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Optimization of crop tissue culture technology and its impact on biomolecular characteristics

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Abstract: Modern agriculture relies on crop tissue culture technology for fast propagation, genetic improvement, and protection of plant species. Conventional media like Murashige and Skoog (MS) usually lack optimal conditions for embryogenesis, necessitating the development of improved media tailored to specific crop requirements. In this article, we introduce an Efficient Grid Identified-Deep Feedforward Neurons (EGI-DFFN) to identify the ideal nutrient and vitamin levels of the crop plants for improving crop tissue culture aimed to improve crop plant growth in a lab setting by predicting callogenesis rate (CGR), embryogenesis rate (EGR), and somatic embryo number (SEN), shoot regeneration rate (SRR), rooting rate (RR). Different concentrations of ionic macronutrients, bio-molecular, and vitamins of the crop plant are the input to the predictive model, which is collected through the laboratory Callus Induction Experiment (CIE). Z-score normalization is used to preprocessing the CIE data to ensure consistent scales across different input features and improve model training performance. DFFN used discriminates to predict complex relationships and interactions between CGR, EGR, SEN, SRR, and RR with EGI tuning. The EGI-DFFN model has significantly improved crop tissue culture growth by accurately predicting the CGR, EGR, SEN, SRR, and RR respectively. The EGI-DFFN model enhances understanding of how ionic macronutrients and vitamins impact plant growth. It identifies optimal concentrations of the biomolecular to enhance somatic embryo formation and plantlet development, providing insights for optimizing crop tissue culture conditions for optimal growth outcomes.

Keywords: crop plant; tissue culture; ionic macronutrients; efficient grid identified-Deep feedforward neurons (EGI-DFFN); biomolecular

1. Introduction

Crop tissue culture technology, an in vitro plant propagation approach, has revolutionized agricultural practices by imparting a controlled environment for the rapid multiplication of plants [1]. This era entails the development of cells of plants, tissues, or organs on the medium of nutrients underneath a sterilized environment, allowing the improvement of entire flowers from small explants. Originally tissue subculture strategies have advanced substantially, and they are now broadly used in crop development applications, plant breeding, and the conservation of plant genetic sources [2]. As global agricultural structures face demanding situations like climate alternate, soil degradation, and growing populace pressures, optimizing tissue subculture generation presents an opportunity to improve crop manufacturing at the same time as keeping or enhancing the quality of vital agricultural traits [3,4].

Optimization of crop tissue culture approaches involves refining the factors that impact increase and improvement in vitro, consisting of the choice of explants, media composition, mild conditions, temperature, and the application of plant

growth regulators (PGRs). These factors directly affect the efficiency of micropropagation and the pleasant of the plants produced [5]. For example, the concentration and aggregate of PGRs together with auxins and cytokinin can drastically influence cellular department, differentiation, and organogenesis. Similarly, adjustments within the nutrient media, which includes macro- and micronutrients, nutrients, and carbon sources, are critical for attaining a high regeneration rate. Continuous efforts have been made to excellent-tune those variables to reap top-quality results for specific crop species, to ensure uniformity, disease resistance, and progressed yield [6].

Beyond the practical application of enhancing crop yields and lowering propagation time, the effect of tissue culture on the biomolecular characteristics of plants has garnered substantial medical hobby [7]. Tissue way of life situations can impact the expression of unique genes related to plant growth, pressure tolerance, and secondary metabolite production. This is particularly important for plants that are precious now not only for their agricultural yield but also for bioactive compounds, which include medicinal vegetation and vegetation utilized in pharmaceuticals or nutraceuticals. Optimized tissue tradition situations can result in increased manufacturing of bioactive molecules like alkaloids, flavonoids, and phenolic compounds, which have critical fitness benefits [8]. Furthermore, genetic balance in tissue-cultured flowers is a vital subject, as soma clonal variations, which occur because of the in vitro surroundings, can result in unwanted genetic mutations. Although those versions can occasionally be high quality by using introducing novel traits, ensuring genetic fidelity in micro propagated vegetation is important for maintaining consistency in crop performance [9]. Molecular strategies along with Deoxyribonucleic Acid (DNA) fingerprinting, Simple Sequence Repeat (SSR) markers and Random Amplified Polymorphic DNA (RAPD) are increasingly being utilized to assess the genetic balance of tissue-cultured vegetation and to display modifications at the biomolecular degree [10].

The maximum commonly used traditional medium in plant tissue way of life is the Murashige and Skoog (MS) medium, which incorporates a balanced mixture of essential nutrients, vitamins, and hormones. However, the effectiveness of this medium can vary depending on the specific crop species and the developmental degree of the plant [11]. While MS medium has been instrumental in tissue way of life fulfillment, it frequently falls quickly in optimizing situations for crucial techniques like embryogenesis and callogenesis. Therefore, there may be an increasing need for media formulations tailor-made to the particular biochemical and physiological needs of numerous crops. Optimization of nutrient concentrations, ionic macronutrients, and vitamins is important to improve embryogenesis charges, rooting performance, and shoot regeneration, all of which might be vital for successful tissue tradition [12].

1.1. Objective of the study

The purpose is to optimize crop tissue culture technology by developing an Efficient Grid Identified-Deep Feedforward Neurons (EGI-DFFN) model. This model aims to identify the ideal concentrations of ionic macronutrients, vitamins,

and biomolecular components necessary for enhancing callogenesis, embryogenesis, and somatic embryo formation. By accurately predicting key growth parameters, the model seeks to improve plantlet development in lab conditions, ultimately aiding in rapid propagation and genetic improvement of crop species.

1.2. Contribution of the study

- 1) To develop an EGI-DFFN model for precise prediction of key tissue culture parameters.
- 2) To enhance the understanding of the relationship between nutrients and plant growth outcomes.
- 3) To optimize conditions for improved somatic embryo formation and plantlet development.

