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Research Article

Artificial Neural Network to Determine the Optimum Nutrient Media Composition in Plant Biotechnology

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Abstract

Growth of various plants in Agriculture industry, whether on-field crop or a plant tissue culture laboratory specimen requires resources and time to obtain the first yield. In traditional way of plant tissue culture huge amount of the resources as well as time is consumed only to adjust growth condition for the minimal threshold of plant required, before actually taking a step further in experimental specimens. Every alteration in the nutrient media consumes further time and resources to obtain the required results by traditional trial and error method. These resources and time spent can be minimised if a virtual computational model can be made instead of performing the actual physical plant growth experiments. This led to the idea of using and creating an artificial neural network to execute the simulation of plant growth and respective yield performance on a computational machine and using those result to actually grow the plant in its optimum yield condition.

Artificial Neural Network is computational technique which mimics the brain synapse signal model into a computer programme which self-learns and provides an output as according to the targeted results. When all inputs and its respective target result models are fed into this computational model and run with maximum accuracy, it then allows a user to try and put any random variable value of initial input and the corresponding result along with effect on actual specimen are displayed on screen. For example, if a computer has the information of what different concentration of 'x' nutrient is affecting the growth of plant, then by putting a value of any concentration of the 'x' nutrient as input will give the exact effect to plant as output. This simple example can be mathematically solved on a two-dimensional graph. But imagine when the medium is made out of several components like x, y, z, a, b, c and each effect the growth of plant. Thus, in this case change in concentration of any component can be modelled and calculated using artificial neural network saving time and other expensive resources to perform each and every experiment individually.

Keywords: Artificial Neural Network (ANN); Optimum; Plant; Nutrient Media; Plant Tissue Culture; Agricultural Advancement; Plant Biotechnology

Abbreviations

ANN: Artificial Neural Network; Demo: Demonstration Version for Software; GB: Gigabyte Computer Memory Storage Unit; Mac: Macintosh Operating System; MB: Megabyte Computer Memory Storage Unit; MLP: Multilayer Perceptron Network; OS: Operating System; USA: United States of America; Vs.: Versus

Introduction

Artificial Neural Network

An Artificial Neural Network abbreviated as ANN is a black box model of computational technology vaguely inspired by the structure and/or functional aspects of biological neural networks [1]. Black Box model implies a system or an object which can be observed overall in terms on the inputs and output given to the system without actually having the knowledge of its internal workings. ANN consist of various interconnected group of artificial neurons. It processes information using a connectionist and response approach to computation. This exactly models the inspirational concept of the functioning of a signal transmitted in a biological neuron by axon, process in cell body and then transmitted by synapse to the next cell by dendrites either to the next cell or output response [2,3].

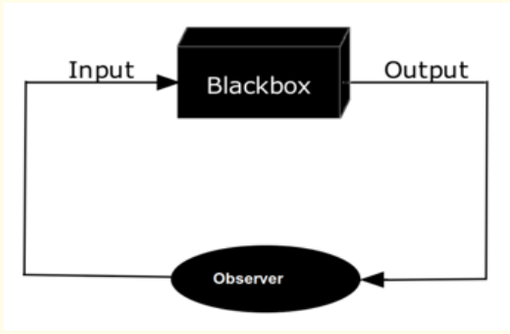
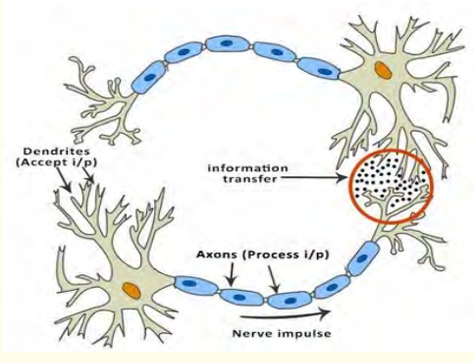


Figure 1: Working Diagram of Black Box Model. Image Courtesy: By Krauss [CC BY-SA 4.0], from Wikimedia Commons.

