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Osman Altay
Manisa Celal Bayar University, Department of Software Engineering, Manisa, Turkey

Inanc Ozgen
Firat University, Department of Bioengineering, Elazig, Turkey

Corresponding Author:
Osman Altay
Manisa Celal Bayar University, Department of Software Engineering, Manisa, Turkey

Predicting the LD50 values of two different vinegars whose insecticidal effect was determined by the spraying method against *Tribolium confusum* Jacquelin du val (Coleoptera: Tenebrionidae) using different artificial neural networks models

Osman Altay and Inanc Ozgen

Abstract
Tribolium confusum Jacquelin du Val pest, which has a wide spread around the world, causes significant damages to stored products. It has of great importance to combat pests such as *Tribolium confusum* Jacquelin du Val with alternative methods rather than using chemicals. Among these methods, carbonization-based by-products are used in pest control. For this purpose, it is important to eliminate the chemical effects by minimizing the duration of the experiment with artificial intelligence methods in order to popularize the use of wood and hazelnut vinegar, which are natural methods. In this study; artificial neural network methods were applied to predict the LD50 value by applying different doses of nut vinegars and wood vinegars whose insecticidal effect was determined by the Spraying Method against *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae). This study is the first to predict the LD 50 value using machine learning methods. Three different models have been proposed to predict the LD50 value with the classification methods. Multi-layer perceptron (MLP) and evolutionary-based, genetic algorithm with neural network (GANN) and evolutionary product unit-based neural network (EPUNN) methods were used to predict the LD50 value. 10-k cross-validation was used to evaluate the models designed in the study. It has been shown that the LD50 value can be predicted with the designed models. EPUNN algorithm performed 6.11 better than MLP and % 2.02 better than GANN.

Keywords: LD50, artificial neural networks models

1. Introduction

Product losses caused by pests in stored products vary between 10-40 percentage. Among these pests, *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) has an important place. This kind has a wide distribution in the world (Park, 1934) [21]. This kind causes significant damage to many stored products (Hill, 1990) [12]. Alternative methods of pest control to chemical control are also important, and many studies on entomopathogens and plant extracts have been conducted around the world (Benzi *et al.*, 2014; Komaki *et al.*, 2017; Saidana *et al.*, 2007; Stamopoulos *et al.*, 2007) [5, 16, 22, 24]. In addition, many alternative methods of combating pests such as the use of radioactive elements, low and high-temperature applications, and adjustment of relative humidity have been used in recent years in pest control (Boylu, 2020) [7]. Some studies have also been conducted on the effects of wood vinegar on pests, diseases, and weeds (Koc, 2020) [15]. In the applications made in the wheat agroecosystem, it has been determined that wood vinegar reduces the total arthropod population and increases the population of predator arachnid species. It has been determined that this change is due to the repellent smell of wood vinegar (Koc, 2020) [15]. In the study conducted to determine the insecticidal effect of different concentrations of wood and hazelnut vinegar against *Tribolium confusum* under laboratory conditions, it was determined that there was a statistically significant difference in mortality rates compared to control applications. In this study, acute toxicity was investigated by both spraying methods (Ozgen *et al.*, 2018) [25]. In determining the LD 50 values of different chemical and plant origin extracts made related to the pest, Polo-PC probit package program and LC / LD50 and LC / LD90 values and confidence intervals are analyzed (Finney *et al.*, 1948) [11]. In addition, LD 50 mortality Finney and LD 50 mortality Miller Tainter methods are mostly used in LD 50 calculations and the MATLAB interactive computer program is also adapted to these calculations (Arambašić *et al.*, 2015) [4].

In many studies, principal component analysis (PCA) is performed with the help of Gen Stat statistics software (Alkan, 2020) [1]. In entomology, machine learning methods are generally used in areas such as identifying pests with neural networks including photo analysis, detecting damage situations with photo analysis of warehouse pests, identifying pests according to their damage in cultivated plants for the use of farmers, and monitoring diseases and pests with remote sensing method (Boniecki *et al.*, 2015; Jiao *et al.*, 2020; Li *et al.*, 2020; Zhang *et al.*, 2019) [6, 14, 17, 27].