The structure of the paper is as follows: Section 2 explains the relevant works; Section 3 provides a detailed explanation of the process of tissue culture; Section 4 details the methodology; Section 5 provides the result; Section 6 gives a discussion of the study; Section 7 concludes the study.

2. Related works

Niazian and Niedbała [13] used Nonlinear nonparametric machine learning (ML) algorithms, including neural networks (NN), random forests (RF), partial least square regression (PLSR) then support vector machines (SVM) to manage complicated data, categorize genotypes, predict quantitative characteristics, and improve breeding practices. Plant breeding and biotechnology research include nondeterministic properties, which made classical statistical approaches impractical for data interpretation. Accurate plant measurement combined with ML analysis could lead to precision agriculture. Zhang et al. [14] used four culture circumstances agar meditation, light duration, culture temperature, and moisture to predict the melon differentiation rate using artificial neural networks (ANN). Using a four-layer NN design with traingdx as the training function, the ideal back propagation neural network (BPNN) was created. ANN and genetic algorithm (GA) combined could improve the circumstances of plant tissue culture with high prediction accuracy, which made it a potential technique for further biological investigations.

Sathyavani et al. [15] examined nutritional inadequacies in leaves of plants including coriander, tomato, pepper, and chili utilizing an Internet of Things (IoT)-based image capture and nutrition analyzer strategy. Images of leaves were processed using an enhanced convolutional neural network (ECNN) to extract patterns. The CNN was incorporated into virtual machines, and the system stored and processed data on the cloud. Wijerathna et al. [16] investigated wheat genetics for a wide range of properties of wheat's enormous chromosomal number and polyploid DNA. However, research on the genetic characterization of diploid species was hampered by its huge genome size. Genes might be added or changed without changing the genetic background by genetic transformation and gene editing techniques, which provide an alternative to traditional breeding methods. It was feasible to transfer beneficial genes to wheat, enhancing trait values including conflict with abiotic and biotic influences, plant architecture, and grain quality. It

was made possible by advancements in the development of gene delivery techniques and plant tissue regeneration.

Mohammed et al. [17] designed and assessed an IoT-mechanical ex vitro acclimatization system (E-VAS) that would improve morpho-physiological attributes while lowering mortality rates and contamination risk. The E-VAS recorded a higher plant survival rate and phenotype quality than the manual system, suggesting that it could be a viable solution for the growing demand for tissue culture-produced planting materials. The capacity of industrial hemp calli grown in vitro explants as a sustainable strength crop become investigated by means of Norouzi et al. [18]. To broaden a crude, Hydrothermal Liquefaction (HTL) experiments had been designed after the explants' elemental and chemical compositions had been analyzed. The hydrocarbons that resemble petroleum, their practical groups, and their chemical additives were located.

Pandey et al. [19] aimed to predict nutrient concentrations and deficiency in hydroponic lettuce using in situ hyperspectral imaging. The research used Salanova Green lettuce grown with varying macronutrient fertility rates and also used PLSR models to predict nutrient concentrations for nitrogen, phosphorous, potassium, calcium, magnesium, and sulfur. Dan and Tomat [20] investigated the formation of antioxidant properties and bioactive compounds in both callus and explants and the development of callus in tomatoes. To create callus on MS media, tomato sprouts' cotyledon and hypocotyl were employed. The findings demonstrated that both explants developed friable callus and produced significant amounts of antioxidant chemicals from ascorbic acid and pigments. Hydrocarbons predominated among the metabolites.

Rahman et al. [21] highlighted the dangers and difficulties involved in employing *Agrobacterium*-mediated transformation to create transgenic crops and offered a thorough study of these crops. It also addressed how cutting-edge biotechnology methods like Clustered Regularly Interspaced Short Palindromic Repeats and CRISPR-associated protein 9 (CRISPR/Cas9) systems were used to successfully change genetic material.

Wijerathna-Yapa and Hiti-Bandaralage [22] emphasized the significance of plant tissue culture for identifying main abiotic stressors, drought, salt, and the emergence of disease resistance. More focus was placed on transgenic technology applications for stress tolerance and metal accumulator screening.

Research gap

In modern agriculture, crop tissue culture technology is critical for hasty propagation, genetic improvement, and maintenance of plant species. However, optimizing the nutrient and environmental conditions to enhance plant growth remains a significant challenge. Conventional media, such as MS, often fail to provide optimal conditions for callogenesis, embryogenesis, and shoot regeneration, limiting the efficiency of plant tissue culture. While recent studies have integrated ML algorithms, such as SVM, RF, and ANN, to improve data prediction and breeding practices, there remains a need for a robust predictive model that can accurately identify the ideal concentrations of macronutrients, vitamins, and

biomolecular components to optimize plant tissue culture. To overcome this issue, the EGI-DFFN model was proposed, which is intended to predict and optimize complex interactions between nutrient levels and growth parameters, thereby improving the precision and efficiency of plant tissue culture technologies.

3. Tissue culture process

The basis of crop tissue culture steps, from the selection of plant tissue to the final transfer to soil for continued growth is illustrated in **Figure 1**.