Most interesting quality of ANN is the possibility of active learning every time the computation is run on a system. In various cases tested till date, ANN performs to be an adaptive system having the capability to change its structure based on external or internal information which flows into the system during the learning phase. Morden Neural Networks are usually non-linear statistical and accurate data modelling tools which are used to model complex relationships between inputs and outputs to find patterns in data [4].



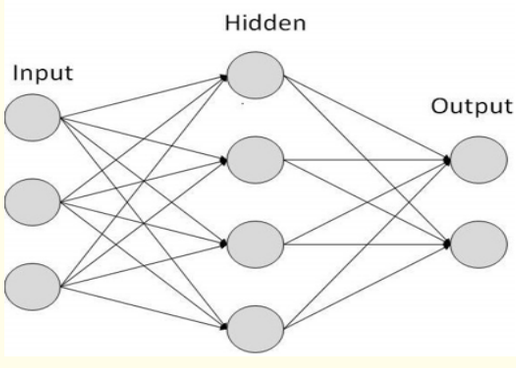


Figure 2: Comparative Diagram between Biological Neuron (left) and Artificial Neural Network (right) Image Courtesy: Tutorial Points.

Example Explanation of Artificial Neural Network

The simplest example to explain the functioning of a Neural Network is, that if a computer is acknowledged by a picture of a dog as input data recognising it as a "DOG" and along with some other picture which does not show the picture of dog as "NOT A DOG". Then by providing more and more pictures of DOG, computer learns external features of DOG having four legs, its facial structure, its tail, and body structure with more and more input data as 'DOG' and ''NOT A DOG''. Finally, when the learning phase is complete, then by providing any random picture to computer, the ANN can now determine the output; that whether the picture has a dog or does not have a dog. Another way to observe its use is by providing computer with hundreds and thousands of random pictures and asking the computer to select or provide only the pictures which has a DOG in them. This huge data processing can be done in less than a minute by ANN which might take a human hours of its time and energy [5].

Conceptual Explanation of Artificial Neural Network

ANN models are based on connections like computing units called 'nodes' which are also known as artificial neurons. These Artificial Neurons have the capacity to receive and transmit signals from one artificial neuron to the other connected artificial neuron. Along with this function, it also has the capacity to process the input signal and then transmit the processed data to the nearest connected artificial neuron. Making each and every connection unit as dependent processor from the information received from the nearest neuron [4,6].

Signal in these artificial neurons is mostly a real number. Computation of the output is some non-linear function of sum of its input. Every artificial neuron is connected by other neuron by the connections which are called as "edges." Edges have weigh system that increases and decreases adjusting with the surrounding inputs and thus information signal is only considered when a minimum threshold potential of the weigh system is reached. This helps in learning pattern of the ANN which also implies a short storage function of every artificial neuron [7].

Various artificial neurons can be clubbed together in layers. And various layers perform a different function based on the input received from either user or neighbouring artificial neuron. Any signal is converted into real numbers and then gets transfers form first layer which is called as "Input layer'' to the final layer also called as "Output layer" through multiple artificial processing neurons called Hidden Layer.

Types of ANN

Multiple Designing of Neural Network created the possibility of number of types of ANN to be known. The research area of ANN is still at its progress and with time and various methods of computing, further types of ANN are being classified. These classifications are based on one or more static components such as number of units in ANN, number of layers in the ANN, units of weigh in ANN and topology of the ANN.

Broadly with its functional topology we can consider having two type of ANN which can be classified further. These two types are namely Feed forward and Feed backward ANN.

