding programmes submitting varieties to NEF. Yields for varieties presented to NEF indicated with *, compared to the US control (Clone Illinois first on the left). Please note yield is presented as tonnes per acre, image of US trial site for this trial with clone Illinois indicated with an arrow.



[redacted]

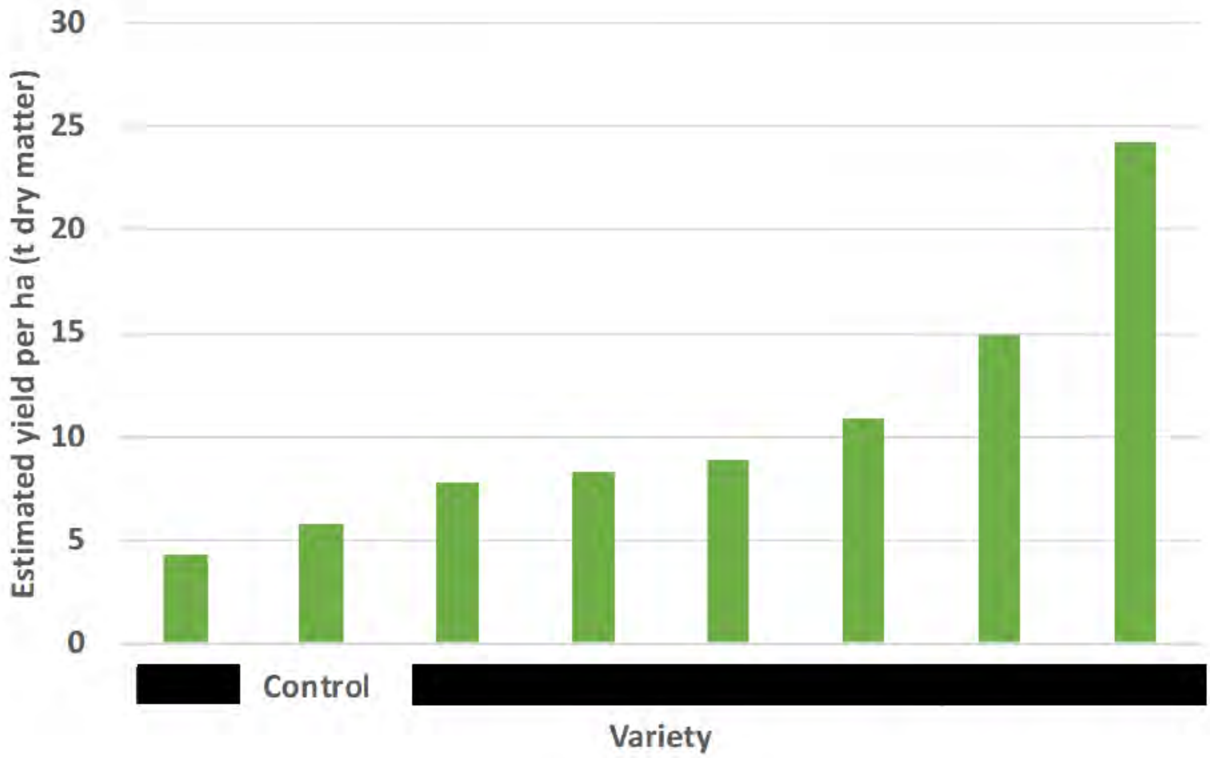
Fig 5. Yield of Saccharum clones in the US as t/ha, compared to Miscanthus (Miscanthus x giganteus control yield are the first data bars on the left). The best variety imported to the UK from this program had a yield of 60 t per ha dry matter.



Variety codes removed for confidentiality

Fig 6. NEF Phase 1 variety yield assessment from specimen trial data, (a) projected yield t/ha dry matter for 7 varieties, compared to a *Miscanthus x giganteus* control, (b) stem numbers per plant.

(a)



(b)