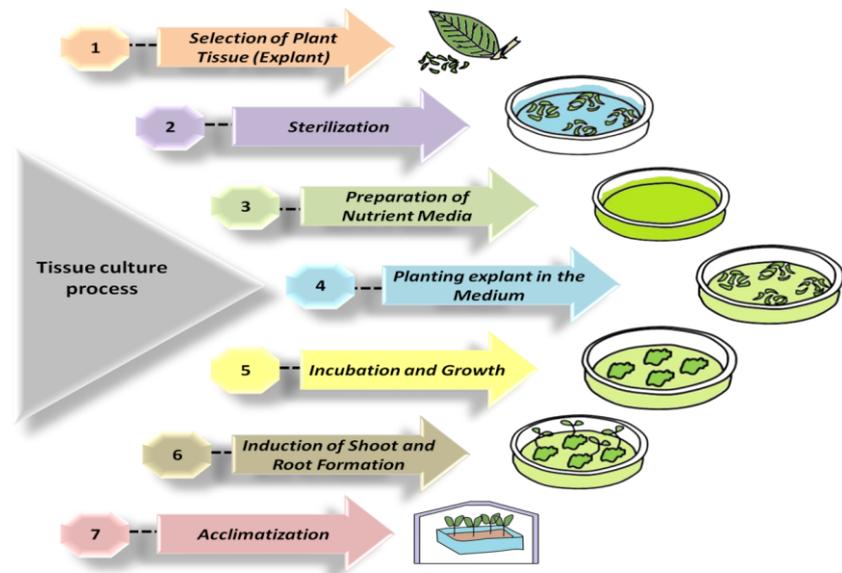


Figure 1. Process of tissue culture.

- 1) Selection of Plant Tissue (Explant): A healthy part of the plant, often a leaf, stem, or root, is chosen as the explant for tissue culture. It is trimmed to the desired size.
- 2) Sterilization: The selected explant is sterilized by washing it with sterilizing agents (such as ethanol or sodium hypochlorite) to eliminate contaminants like bacteria and fungi.
- 3) Preparation of Nutrient Media: A nutrient-rich medium (usually MS medium) containing essential vitamins, minerals, hormones, and sugars is prepared in Petri dishes or flasks. This medium supports the enlargement of the plant tissues.
- 4) Planting explant in the Medium: The sterilized explant is transferred to the prepared medium under sterile conditions, often in a laminar flow hood to prevent contamination.
- 5) Incubation and Growth: The Petri dishes or flasks containing the explant are placed in a controlled environment (usually under specific light and temperature conditions). Over time, the explant begins to form callus (undifferentiated cells).
- 6) Induction of Shoot and Root Formation: Once a callus is formed, the nutrient medium is altered with plant growth hormones (auxins and cytokinin) to induce the formation of shoots and roots, developing into a plantlet.
- 7) Acclimatization: The developed plantlets are carefully detached from the tissue culture containers and transferred to soil or another growing medium. They are

acclimatized to outdoor conditions in a greenhouse or controlled environment before being planted in the field.

4. Methodology

The methodology for this study encompasses the detailed experimental design, data collection procedures, parameter explanations, and preprocessing techniques used to evaluate and optimize tissue culture conditions. These components are thoroughly outlined in this section, providing a comprehensive overview of how the Efficient Grid Identified-Deep Feedforward Neurons (EGI-DFFN) approach was applied to enhance crop tissue culture outcomes. **Figure 2** illustrates the flow of the overall methodology.

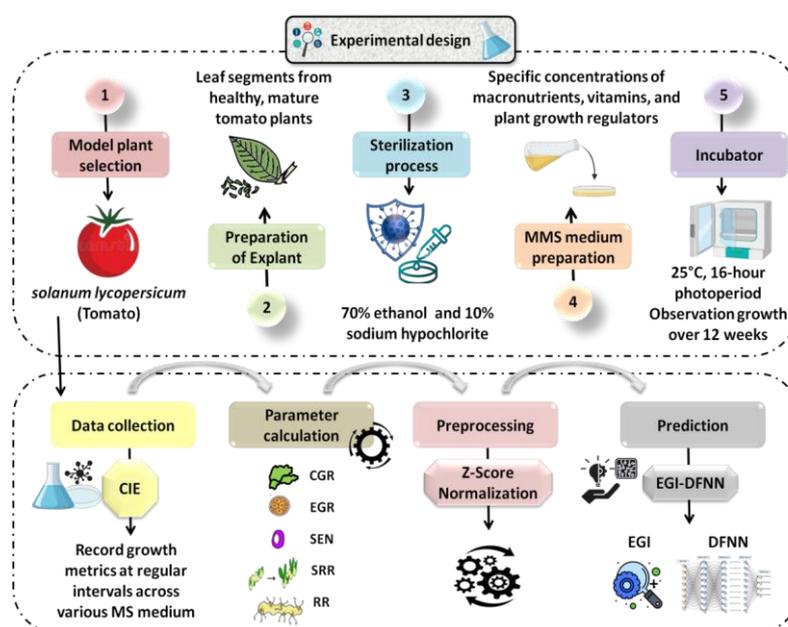


Figure 2. Overall methodology.

4.1. Experimental design

In this study, *solanum lycopersicum* (tomato) was selected as the model plant species for tissue culture optimization due to its well-documented regenerative capacity and relevance to agricultural research. Healthy, mature tomato plants were used as the source for explants, which were prepared from leaf segments. To ensure sterility, the explants were sterilized using a 70% ethanol wash for 30 s, and a 10-*minute* soak in 10% sodium hypochlorite. With sterile distilled water the explants were then rinsed thoroughly to remove any residual sterilizing agents. The explants were placed on a Modified MS (MMS) medium (**Table 1**), optimized for tomato callus induction, containing specific concentrations of macronutrients, vitamins, and plant growth regulators (BAP, NAA, and 2,4 – D). The cultures were incubated at 25°C under controlled conditions with a 16-hour photoperiod and observed regularly for callus formation and growth over 12 weeks (3 months) to evaluate the impact of different concentrations on tissue culture outcomes.

Table 1. Composition of standard and MMS medium.