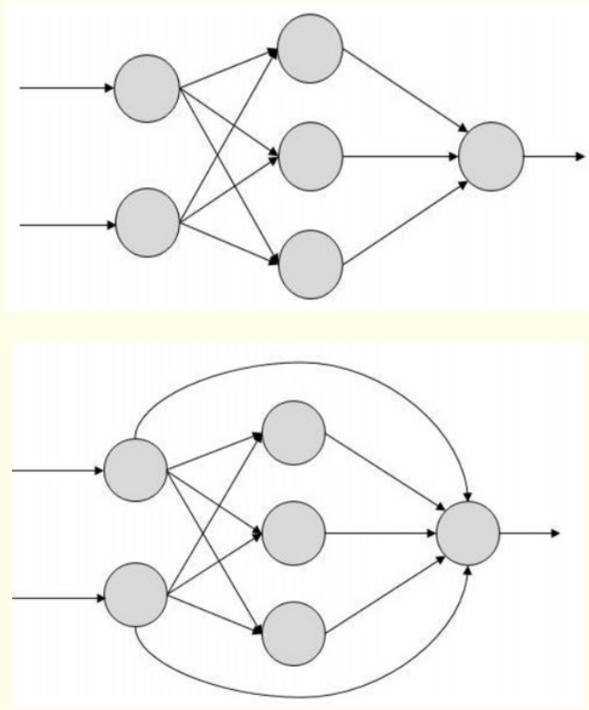


Figure 3.1: Types of ANN, Feed Forward Image Courtesy: Tutorial Points.

Feed forward ANN has the unidirectional flow of information. Every unit or artificial neuron only received information from one end and send to next preceding layer. The arrows in the figure 3.1 and 3.2 shows the connection between two artificial neuron and pathway of their respective signal transmission. These types of ANN can be used in batch systems where only reaction is started at a time does not produce an output or product that will interfere with the initial input. In other words, initial input is independent of final outcome and input layer dose not receive any information from the output or hidden layers.

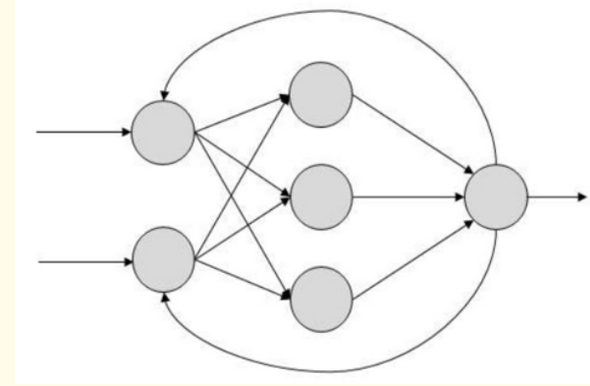


Figure 3.2: Types of ANN: Feed Backward ANN. Image Courtesy: Tutorial Points.

Feedback ANN has the functional component of loops of signal coming from its preceding neurons to pervious neurons. These are mostly used in continuous cultures where any output or its products have an impact on the input. Also, these types have the capacity to access memories and the interlinking of the responses is possible which makes it more suitable in Morden computing. The ANN dose not exactly replicate the function of actual biological neuron as the memory and logical part of transmission of signal in biological neuron is still matter of ongoing research and thus it is only inspired model and is used in modern computing.

Plant Tissue Culture Requirements

Growing plants in laboratory requires resources like nutrient medium, natural or artificial light, container to hold the plant, sterile atmosphere to avoid contamination, skilled labour, controlled observation periods and most important time consumed for the growth of plant which varies from species to species [8]. To test every small alteration in the nutrient media all the resources has to be spent and the tissue culture process has to be repeated until the targeted result is obtained. This implies a simple experiment can take weeks or months of a researcher's time to first figure out the optimum plant stability growth condition and then continue the further part of the targeted experiment, initially aimed by the researcher. Along with the mentioned possibility, if at all human error in performance and care occurred then the experiment has again to be repeated, implying more resources to be spent which may not be cost effective and prolong the research time.