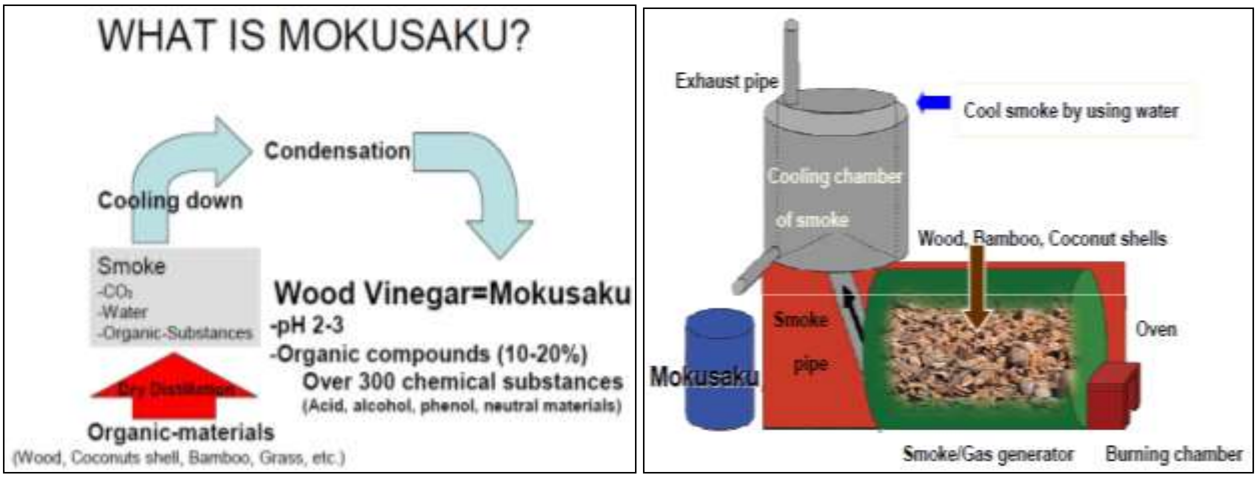
When the literature is reviewed, there are no studies to predict the LD50 value using machine learning methods in insect species that are storage and storage pests. One of the main advantages of machine learning methods is that they have the ability to autonomously solve large nonlinear problems using data sets from multiple different sources (Chlingaryan *et al.*, 2018) [9]. Neural network (NN) is one of the basic machine learning methods applied in almost every field of science. Multi-layer perceptron (MLP), which is a sub-class of NNs, has been chosen in many studies because of its easy programming, high-quality results and universal approach (Curteanu and Cartwright, 2011) [10]. Different versions of NNs have been developed using evolutionary algorithms. Evolutionary algorithms are used to better perform various tasks such as link weight training, architectural design, and learning rule (Yao, 1999) [13]. The Genetic algorithm with neural network (GANN) has been proposed to improve the architectural design process of neural networks using the genetic algorithm (Miller *et al.*, 1989) [20]. In addition, in a different study, an evolutionary algorithm was used to determine the design of the product unit based neural network, a special type of NN, and to determine the coefficients of the designed model (Miller *et al.*, 1989) [20]. Evolutionary programming optimization method, one of the evolutionary algorithms, is used in the evolutionary product unit-based neural network algorithm (EPUNN). Among these methods, especially MLP is successfully used in studies such as indoor and outdoor temperature of greenhouses, indoor air relative humidity, wind speed and protection from frost effects of indoor plants (Castañeda-Miranda *et al.*, 2017) [8], and the instantaneous thermal efficiency (ηith) of a solar energy using weather and operational data (Mashaly *et al.*, 2016) [19].

The aim of this study is to predict LD50 values by classification method using different artificial neural network algorithms. As far as we know, this study on predicting the LD50 value using machine learning methods is the first. Three different machine learning methods were used to predict the LD50 value. These are MLP, GANN and EPUNN algorithms. In section 2, the data set used in the study and the experimental acquisition of the data set is explained in detail. In section 3, artificial neural networks and evolutionary-based artificial neural networks and metrics used in the evaluation of classification methods are explained in detail. In section 4, parameters and results of the algorithms used in the study are given and the results are discussed. Finally, section 5 summarizes the main findings.

2. Data Acquisition

In the material of the study; *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) adults, flour and bran mixtures, nut vinegar obtained by carbonization of hazelnut shells with the carbonization approach, and wood vinegar obtained by carbonization of broiler chicken breeding waste were used. *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) adult is shown in Figure 1. Hand Flow Chart of Hazelnut and Chicken Vinegar and Simple demonstration of the derivation of vinegar are shown in Figure 2. Studies; It was conducted at 65% humidity and 25C ± 2 C °. A mixture of wheat flour-wheat bran at a ratio of 2:1 was used as a nutrient for cultivation.