Component		Standard MS Medium	MMS Medium for Tomato
Macronutrients (mg/L)	Nitrogen (NH_4NO_3)	1650	1200
	Potassium (KNO_3)	1900	1700
	Phosphorus (KH_2PO_4)	170	150
	Calcium ($\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$)	440	500
	Magnesium ($\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$)	370	400
	Sulfur ($\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$)	370	370
Micronutrients (mg/L)	Boron (H_3BO_3)	6.2	5.0
	Manganese ($\text{MnSO}_4 \cdot \text{H}_2\text{O}$)	16.9	20.0
	Zinc ($\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$)	8.6	10.0
	Iron ($\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$)	27.8	30.0
	EDTA – Fe	37.3	40.0
	Copper ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$)	0.025	0.025
	Cobalt ($\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$)	0.025	0.025
Vitamins (mg/L)	Thiamine – HCl (Vitamin B1)	0.1	0.5
	Pyridoxine – HCl (Vitamin B6)	0.5	0.5
	Nicotinic acid	0.5	0.5
	Myo – inositol	100	100
Plant Growth Regulators (mg/L)	6 – Benzylaminopurine (BAP)	0	2.0
	Naphthalene Acetic Acid (NAA)	0	1.0
	2,4 – Dichlorophenoxyacetic acid (2,4 – D)	0	1.0
Sucrose (%)		3.0	3.0
Agar (g/L)		7.0	7.0
pH		5.8	5.8

4.2. Data collection

Data were collected using the Callus Induction Experiment (CIE) to evaluate the impact of different concentrations of ionic macronutrients, biomolecular substances, and vitamins on tissue culture outcomes. Key growth metrics, including CGR, EGR, SEN, SRR, and RR, were measured at regular intervals throughout the experiment. These metrics were recorded for each explant across various media formulations, which were designed to identify optimal nutrient and vitamin levels for enhancing tissue culture efficiency in tomatoes.

4.3. Parameter explanation

Callogenesis Rate (CGR): The CGR measures the effectiveness of a tissue culture medium in inducing callus formation from explants. It is calculated as the proportion of explants that effectively increase callus tissue. Specifically, CGR is decided by way of dividing the number of explants that produce callus with the aid of the overall range of explants used in the experiment after which multiplying by one hundred. This price gives an illustration of ways well the medium supports the

preliminary level of callus induction, which is crucial for subsequent plant regeneration strategies Equation (1).

$$\text{CGR} = \left(\frac{\text{Number of explants forming callus}}{\text{Total number of explants}} \right) \times 100 \quad (1)$$

Embryogenesis Rate (EGR): The EGR assesses the potential of the callus tissue to distinguish into somatic embryos. The charge is calculated with the aid of dividing the number of callus tissues that turn into embryos using the overall variety of callus tissues after which multiplying with the aid of a hundred. EGR is a vital metric for comparing the medium's effectiveness in promoting embryogenesis which is a key step in generating plantlets from callus tissues Equation (2).

$$\text{EGR} = \left(\frac{\text{Number of callus tissue forming embryos}}{\text{Total number of callus tissue}} \right) \times 100 \quad (2)$$

Somatic Embryo Number (SEN): SEN refers to the whole remember of somatic embryos produced in step with explant or in keeping with the unit place of callus. It offers a quantitative degree of the performance of the embryogenesis culture. SEN is calculated through at once counting the wide variety of somatic embryos that shape on each explant or in each callus vicinity. A better SEN indicates a greater successful embryogenesis and a greater potential for generating multiple plantlets from a single explant.

Shoot Regeneration Rate (SRR): SRR measures the proportion of explants or callus tissues that correctly produce shoots. This rate is calculated using dividing the number of explants or callus tissues that form shoots via the overall quantity of explants or callus tissues after which multiplying by using a hundred. SRR is crucial for figuring out how efficiently the medium supports shoot improvement, which is vital for in addition plant boom and improvement Equation (3).

$$\text{SRR} = \left(\frac{\text{Number of explant/callus forming shoots}}{\text{Total number of explant/callus}} \right) \times 100 \quad (3)$$

Rooting Rate (RR): RR evaluates the share of shoots that broaden roots. It is calculated by dividing the range of shoots that shape roots with the aid of the entire range of shoots and then multiplying using a hundred. RR is an important parameter for assessing the effectiveness of the medium in promoting root formation, which is important for stabilizing plantlets and preparing them for the switch to soil or other increased environments Equation (4).

$$\text{RR} = \left(\frac{\text{Number of shoots forming roots}}{\text{Total number of shoots}} \right) \times 100 \quad (4)$$

4.4. Pre-procession using Z-score normalization

This statistical normalization technique addresses variability in data collected from the CIE by transforming the feature values into new normalized values through the application of Equation (5), which is derived from the standard deviation and mean for the considered feature.

$$v' = \frac{v - \mu}{\sigma} \quad (5)$$

For each defined characteristic, σ represents its standard deviation and μ is its mean value. This normalization was used for this study's important measurements, which included SEN, CGR, EGR, SRR, and RR. When the Z-score normalization approach is used, principles that are precisely identical to the mean are converted to zero, values above the mean become visible as positive information, and values below the mean appear as negative numbers.

4.5. Prediction using EGI-DFFN

The proposed approach combines Efficient Grid Identified (EGI) optimization with Deep Feedforward Neurons (DFFN) to enhance crop tissue way of life era. EGI's grid search first-class-tunes the DFFN version parameters, permitting specific optimization of nutrient and vitamin stages. This integrated approach accurately predicts key growth metrics, providing valuable insights for optimizing tissue tradition conditions and enhancing crop improvement.

4.5.1. Deep feedforward neurons (DFFN)

DFFN are well-suited for optimizing crop tissue culture technology due to their capability to form complex, non-linear relationships among input features and desired outcomes. In this context, DFFN can effectively analyze and predict the impact of various nutrient and vitamin levels on key growth metrics such as CGR, EGR, SEN, SRR, and RR. The model's depth allows it to capture intricate interactions and dependencies among these variables, leading to more accurate and insightful predictions.

A mathematical model called the DFFN was created by simulating how the human brain interprets data. Three main layers make up most DFFN models an input layer (IL), a few hidden layers (HL) then an exit layer (EL). The numeral of subterranean levels determines the architecture's depth. When there are several HL and neurons in each HL, DFFN may represent a dataset at multiple levels. **Figure 3** displays the schematic of a generic DFFN model.