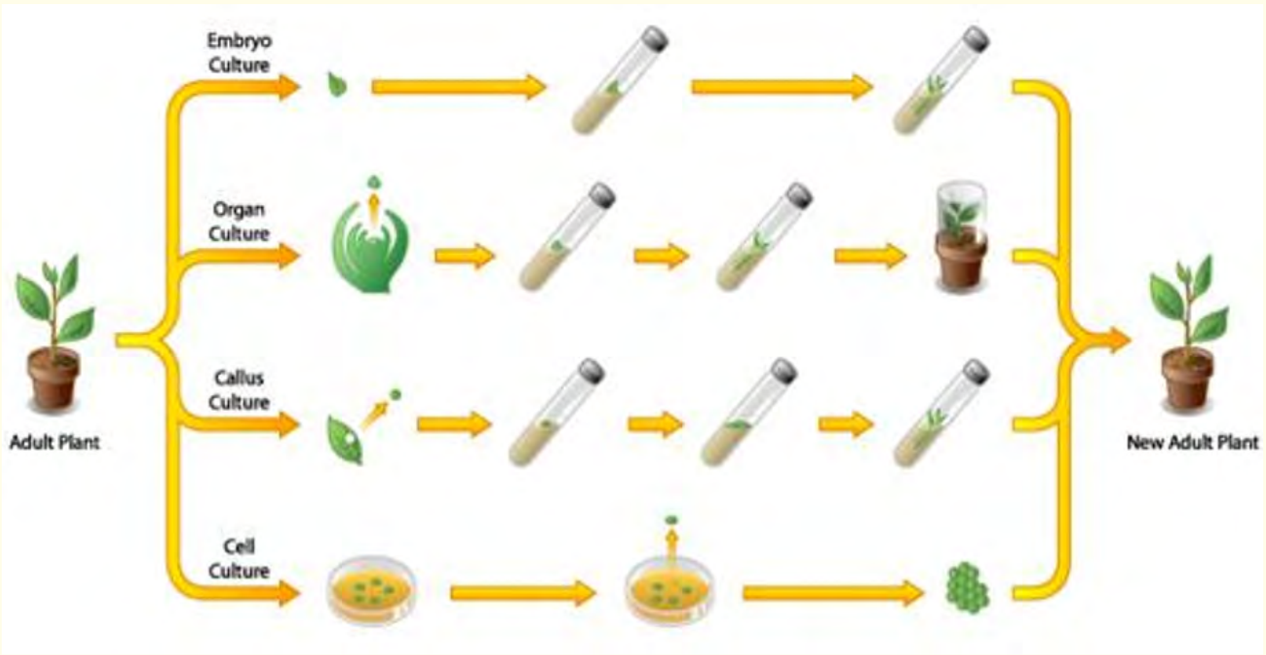


Figure 4: Steps in Plant tissue culture. Image Courtesy: TNAU Agritech.

Plant Tissue Culture Procedure

Basic Steps in plant tissue culture starts form choice of explant. Explant in biology is cell, organ or piece of tissue which can be used to transfer and regrow into whole specimen on adequate supply nutrient medium in vitro. Plant have the property of totipotency which is capability of cell to divide and differentiate to form a working specimen [9]. Thus, in case of plants, explant can be taken from various parts and portion of plant's shoot, steams, leaves, flowers, roots or any undifferentiated plant cell mass. This is then exposed to various types of nutrient media to explore the property of totipotency [10]. Not all parts of plant grow readily. This varies from species to species and also within plant part, which implies choice of explant is an import factor. Other depending factor include optimised nutrient medium, different regeneration time and exposure of light. Most important part of growth of plant in tissue culture laboratory is that aseptic conditions have to be followed as there is a higher chance of contamination due to competition to consume nutrient media by other microbes [11].

After successful growth of plant in tissue culture medium in laboratory, the plant is grown further by exposing it to natural light and environment [12]. This process is termed as hardening. When a tissue culture plant accepts the natural environment then it is transferred into a pot with soil or land directly. The overall process consumes several weeks of time depending on factors like choice of plant species, choice of explant, and skilled labour to handle the tissue culture protocol.

Conceptual Use of ANN for Optimum Nutrient Media Composition

A way to reduce the time and resources required in plant tissue culture consumption, is by using an ANN and by making an effort to carry out the plant growth in various nutrient concentration experiment only once globally. Then making this result obtained by tissue culture experiment available as peer reviewed input data to be used all over the world. Now this data can be used in any standard computer using ANN compilers. Any required targeted changes can be modelled into the ANN and Change in input and output can be observed by running the experiment on simulation mode. The behaviour of plant growth in absence, excess or optimal concentration of any nutrient media can be studied [13]. This simulation can be done in minutes and would save the time and resources of vast number of scientist all over the globe performing individual experiment by tradition trial and error methods [14]. When satisfied by the simulation, the actual plant can be grown in the laboratory by using the results obtained from the simulation having already known the optimum and exact nutrient media required for the growth of the plants [15,16].