Fig 1: Habitus of *Tribolium confusum* Jacquelin du Val Adult



a b

Fig 2: Hand Flow Chart of Hazelnut and Chicken Vinegar b) Simple demonstration of the derivation of vinegar

T. confusum adults produced in the laboratory (1-part flour and 1-part bran in culture vessels) were taken with the help of an aspirator and placed in application containers. The solutions (3% and 5% for hazelnut and chicken vinegar) prepared in three sprays (10 each) were applied on the individuals placed on the filter papers by placing individual filter papers in each of the adjusted containers and the individuals were dried for ten minutes expected. Each culture dish was placed in five different positions (5, 10, 15, 20 and 25 cm from the light) in an air conditioning cabinet (65% humidity, 25C ± 2 ° C) where controlled conditions were provided. For the control group; Pure water was sprayed on the samples. To each group; On the same day, the same amount of flour-bran mixture was given as food and counts were made on the 1st, 7th, 14th and 21st days of the applications. The counts continued until the death of 50% of the individuals. In the study, an air conditioning cabinet with core ID-600 branded, 2500-3500 lux fluorescent lighting, programmable day and night temperature and hygrometer was used. In the study, a lighting period with 16 hours light and 8 hours dark conditions was used. The lighting of the climate cabinet is designed to be opened and closed from the side. The exterior and interior view of the climate cabinet is shown in Figure 3.

a

b

Fig 3: a) Climate Cabinet b) Containers in the climate cabinet at 5, 10, 15, 20 and 25 cm from the light.

3. Artificial neural networks

In this section, the artificial neural network models and evolutionary algorithms used in the study are explained in detail. In addition, the methods and metrics used in the evaluation of the designed artificial neural network models are explained.

3.1 Multi-Layer Perceptron

Artificial neural networks (ANN) are an intelligent and nonparametric mathematical model inspired by the nervous systems. ANN is successfully applied in problems such as classification, regression, pattern recognition. ANN is successfully applied in many fields such as medicine, construction, materials. The learning process in ANN has a great effect on the performance of the designed model. Feed forward neural networks are a well-known and widely used class of ANN-based neural models. A series of processing elements called neurons form the basis of feed forward neural networks. Neurons are scattered across multiple stacked layers within the designed model, where each layer is linked to the next layer.

MLP is one of the most widely used in feed forward neural networks. It is expected to achieve a better success rate than models designed with MLP due to its ability to make nonlinear matches between input and output in MLP. The basic architecture of MLP constitutes the first layer input layer fed with input variables and the last layer constitutes the output layer. The layers between the input and output layers are called hidden layers. The connection between the layers consists of values between [-1, 1] which are called weight. Each node of the MLP is processed with the addition and activation function. The addition process is shown in the Eq. (1).

$$S = w_i + \beta \quad (1)$$

Here w value shows weight values and β value shows bias value. There are different equations for the activation function in ANN models. As in ANN, there are different activation functions in MLP as well. The activation function directly affects the MLP's ability to obtain solutions, efficiency and speed (Ulas *et al.*, 2020) [26]. There is no general rule to choose the activation function and it is determined by trial and error. Tanh activation function is used in this study. The basic MLP structure is shown in Figure 4.