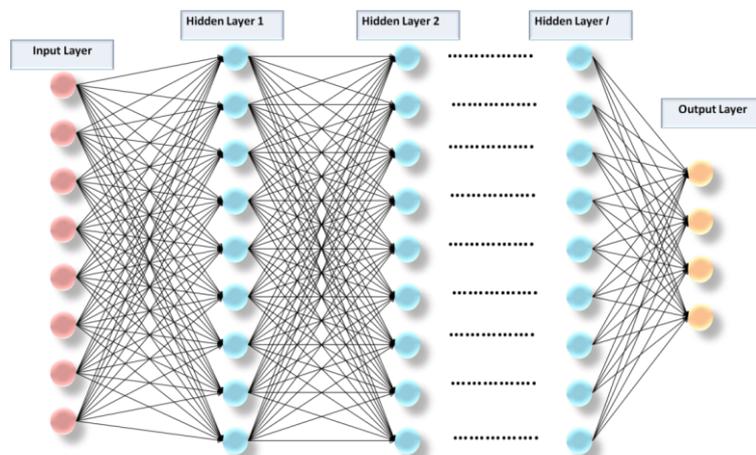


Figure 3. DFFN architecture.

DFFN algorithm.

The data collected from the CIE focuses on various concentrations of ionic macronutrients, biomolecules, and vitamins relevant to crop tissue culture. Input data

W includes the specific nutrient levels and their effects on tissue culture outcomes. Output data Y represents the observed growth metrics (CGR, EGR, SEN, SRR, and RR).

The training database is formed using data, which includes both the input $W = \{W_j | j = 1, 2, \dots, M_{IN}\}$ and output $Y = \{Y_s | s = 1, 2, \dots, S\}$ datasets, which are used to create the training database. M_{IN} represents the different nutrients and vitamin concentrations, and S represents the various experimental conditions and outcomes. W_j has S time samples for every kind of input variable. $W_j = \{W_{j,s} | s = 1, 2, \dots, S\}$. $M_{G,l}$ neurons make up the l^{th} HL out of m HL. Consequently the $i_1^{th} (i_1 | i_2 = 1, 2, \dots, M_{G,1})$ neuron for the $s^{th} (s = 1, 2, \dots, S)$ training sample. The first HL is computed by Equation (6).

$$Z_1, i_1 = e(\sum_{j=1}^{j=M_{IN}} (X_{IN,j,1,i_1} W_j)) \quad (6)$$

Using Equation (7) i_l^{th} the neuron in the $l^{th} (m \geq l \geq 2)$ HL can be obtained. The neuron z in the output layer is determined by Equation (8).

$$Z_l, i_l = e(\sum_{j_{l-1}=1}^{j_{l-1}=M_{G,l-1}} (X_{l-1,i_{l-1},l,i_l} W_{l-1,j_{l-1}})) \quad (7)$$

$$Y = e\left(\sum_{i_m=1}^{i_m=M_{G,m}} (X_{m,i_m,p} Z_{m,i_m})\right) \quad (8)$$

where the number of neurons in the IL (j), the quantity of HL (l), the quantity of neurons in the l^{th} HL i_l , and the activation function (e) are all represented. Additionally, X represents the connection weighting factors between the two neurons. Here $X_m, i_m, \text{ and } p$ indicate the connection weights among the last HL and the output layer, which predicts the growth metrics (CGR, EGR, SEN, SRR, and RR).

Activation Function (AF).

The sigmoid function Equation (9), hyperbolic tangent (\tanh) function Equation (10), rectified linear unit (ReLU) function Equation (12), and exponential linear unit (elu) function Equation (11) are the four AF that are tested to improve prediction performance. These are the possible expressions for the four AF.

$$e(w) = \frac{1}{1 + f^{-w}} \quad (9)$$

$$e(w) = \tanh(w) \quad (10)$$

$$e(w) = \begin{cases} f^w - 1 & \text{for } w < 0 \\ w & \text{for } w \geq 0 \end{cases} \quad (11)$$

$$e(w) = \begin{cases} 0 & \text{for } w < 0 \\ w & \text{for } w \leq 0 \end{cases} \quad (12)$$

These functions help capture non-linear relationships between nutrient levels and growth outcomes. The choice of AF can be adjusted based on which provides the best predictive accuracy for the crop tissue culture metrics.

4.5.2. Efficient grid identified (EGI) optimization

An optimization method called ECI is used to iteratively go through every conceivable combination of a set of variables. It's one of the most fundamentally straightforward and basic optimization algorithms, yet despite this, it's still quite strong and ensures that the paramount possible explanation for trouble will be found. To identify the optimal combination, all potential combinations of parameter values are created into a grid and tested one at a time. The term "grid search" refers to the grid of parameters that are established before the optimization/search process.

The EGI optimization is highly suitable for the objective of enhancing crop tissue culture technology. EGI excels in fine-tuning model parameters, which is crucial for accurately predicting key growth metrics like CGR, EGR, SEN, SRR, and RR. It effectively manages the complex interactions between various nutrients and vitamins, ensuring precise optimization of these inputs. EGI's computational efficiency and scalability handle extensive experimental data from CIE, leading to improved model accuracy and a deeper understanding of nutrient impacts on plant growth.