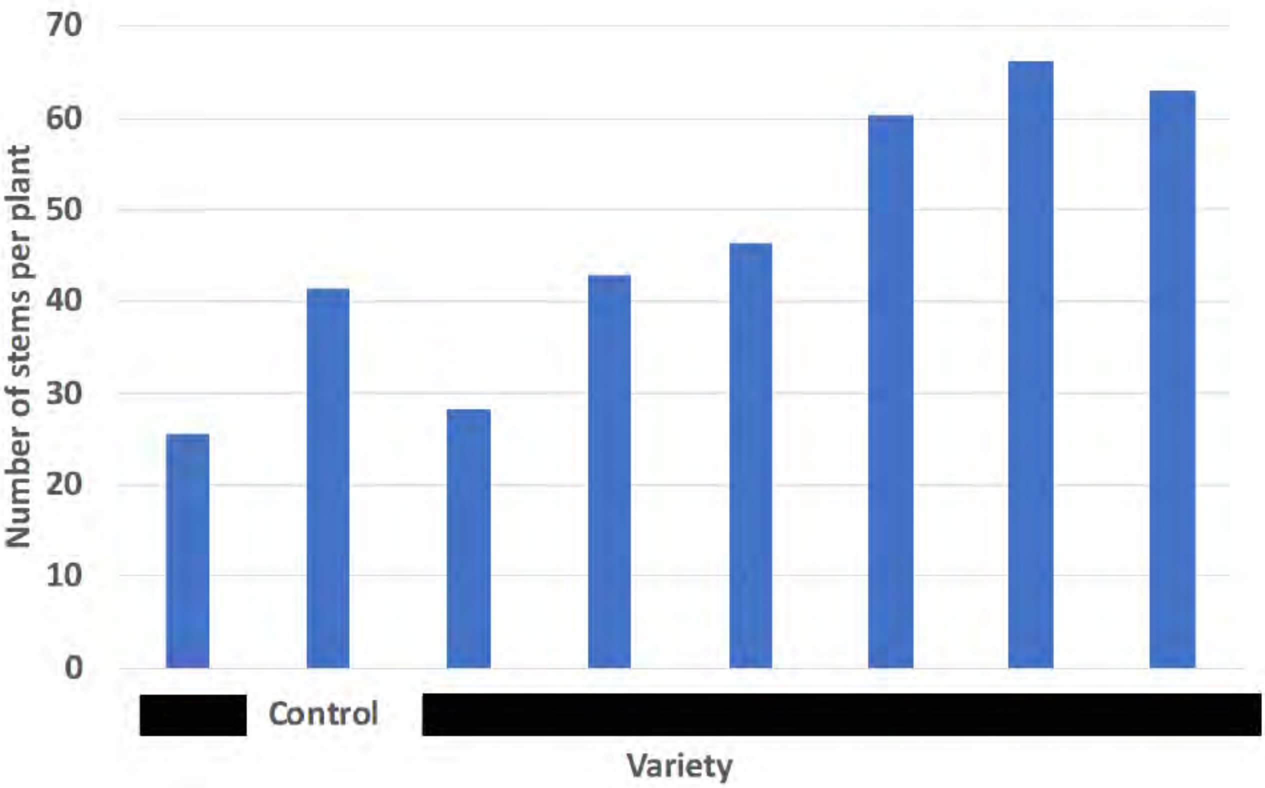


Fig 7. Specimen plot yields from 3 replicate plants (Taunton, January 2022)

Variety	Stems/plant	% moisture content.	Yield t/ha	Yield as % of Control
[redacted]	26	51	4.28	74
Miscanthus x giganteus (Control)	41	69	5.78	100
[redacted]	28	51	7.79	135
[redacted]	43	48	8.30	144
[redacted]	46	48	8.85	153
[redacted]	60	60	10.89	173
[redacted]	66	43	14.89	258
[redacted]	63	66	24.29	420

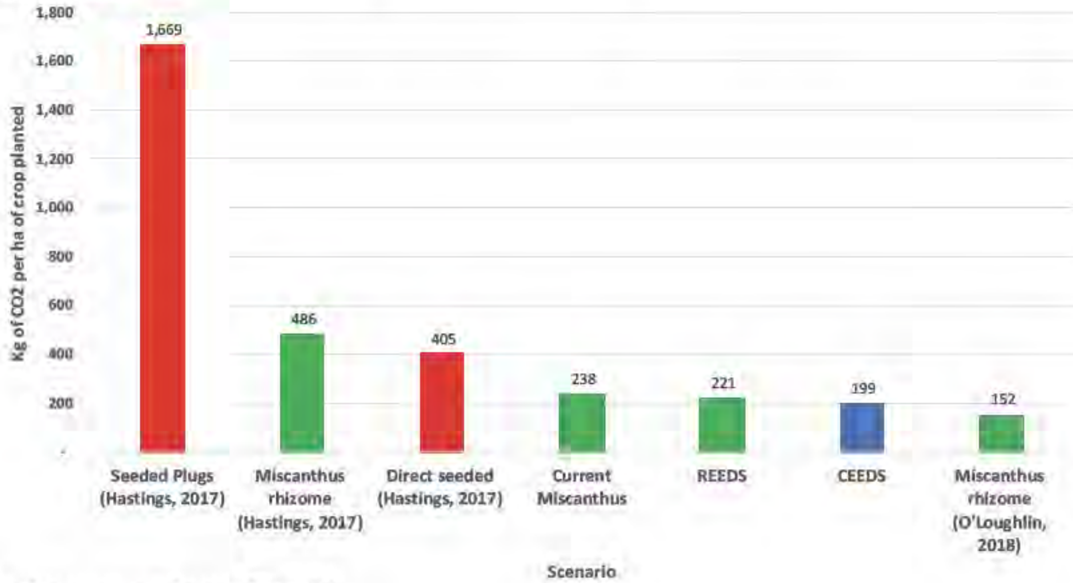
Fig 8. Specimen Trial plots, Taunton January 2022. The three replicates of Miscanthus variety [redacted] are identified by red dots, with the control (Miscanthus x Giganteus) indicated by a red arrow.