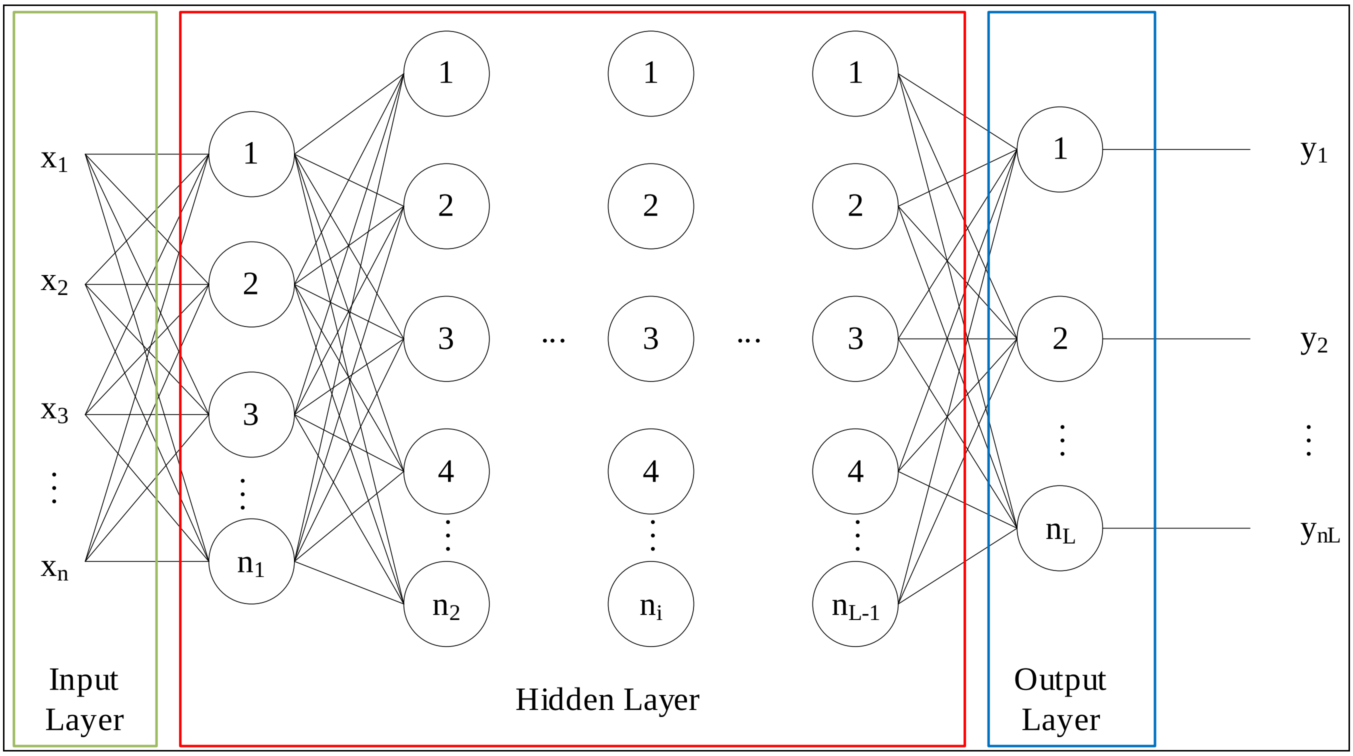


Fig 4: Architecture of MLP

3.2 Evolutionary based artificial neural networks

Evolutionary algorithms are in the class of population-based stochastic search algorithms inspired by natural evolution. Evolutionary programming and genetic algorithms are in this class. The common feature of the two algorithms is population-based search strategies. Evolutionary algorithms can usually produce satisfactory solutions in solving large complex problems that produce many optima. Evolutionary algorithms are less likely to catch the local minimum than gradient based search algorithms. Since evolutionary algorithms do not need gradient information, they are suitable for solving problems in situations where this information is not available or where it is difficult to obtain or predict. They are better than many other search algorithms because they can find solutions for problems that do not have a clear or precise objective function (Yao, 1999) [13]. Evolutionary based artificial neural networks consist of three different structures. These are link weights, architectures and learning rules.

3.2.1 Genetic algorithm based artificial neural networks

Genetic algorithm-based artificial neural networks combine two adaptive processes. These are the area of network architecture and individual network learning to evaluate genetic search and selected architecture. Thus, in our method, as in biological systems, learning cycles in individuals are settled for evolutionary cycles in populations. In each learning cycle, one by one artificial neural networks present the concretization of a specific network architecture with input-output values defining the task. The back propagation learning algorithm compares the actual outputs produced with the desired outputs. The network changes the connection weights to achieve the desired input / output values. Each evolution cycle processes a network design population according to their respective fitness values (calculated during learning cycles) to obtain the offspring population of more highly tailored network designs (Miller *et al.*, 1989) [20].

As shown in Figure 5, the connection topology shows the matrix in which the type of connection of each input from neuron to neuron is encoded (8 * 8). The matrices in the connection input are binary as 0 or 1. 0 indicates no connection and 1 indicates connection. The presence of the link indicates that the weight can be changed through learning. It is also shown in Figure 5 that it is transformed into a chromosome for the genetic algorithm. It also shows how the bit sequence is networked in Figure 5. The first formation of connections that could be learned was initiated with small random weights. Given a set of training examples and a binary string representation for possible network architecture, a basic genetic algorithm follows the steps in the Table 1 (Sekkal *et al.*, 2011) [23].