EGI-DFFN: The integration of the EGI optimization with the DFFN method offers a robust solution for optimizing the crop tissue culture technology. The DFFN's functionality to version complex, non-linear relationships between nutrient levels and more desirable metrics is advanced through the use of EGI, which meticulously searches through parameter combinations to find high-quality settings. This combination allows for unique tuning of input variables, inclusive of ionic macronutrients and vitamins, which right away impact CGR, EGR, SEN, SRR, and RR. By leveraging EGI's efficient grid search capabilities, the DFFN model could provide tremendous experimental records from CIE and efficaciously expect growth effects, as a result imparting deeper insights into how particular nutrient combinations affect plant improvement. This integrated approach ensures that the version no longer simplest predicts with excessive accuracy but moreover offers actionable steerage for optimizing crop tissue culture of life conditions, on the quit improving increase and improvement in a lab placing. The proposed approach's pseudocode is provided in Algorithm 1.

Algorithm 1 EGI-DFFN

```

1: import numpy as np
2: import tensorflow as tf
3: from sklearn.model_selection import ParameterGrid
4: from sklearn.metrics import mean_squared_error
5: def sigmoid(x):
6:     return  $e(w) = \frac{1}{1+e^{-w}}$  (Equation 9)
7: def relu(x):
8:     return np.maximum(0, x)  $e(w) = \begin{cases} 0 & \text{for } w < 0 \\ w & \text{for } w \leq 0 \end{cases}$  (Equation 11)
9: def create_dffn_model(input_shape, activation_function, num_hidden_layers,
10: num_neurons):
11:     model = tf.keras.Sequential()
12:     model.add(tf.keras.layers.InputLayer(input_shape = input_shape))
13:     for _ in range(num_hidden_layers):
14:         model.add(tf.keras.layers.Dense(num_neurons, activation = activation_function))
15:     model.add(tf.keras.layers.Dense(5)) # Output for CGR, EGR, SEN, SRR, RR

```

Algorithm 1 (Continued)

```

16:  model.compile(optimizer = 'adam', loss = 'mean_squared_error')
17:  return model
18:  def egi_optimization(X_train, y_train, X_test, y_test, param_grid):
19:      best_score = float('inf')
20:      best_params = None
21:      for params in ParameterGrid(param_grid):
22:          model = create_dffn_model(
23:              input_shape = (X_train.shape[1],),
24:              activation_function = params['activation_function'],
25:              num_hidden_layers = params['num_hidden_layers'],
26:              num_neurons = params['num_neurons'])
27:          model.fit(X_train, y_train, epochs = 10, batch_size = 32, verbose = 0)
28:          y_pred = model.predict(X_test)
29:          score = mean_squared_error(y_test, y_pred)
30:          if score < best_score:
31:              best_score = score
32:              best_params = params
33:  return best_params

```

5. Result

This section details the experimental setup and findings related to optimizing tomato tissue culture using the EGI-DFFN model. It includes system configuration specifics, growth metrics comparison between MS medium and MMS medium over 12 weeks, and graphical comparisons of key parameters such as CGR, EGR, SEN, SRR, and RR.

5.1. System configuration

Table 2. Hyperparameter setting.

Hyperparameter	Description	Search Range	Optimal Value
AF	The AF used in HL	<i>sigmoid, tanh, ReLU, elu</i>	<i>ReLU</i>
Number of HL	The number of HL in the DFFN model	1–5	3
Number of Neurons	The number of neurons in each HL	32, 64, 128, 256	128
Batch Size	The number of training samples per batch	16, 32, 64	32
Learning Rate	Controls how much the model is adjusted during training	0.001, 0.01, 0.1	0.001
Optimizer	Optimization algorithm used during training	<i>adam, rmsprop, sgd</i>	<i>adam</i>
Parameter Grid Size	Grid size for EGI optimization (number of parameter combinations evaluated)	Various combinations of AF, neurons, and layers	-

In this experiment, Python version 3.8 is used to increase and optimize a DFFN version, integrating EGI optimization to determine the ideal nutrient and diet ranges for improving crop tissue subculture metrics such as CGR, EGR, SEN, SRR, and RR. The setup was performed on a gadget prepared with 16 GB of RAM and a solid-state drive (SSD) to ensure green statistics processing and model education. For statistics manipulation and evaluation, libraries together with NumPy for numerical computations and TensorFlow/Keras were employed for constructing and schooling

the DFFN model. Scikit-research was used for parameter grid search and overall performance assessment. **Table 2** provides the hyperparameter tuning used in the EGI-DFFN model for optimizing crop tissue culture metrics such as CGR, EGR, SEN, SRR, and RR.

5.2. Growth metrics over 12 weeks for tomato plants

Table 3 compares growth metrics for tomato plants cultured in MS-medium and MMS medium, both enhanced with EGI-DFNN. Over 12 weeks, the MMS medium consistently outperforms the MS medium in CGR, EGR, SEN, SRR, and RR demonstrating its superior efficacy in promoting plant development.

Table 3. Comparative growth metrics of tomato plants in MS Medium vs. MMS medium.

No. of. Weeks	Parameter	MS medium + EGI-DFNN (%)	MMS medium + EGI-DFNN (%)
2	CGR	10(%)	20(%)
	EGR	5(%)	15(%)
	SEN	5 embryos per culture	10 embryos per culture
	SRR	3 shoots per culture	7 shoots per culture
	RR	8(%)	15(%)
4	CGR	20(%)	35(%)
	EGR	10(%)	25(%)
	SEN	15 embryos per culture	25 embryos per culture
	SRR	7 shoots per culture	15 shoots per culture
	RR	15(%)	30(%)
6	CGR	30(%)	50(%)
	EGR	20(%)	40(%)
	SEN	25 embryos per culture	35 embryos per culture
	SRR	15 shoots per culture	25 shoots per culture
	RR	25(%)	40(%)
8	CGR	40(%)	60(%)
	EGR	30(%)	50(%)
	SEN	35 embryos per culture	50 embryos per culture
	SRR	25 shoots per culture	35 shoots per culture
	RR	35(%)	50(%)
10	CGR	50(%)	70(%)
	EGR	40(%)	60(%)
	SEN	45 embryos per culture	65 embryos per culture
	SRR	30 shoots per culture	40 shoots per culture
	RR	40(%)	55(%)
12	CGR	55(%)	75(%)
	EGR	50(%)	70(%)
	SEN	50 embryos per culture	80 embryos per culture
	SRR	35 shoots per culture	45 shoots per culture
	RR	45(%)	60(%)