Fig 9. Estimated cost from 1992 for *Miscanthus* seeded hybrid breeding projects in the UK The number of varieties at registration stage (not commercial) from three programs with an estimated investment cost per variety.

Program	Cost of R & D projects (£)	Number of projects
DEFRA	4,753,833	16
BBSRC	13,493,061	43
Other (UK and EU)	8,013,451	4
Total (£)	**26,260,345**	63
Number of varieties from these programs at registration stage		**7**
Estimated cost per variety to date (£)		**3,751,478**

Fig 10. Images of NEF CEEDS development plantings of sugarcane in Brazil. Showing CEEDS planted against a commercial control of conventional stem planting. A CEEDS planter developed for sugarcane, the CEEDS product for scale in a persons hand, and the vigorous growth from a germinated CEEDS, all under field conditions.

Fig 11. CO_2 emissions (Kg CO_2) per ha of planted crop, for different *Miscanthus* establishment scenarios.



References from Fig 11

[Hastings, 2017] Economic and Environmental Assessment of Seed and Rhizome Propagated Miscanthus in the UK. Hastings A, et al. Frontiers in Plant Science. Volume 8, 2017.
[O'Loughlin 2018] O'Loughlin, John & Mcdonnell, Kevin & Finnan, John. (2018). Quantifying the economic and greenhouse gas balance advantages of establishing miscanthus from stem cuttings. Biomass and Bioenergy. 109. 147-154. 10.1016/j.biombioe.2017.12.010.

[redacted]

Fig 13. Number of mini rhizomes produced per ha with CEEDS production in the UK compared to conventional *Miscanthus* field rhizome production.

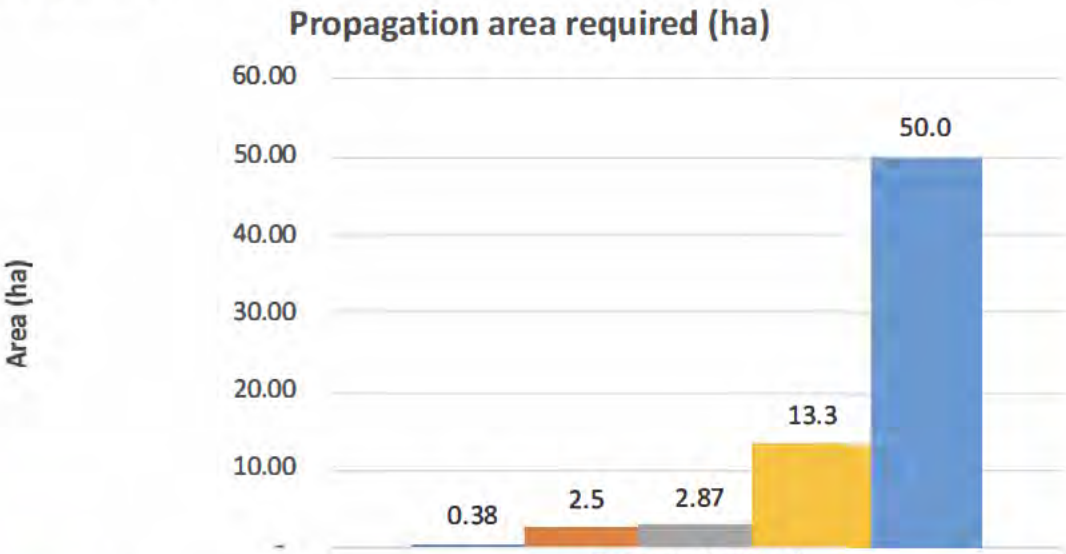
Propagation option	Rhizomes per m-2	Rhizomes per ha (Million)
Conventional field rhizome yield	40	0.4
CEEDS rhizome yield from Phase 1 evaluation	2,023	20.23

Fig 14. Prior work conducted by NEF in the US to propagate *Miscanthus* commercially at higher planting densities as precursor work to REEDS, on higher grade land with water and nutrient inputs. This resulted in 3 times the yield, up to 1.5 million rhizome cuttings per ha.