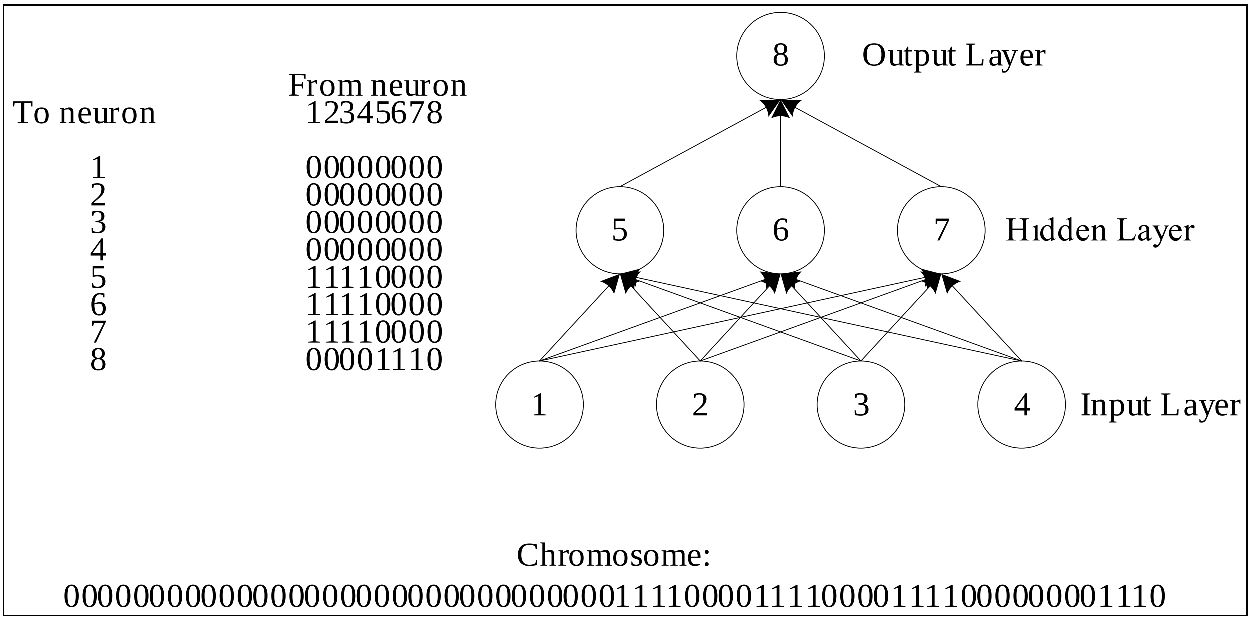


Fig 5: Architecture of Genetic Algorithm Based Artificial Neural Network

Table 1: Steps of Genetic Algorithm

Step 1: Select the size of a chromosome population, the probabilities of crossover and mutation, and define the number of training periods.
Step 2: Define a fitness function to measure the performance or consistency of a single chromosome in general. The accuracy of the network should be based not only on its accuracy but also on the speed, size and complexity of learning. However, the performance of the network is much more important than its size, and therefore the fitness function is still mutually definable as the sum of squares of errors.
Step 3: Generate a random starting chromosome population.
Step 4: Convert the code of an individual chromosome into a neural network. Our sine networks are restricted to feed forward, ignore all feedback links specified on the chromosome. Set the net starting weights to small random numbers. Train the network on a set of training samples for a specified number of periods using the backpropagation algorithm, calculate the sum of the squared errors, and determine the fit of the network.
Step 5: Repeat step 4 until all individuals in the population have been considered.
Step 6: Select a pair of chromosomes for mating with a probability proportional to their fitness.
Step 7: Create a pair of offspring chromosomes by applying the crossover and mutation of the genetic operator. A crossover operator selects a random array of rows and alternates the corresponding rows between the two parents, creating two offspring. A mutation operator detects one or two bits on the chromosome with a slightly lower probability.
Step 8: Insert the formed offspring chromosome into the new population.
Step 9: Repeat step 6 until the size of the new chromosome population is equal to the size of the first population, and then replace the first (parent) chromosome population with the new (offspring) population.
Step 10: Go to step 4 and repeat the process until a certain number of generations are considered.