Current Experiments Done Globally

ANN is used in various chemical engineering and biotechnology computation process and been implemented as a successful computational model instead of carrying out each process individually [17]. In the year 2005, Research result has been accepted on the

topic use of ANN for Corn and Soybean Yield Prediction performed by University of Maryland, USA [18]. Year 2009 marked the Prediction of average regional yield and production of Wheat in Argentine Pampas using ANN in Argentina [19]. In Year 2011 Authors of this paper i.e., we performed to study and replicate the experiment in Bioinformatics laboratory, Nagpur University, India as our Final Year Research Thesis on Optimization of Medium Composition for Thermo-stable Protease Production using ANN. This was originally performed and published in year 2008 at African Journal of Biotechnology [20]. Our observation and result closely matched that of the original experiment proving that Computational technique using ANN simulation can be replicated in any part of the world using different software or compliers once base research data is generated.

Materials and Methods

Materials Needed

A system with Basic ANN compilers is needed to design and run the computing. Some of the top names include Neural Designer, Neuroph, Darknet, Deep Learning Kit, DTERG, Synaptic and Torch.

Get Data: Graph Designer. This software can be used if user wishes to do the reverse of plotting a graph. Get data can create data points from the actual graph and present you with numerical values corresponding to the points in graph.

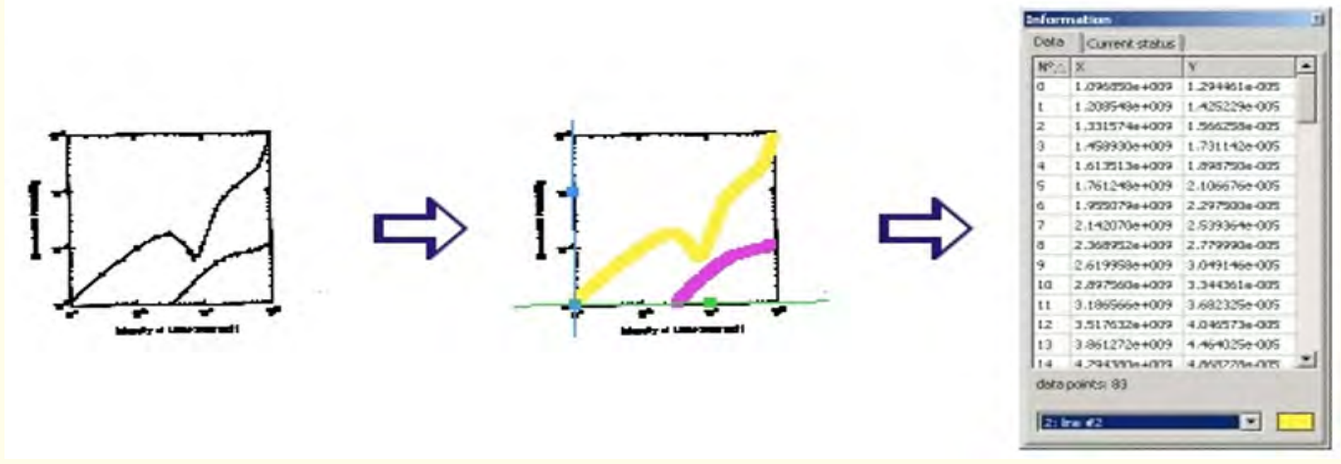


Figure 5: Get Data Graph Designer working Diagram. Image Courtesy Get Data Website.

Research Data: Actual Research Data is needed as input variable and corresponding output result. The data is required to be feed to teach the ANN so that computer compilers can learn the initial phase of the experiment data dependency. This data can be obtained by performing the actual experiment physically for the first time and/or requesting the authors of various papers published already to use their data for your research at the initial stage [21]. Traditional Plant Tissues culture paper published till date provides only the optimum growth condition data, where the result for the experiment was at its peak. And thus, accepting the new era of ANN requires more and more experiments to be done globally on plant biotechnology, generating and publishing data to be freely available to use by researchers over the globe. A step has already been initiated in this direction in departments like chemical engineering and process biotechnology and only few papers published in Plant tissue culture.