[redacted]

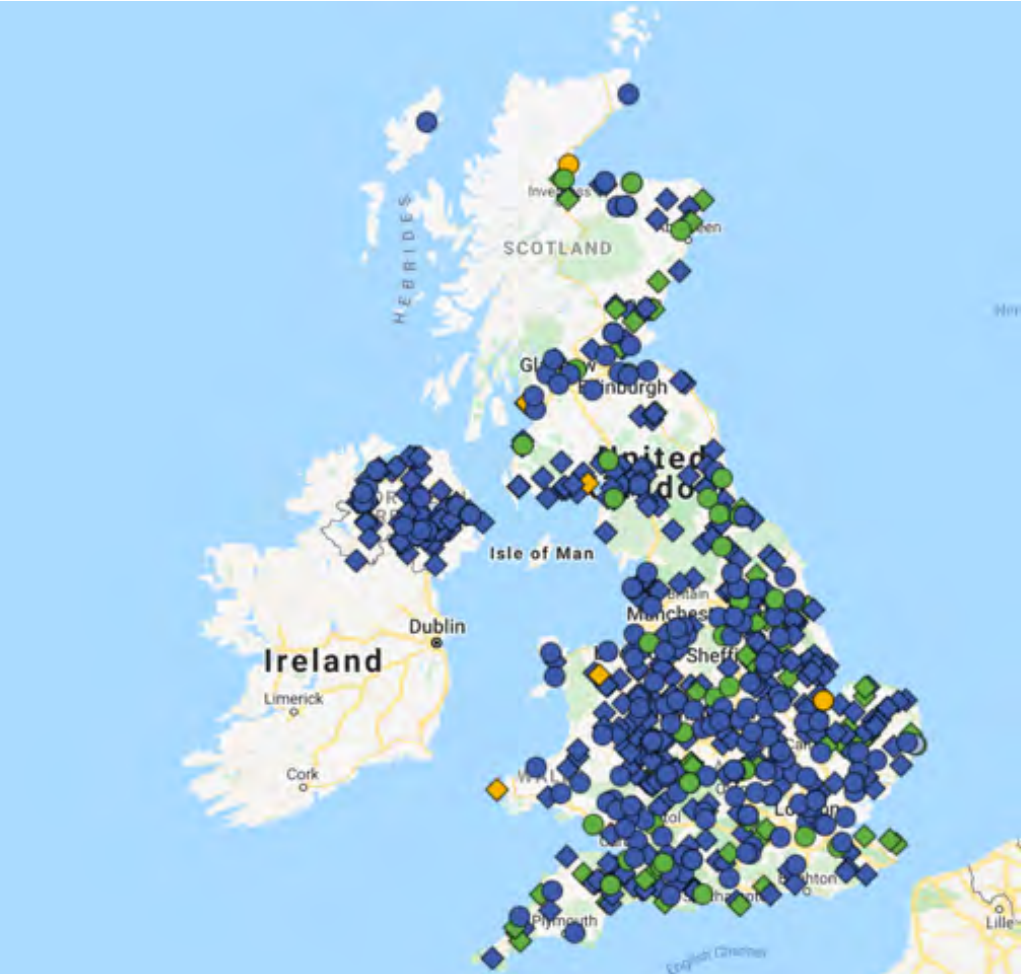
[redacted]

Fig 17. Area required by different *Miscanthus* propagation options to produce material for 1,000 ha of planting.



	Area required (ha)
CEEDS v2	0.38
Seeded Plugs	2.5
CEEDS	2.87
REEDS v2 Rhizome	13.3
Current Rhizome system	50.0

Fig 18. Map of UK AD sites.



[redacted]

[redacted]

Fig 21. NPV of *Miscanthus* with different yield and establishment cost. All models used 20 years of production, with a ramp up and a decline in yield from years 15 to 20. Offtake of biomass sold ex farm gate at £75 per odt. Results show yield increase results in the greatest increase in farmer NPV, and associated update potential.

Scenario	NPV (£)	Planting cost £/ha
Commercial Giganteus (Rhizome) 10 t ha	4,331	1580
Hybrid Miscanthus (plug plants) 10 t ha *	4,831	1050
Hybrid Miscanthus (plug plants) 15 t ha *	8,252	1050
Vegetative Miscanthus 20 t ha - Current establishment cost	11,174	1580
Vegetative Miscanthus 25 t ha - Current establishment cost	14,595	1580
Vegetative Miscanthus 20 t ha - 30% Lower establishment cost	11,674	1050
Vegetative Miscanthus 25 t ha - 30% Lower establishment cost	15,095	1050

FIG 22 - NEF PROJECT GANTT CHART

[redacted]

FIG 22 - NEF PROJECT GANTT CHART (CONTINUED)

[redacted]

Fig 23. Risk Assessment Table

[redacted]

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