3.2.2 Evolutionary product unit based neural networks

EPUNN is the evolutionary programming integrated into the product-unit neural network, which is one of the feed forward neural networks. The advantage of the product-unit neural network is that it increases the information capacity and can create high-order input combinations. The presence of combinations of high-order inputs, on the other hand, can cause small errors that may occur during the training of the network to cause larger errors in the progressive structure. The basic structure of the product unit neural network is shown in Figure 6. One input layer consists of k nodes. Each input variable has a node. There is a hidden layer consisting of m nodes and an output layer from the number of j nodes for each class. There is no link between the nodes of a layer. Also, there is no connection between the input and output layers. The activation function of the node in the jth iteration of the hidden layer is calculated with the following Eq. (2) (Martínez-Estudillo *et al.*, 2008) [18].

$$B_j(x,w_j) = \prod_{i=1}^{k} x_i^{w_{ji}} \tag{2}$$

where w_{ji} $w_j = (w_{j1}, \dots, w_{jk})$ is the weight of the link between input node i and hidden node j. The activation function of the output layer l is given Eq. (3).

$$\beta_0^l + \sum_{j=1}^{m} \beta_j^l \, B(x,w_j) \tag{3}$$

where β_j^l is the weight of the link between hidden node j and output node l. β_0^l is the bias value. The transfer function of all hidden and output nodes is the identity function. Thus the transfer function is given Eq. (4).

$$f_1(x;\vartheta_l) = \beta_0^l + \sum_{j=1}^{m} \beta_j^l \, B(x,w_j), l = 1,2,3,\dots,j \tag{4}$$

Here it represents the values of $\vartheta_l = (\beta^l, w_1, \dots, w_m)$ and

$\beta^l = (\beta_0^l, \beta_1^l, \beta_2^l, \dots, \beta_m^l)$.

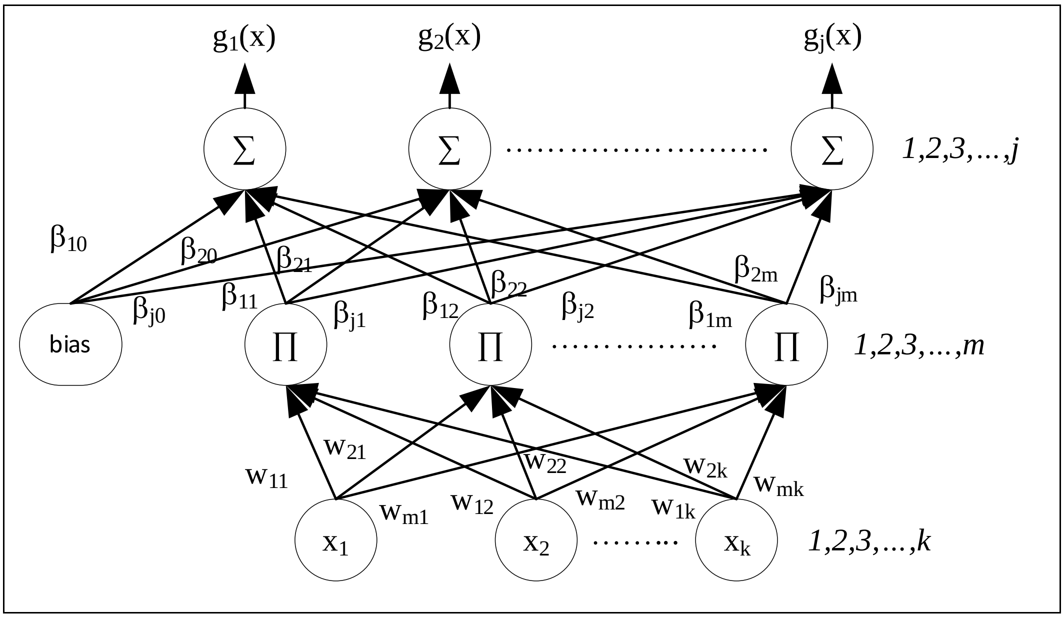


Fig 6: The basic structure of the product unit neural network

In the study, soft max activation function and cross-entropy error function were used to obtain the highest performance rate. But the cross-entropy function used has some disadvantages. These disadvantages; the existence of a large number of local optimum values and the Hessian matrix of the error function is generally uncertain. In addition, the optimal number of hidden nodes for the model is unknown. Evolutionary programming has been used to eliminate these drawbacks. Using evolutionary programming, it is aimed to design the network structure and determine the training weights in the best way. With evolutionary programming, the number of basic functions of the model, associated coefficients and corresponding exponents are determined. Search begins with the first population of product unit neural networks. In each cycle, the population is updated with the population replacement algorithm. In other words, it goes through crossover and mutation processes. Crossover is not used because of the potential disadvantages of artificial networks in their evolution. Because of these features, the algorithm designed is in the evolutionary programming class. The general steps of evolutionary programming are given in the Table 2. The program is stopped if the number of generations is reached or if the variance of the best 10 percent of the population is less than 10^{-4} (Martínez-Estudillo *et al.*, 2008) [18].