5.3. Comparison phase

5.3.1. CGR on MS and MMS medium

Figure 4 gives a graphical representation of the CGR over 12 weeks for both the preferred MS medium and a changed medium tailored for tomato tissue's way of life. CGR percentages suggest the proportion of callus growth discovered. In the changed medium, CGR is consistently better compared to the MS medium, reflecting the enhanced effectiveness of the changed medium in selling callus development. For instance, at week 12, the CGR is 55% for MS-medium and 75% for the MMS medium. This trend demonstrates that the optimized medium notably quickens and improves callus increase at some point in the subculture duration. This improvement is expected through the use of the proposed method, EGI-DFFN, which aims to discover the best nutrient and nutrition levels to decorate tissue subculture results.

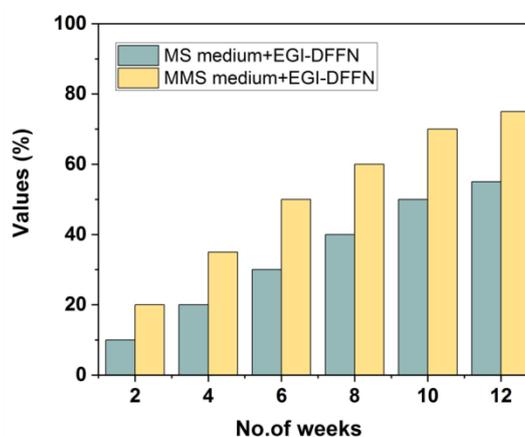


Figure 4. Comparison of CGR levels in both mediums.5.3.2. EGR on MS and MMS medium.

5.3.2. EGR on MS and MMS medium

Figure 5 illustrates the EGR over 12 weeks. EGR probabilities constitute the boom of embryos in cultures. The MMS medium suggests advanced performance as compared to the MS-medium, with a slow increase in EGR from 15% at week 2 to 70% at week 12, while the MS-medium levels from 5% to 50% over the same duration. This indicates that the MMS medium notably complements embryo improvement, main to a better and improved growth of embryos throughout the traditional length. These findings are anticipated by the use of the proposed method for medium optimization.

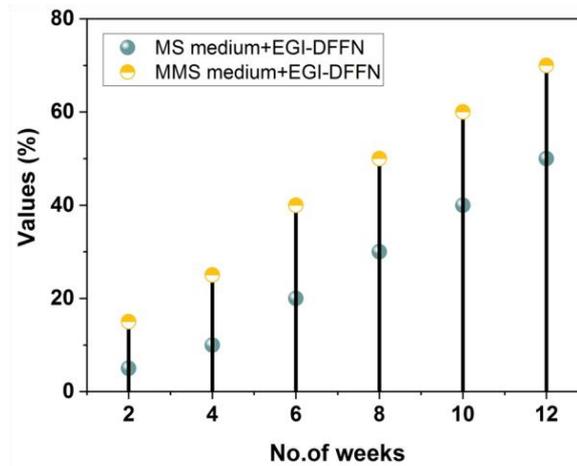


Figure 5. Comparison of EGR levels in both mediums.5.3.3. SEN on MS and MMS medium.

5.3.3. SEN on MS and MMS medium

Figure 6 represents data on the variety of embryos in step with traditional SEN at diverse weeks. The MMS medium yields a better range of embryos in comparison to the MS medium. At week 12, the MMS medium results in 80 embryos per culture, while the MS medium consequences in 50 embryos. This demonstrates the efficacy of the changed medium in assisting and growing embryo production, making it an extra powerful alternative for producing a higher yield of embryos in tissue tradition. These effects are anticipated using the proposed optimization technique for the medium.

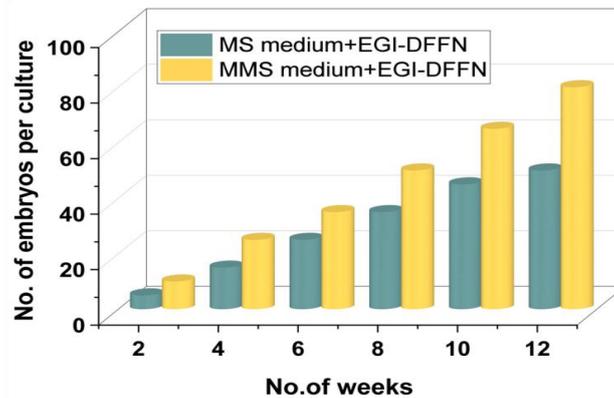


Figure 6. Comparison of SEN level in both mediums.5.3.4. SRR on MS and MMS medium.

5.3.4. SRR on MS and MMS medium

Figure 7 represents the variety of shoots in step with traditional SRR at different weeks. The MMS medium constantly produces greater shoots as compared to the MS medium. For example, at week 12, the changed medium consequences in 45 shoots were in line with culture, whereas the MS-medium consequences were in 35 shoots. This suggests that the MMS medium enhances shoot formation and development, demonstrating its advanced performance in selling shoot growth at

some point in the tissue way of life. These observations predict the usage of the proposed optimization EGI-DFFN approach.