Experimental Methodology

A basic experiment to find the optimum nutrient media composition for the highest yield in plant growth can be done by taking various components of nutrient media as input source for ANN and the result with highest yield as output. Designing of the ANN to be feedback or feedforward mechanism can be determined if the output effects the input at any layer of the ANN. After giving the values of both input and targeted output to the ANN compliers, the ANN is run in the system. The first run of the system gives us an actual vs predicted result of the ANN in use. Higher the number of input variables used, higher is the accuracy. Also care should be taken while entering the data as ANN being Black Box model only works on pattern recognition of Input and Output data. Error in feeding any input value may result in accuracy of prediction of the ANN.

Run order	Experimental values				Protease activity (U/ml)
	Corn Starch	Soyabean meal	Glucose	Yeast extract	
1	12.0	6.0	3.0	3.0	5233
2	12.0	6.0	3.0	5.0	4921
3	12.0	6.0	5.0	3.0	4096
4	12.0	6.0	5.0	5.0	4350
5	12.0	14.0	3.0	3.0	5470
6	12.0	14.0	3.0	5.0	5236
7	12.0	14.0	5.0	3.0	5680
8	12.0	14.0	5.0	5.0	5514
9	28.0	6.0	3.0	3.0	5896
10	28.0	6.0	3.0	5.0	5364
11	28.0	6.0	5.0	3.0	5299
12	28.0	6.0	5.0	5.0	4703
13	28.0	14.0	3.0	3.0	4959
14	28.0	14.0	3.0	5.0	5231
15	28.0	14.0	5.0	3.0	4669
16	28.0	14.0	5.0	5.0	3008
17	20.0	10.0	4.0	4.0	6803
18	3.5	10.0	4.0	4.0	5142
19	36.5	10.0	4.0	4.0	4407
20	20.0	1.8	4.0	4.0	5397
21	20.0	18.2	4.0	4.0	4787
22	20.0	10.0	1.9	4.0	5687
23	20.0	10.0	6.1	4.0	4536
24	20.0	10.0	4.0	1.9	4937
25	20.0	10.0	4.0	6.1	5445
26	20.0	10.0	4.0	4.0	6630

Table 1: Experimental Data Run Order we used to test working for ANN in year 2011, Original contributed by Guangrong., *et al* [20].

To test the working of ANN, we use the research data from experiments of Guangrong., *et al.* on getting the optimum medium compostion for production of Thermo Stable Portease Production. Protease Produciton being the corresponding output targeted result like final yield in plant growth models. This was done by using the Graphical Values converted into numberial data using Get Data application. The final protease activiy calculated in units of U/ml was the data for the output layer of ANN. The output layer depends on the nutrient media compostion of four different sources of Carbon and Nitrogen. Thus we had four different input variable namely Corn Starch, Soyabean Meal, Glucose and Yeast Extract in units g/l. Each of the input variable reacted with each other in different concentrations givng us a different output layer. Input and Output Research Data from Run order 1 till Run order 22 in the table 1 is the the actual exprimental data, and from Run order 23 to 26 is the manual values we entered in the system to pridict the working of the ANN. It can be observed that in real experiment Run 17 gives the maximum protease production by its higherst corresponding activty in Output layer. The same when tested with Run 26 gave us the closely matching result as expected for the highest activity, giving the input in Run 17 is the optimum medium compostion for the sample data is learned by the ANN. More input data would provide better and accurate result.

Result and Discussion

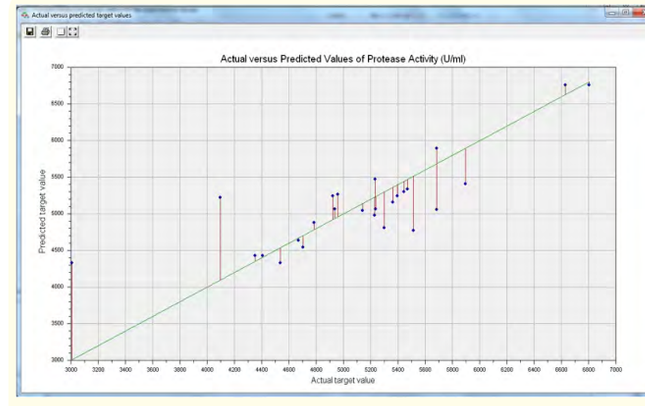


Figure 6: Actual vs Predicted target value of ANN.