Table 2: Steps of evolutionary programming

Step 1: Generate a random population of size N_p.
Step 2: Repeat until the stop criterion is met.
(a) Calculate the fitness of each individual in the population.
(b) Rank the individuals according to their suitability.
(c) The best individual is copied into the new population.
(d) The best 10% of the population individuals replicate and replace the worst 10% of the individuals.
Meanwhile over the population
(e) Apply the parametric mutation to the best 10% of individuals.
(f) Apply structural mutation to the remaining 90% of individuals.

3.2 Classification Evaluate Metrics and Confusion Matrix

The special tabular form used in visualization of the performance of machine learning algorithms used for classification is called a confusion matrix. It is also known as the error matrix. Thanks to the confusion matrix, it is useful to see whether the two classes are mixed with each other and are labeled incorrectly. P in the confusion matrix indicates positive classes and N indicates negative classes. T and F values also show true or false predictions. That is, TP represents true positives, TN represents true negatives, FP represents false positives and FN false negatives (Altay *et al.*, 2020). An example confusion matrix is shown in the Table 3.

Table 3: Simple presentation of confusion matrix

	Predicted 0	Predicted 1
Actual 0	TN	FP
Actual 1	FN	TP

In order to evaluate the algorithms used in this study, 5 different evaluation of binary classification metrics were used (Altay and Ulas, 2019) [2]. The equations of the sensitivity, specificity, precision, accuracy and f1score metrics are between Eq. (5) and Eq. (9), respectively.

$$sensivity = \frac{TP}{TP+FN} \quad (5)$$

$$specificity = \frac{TN}{TN+FP} \quad (6)$$

$$precision = \frac{TP}{TP+FP} \quad (7)$$

$$accuracy = \frac{TP+TN}{TP+TN+FP+FN} \quad (8)$$

$$f1\ sore = 2 \times \frac{precision \times sensitivity}{precision + sensitivity} \quad (9)$$

In the study, it was used to evaluate models based on k-cross validation. It predicts how well predictive models will perform in practice. The data set is divided into k parts. Then, each of these parts is divided into test data (data not used during training) and training data. The results are obtained by applying the test data to models trained with training data that they will see for the first time. An example of 10-k cross validation is shown in the Figure 7.

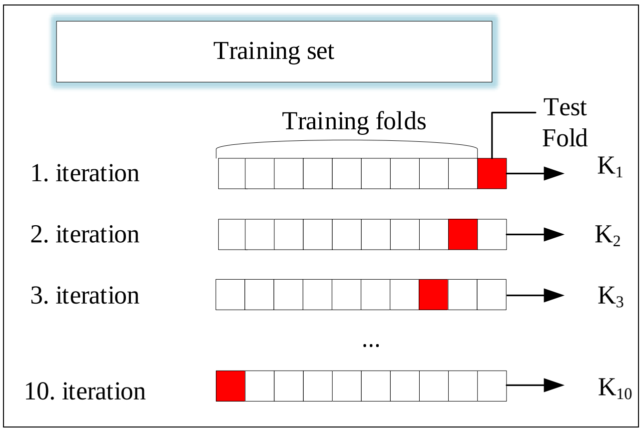


Fig 7: 10-k cross validation

4. Result and Discussion

In this study, three different input parameters are used to predict the LD50 value. The first input parameter is from 3% chicken, 5% chicken, 3% hazelnut and 5% hazelnut vinegar concentrations, the second input parameter is 1, 7, 14 and 21 days, which includes the pest's counting days after the application of vinegar, The third input parameter (1, 2, 3, 4 and 5) consists of the distance of the light source (5, 10, 15, 20 and 25 cm distance to the light, respectively) to the culture vessels containing *T. confusum* individuals where nits are applied. It is the classification of the daily number of *T. confusum* measurements in the vessels treated with vinegar as the output parameter according to the LD50 value.