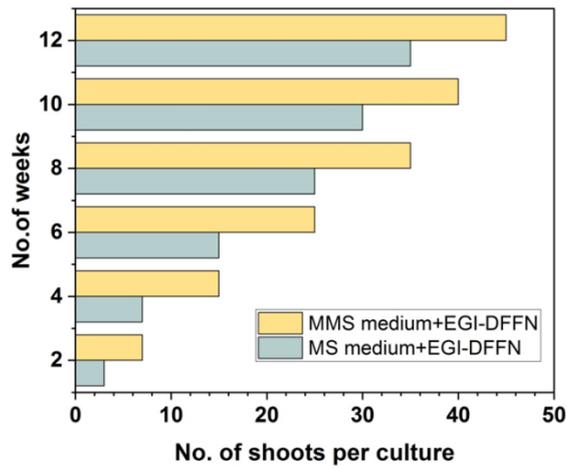


Figure 7. Comparison of SRR level in both mediums.

5.3.5. RR on MS and MMS medium

Figure 8 illustrates the RR possibilities for each MS-medium and changed medium over a 12-week length. The MMS medium exhibits a better rooting price compared to the MS medium. For instance, at week 12, the rooting rate is 60% with the changed medium, as compared to 45% with the MS medium. This development shows that the changed medium provides more effective situations for root improvement, facilitating a higher establishment of plant roots at some stage in the tissue culture method. These effects are anticipated in the usage of the proposed technique for optimizing tissue tradition conditions.

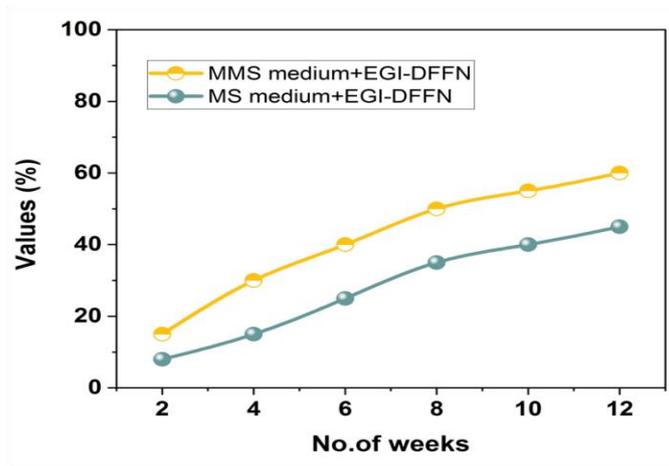


Figure 8. Comparison of RR level in both mediums.

6. Discussion

This study highlights the significant advantages of utilizing the MMS medium, optimized using the EGI-DFFN approach, for tomato tissue culture. The MMS medium consistently demonstrated superior performance across all key growth metrics—CGR, EGR, SEN, SRR, and RR—when compared to the traditional MS

medium. The enhanced effectiveness of the MMS medium can be attributed to the precise tuning of ionic macronutrients, biomolecular substances, and vitamins, as identified by the EGI-DFFN model. By integrating complex nutrient interactions into the predictive model, the EGI-DFFN approach allows for a highly optimized culture environment tailored specifically to the physiological needs of the plant tissue. One of the primary reasons the MMS medium is more effective than the standard MS medium is its ability to provide the optimal balance of essential nutrients that directly affect cellular processes such as callogenesis and embryogenesis. While the traditional MS medium offers a general-purpose formula, it may not fully support the specific nutrient requirements necessary for the rapid and healthy development of certain crops, like tomatoes. In contrast, the MMS medium, fine-tuned by the EGI-DFFN model, delivers a more targeted nutrient profile, which enhances metabolic activity and cellular differentiation. This leads to higher rates of SEN and more efficient SRR, and RR, ultimately accelerating the entire tissue culture process.

Moreover, the MMS medium ensures that the concentrations of key vitamins and biomolecular compounds are aligned with the plant's specific growth demands, which promotes not only faster but also healthier tissue development. For example, the availability of precisely calibrated macronutrients supports stronger callus induction CGR and more robust embryo growth EGR, minimizing the variability often observed in tissue cultures grown on MS medium. This ability to fine-tune nutrient levels directly contributes to a higher success rate in tissue subcultures, reducing waste and improving efficiency in both research and commercial applications. Additionally, the predictive modeling used in this study provides a valuable tool for systematically optimizing media compositions. By leveraging machine learning algorithms, like EGI-DFFN, researchers can identify the most effective nutrient formulations without the need for trial-and-error experimentation, saving both time and resources. This approach could have broad applications, potentially benefiting a wide range of crop species by offering tailored media solutions that significantly improve tissue culture outcomes.

7. Conclusion

The EGI-DFFN model marks a pivotal advancement in crop tissue culture technology, offering a highly effective framework for optimizing nutrient and vitamin concentrations to significantly improve key growth metrics CGR, EGR, SEN, SRR, and RR. By integrating and analyzing the complex relationships between biomolecular factors and plant growth, the model enables precise tuning of culture conditions tailored to specific crop needs. This innovation not only accelerates propagation and genetic enhancement in a controlled lab setting but also contributes to a deeper understanding of plant physiology under tissue culture conditions. The significance of this study lies in its ability to enhance crop propagation strategies with improved accuracy and efficiency, thereby supporting agricultural sustainability and productivity. By offering practical applications that can be readily integrated into laboratory protocols, the proposed model serves as a valuable tool aiming to optimize plant growth and improve crop quality. This study demonstrates how

computational models can drive innovation in biotechnology, providing scalable solutions for faster, more efficient crop development.

Limitations and future works

Despite the substantial progress made, the EGI-DFFN model encounters certain limitations, particularly in handling the computational complexity involved in optimizing multiple parameters. The high number of combinations led to increased computational needs and time requirements. To address this, future research should explore ways to refine the model, potentially by utilizing more advanced optimization algorithms of parallel computing techniques to reduce computation time. Additionally expanding the model's applicability to a wider range of crop and environmental conditions would further validate its robustness. Integration of real-time data and adaptive learning techniques would also improve the model's flexibility, allowing it to dynamically respond to varying growth conditions. These advancements could pave the way for even broader applications in crop biotechnology, enhancing both theoretical insights and practical outcomes in crop propagation and genetic improvement.

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