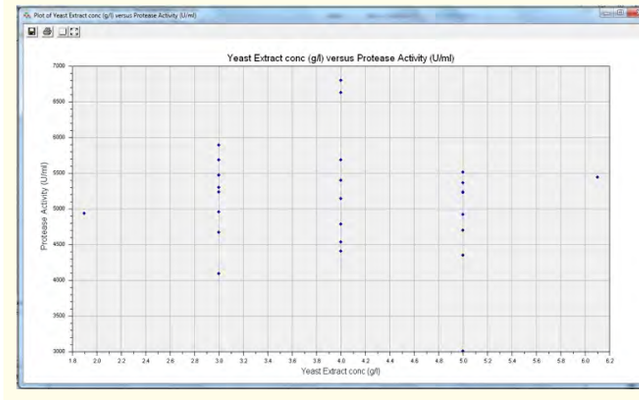


Figure 7: Screenshot of an Example Graph Produced by DTERG Software Comparing one variable (Yeast Exact conc) vs. final product output (protease activity).

The output of ANN produces various multi-dimensional graphs which are difficult in representation on 2-dimensional plane having x and y axis. Figure 7 shows such an example where Yeast Extract (one of input variable) concentration is plotted in x axis and Protease activity (corresponding result) on y-axis. This is because every one type of concentration of input effect the other type of concentration of input which in turn effects the results. Thus, for our observation and understanding Graphs can only be made on two-dimensional plane to compare and observe how either one input variables corresponds and effects to other input variable type. Or how one input type affects the overall output. When Graph are plotted having the increasing one variable concentration on x axis and increasing other variable or result values on y-axis, it produces all the dots of interaction which are observed while computing. The result sometimes is seen as random dots in the 2-Dimensional plane which does not form any linear line. An observer must understand at this stage that the nonlinear or random dots in a graph are produced due to other input variable (corn starch, glucose and soya bean meal in above example) are simultaneously acting on the reference variable of observation and causing the placement of dots at a certain point in the graph. In the 2-Dimensional graph the only point of observation can be made is the value where highest yield or interaction is observed and its average gives the concentration where output yield is maximum. These values from Graphs can be converted into numbers of its highest accuracy by using Get Data software or by simple observation techniques comparing x-axis to y-axis.

Another example results of various concentration for the Yield production of corn and soybean using ANN can be studied in research paper published on 11 September 2004 by University of Maryland USA [18]. Use of ANN in Plant tissue culture is still a developing concept and thus very few papers can be found which have the data as required by the use for ANN compliers. Thus, for other plant species more physical experiments can be carried out and data can be published which will benefit the ease of using ANN by scientist of the future in Plant Biotechnology and Agricultural Industry.

Conclusion

Artificial Neural Network is usually used to model complex relationships between various input and output and also to find patterns in data. A neural network can perform task that linear computational programme cannot. Thus, an ANN refers to a multilayer perception network which consist of input layer, hidden layer and an output layer.

To find the optimum concentration of nutrient medium require for high yield plant growth, various concentration of nutrient media is used as input into the input layer of the ANN and also corresponding result yield is fed as output to the concentration of respective input. The ANN when run on these data performs the co-relation of input vs. output and learns the dependency of each other. At the end of learning phase, the concentrations of highest yield can be selected and used in actual experiment as they are the optimum concentration of nutrient media for highest possible yield. In the same experiment, increase in concentration of any nutrient medium can also be studied to cause what changes it brings to the final yield by simulation growth. The ANN also determines the accuracy at which it works and thus relicense to the result can be interpreted.

Using ANN in Plant Biotechnology and Agriculture industry will save various resources for the experimental part of research and collective data sharing will make growth of industry at a rapid rate to suit the constant rise in demand of the increasing population.

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