Three different machine learning methods, MLP, GANN and EPUNN, were used in the models designed to predict the LD50 value. In the model designed using MLP and GANN algorithms, hidden layer 2 and hidden node 15 are taken. Hard limit is used as activation function. In the GANN algorithm, the percentages of initial connectivity, backprop mutation, random mutation and structural mutation values were taken as 0.5, 0.25, 0.1 and 0.1, respectively. In the EPUNN algorithm, the number of hidden nodes was taken as 4 and the product unit was used as the activation function. The number of generations is taken as 400 in both GANN and EPUNN methods developed by evolutionary algorithm.

The 10-k cross validation method was used for testing the designed models. Thanks to the 10-k cross validation method, all the data in the data set are used both in training data and test data. The training set and the data set were randomly distributed. The same training and test data were used in 3 different methods designed for equal comparison.

Confusion matrices of models designed using MLP, GANN and EPUNN algorithms are given in the Table 4. When the algorithms are considered in general, it has been observed that the prediction of the classes where the LD50 value is exceeded is more successful than the prediction of the classes where the LD50 value is not exceeded. Especially the MLP algorithm has achieved a hundred percent success in predicting this class.

Table 4: Confusion matrices

MLP-BP				GANN				EPUNN			
	Predict				Predict				Predict		
		0	1			0	1			0	1
Actual	0	43	14	Actual	0	50	7	Actual	0	50	7
	1	0	31		1	3	28		1	2	29

The results of 5 different binary classification metrics used are shown in the Table 5. When compared according to the sensitivity value, the MLP algorithm gave the best result. While GANN and EPUNN algorithms give good results at Precision, it has been observed that MLP algorithm has a lower performance. EPUNN algorithm has achieved the best success rate in precision, accuracy and f1score metrics. EPUNN algorithm performed 6.11% better than MLP algorithm and 2.02% better than GANN algorithm in f1 score metric used in general evaluation of models designed especially based on classification problem.

Table 5: Binary evaluation metrics results

Algorithm	Sensitivity	Specificity	Precision	Accuracy	F1 Score
MLP-BP	1.0	0.7544	0.6889	0.8409	0.8158
GANN	0.9032	0.8772	0.8000	0.8864	0.8485
EPUNN	0.9355	0.8772	0.8056	0.8977	0.8657

The dose-response relationship of drug responses is important in pharmacology and toxicology. These answers are also very important in determining the effects of pesticides against pests. Predicting these values correctly for safety and efficacy is valuable in determining the effectiveness of the drug. The reliability of studies and analyzes regarding the small number of doses in studies is very low. However, the use of machine learning methods with this study is valuable in terms of decision mechanism and reliability and it is shown that they can be used in practice. Thanks to the optimization of culture containers, air conditioning cabinets, labor and time applied methods, which constitute the breeding environment of the insect used in the study, it has evolved into a process that can be overcome in a short time. Creating this environment and conducting the experiment involves high costs in terms of time, cost and technical manpower. In order to establish an insect culture, 1 progeny of the species must be produced. This type; the pre-mature period of 25-32 days, especially the culture boxes and labor, for the production of reproduction, 10-12 days have passed for mating maturity as a result of oviposition, the average time from the eggs left by the individuals who reach the mature period to the formation of new individuals is 35+ A minimum trial start time of 35+35 = 70 days has been established. A minimum working period of 70 + 21 days (91 hours) was spent for the conduct of these trials, including the change of nutrients, selection of species, administration of drugs, and a daily 1-hour study period and a 21-day administration period. Considering all the disadvantages that occur in the creation of the data set, it seems that the application of machine learning methods in such data sets is of great importance. It has been shown in the study that these disadvantages can be removed and the pest can be combated by organic means.

5. Conclusion

In this study, two different carbonization products of organic origin were found on T. comfusum, which is an important storage pest, in terms of pest control. In dose trials, vinegar concentrations of 3 and 5 percent were applied under controlled conditions. The mortality rates of the individuals were determined daily and the percentage mortality rates LD50 values were determined. The experimentally determined LD50 value has been predicted with the evolutionary-based artificial neural networks and artificial neural networks. Models designed to predict the LD 50 value are MLP, EPUNN and GANN methods. As a result of the study, it has been shown that machine learning methods can successfully predict mortality rates of T. comfusum beetle by using hazelnut and chicken vinegar. As a result of the designed models, EPUNN method gave the best result according to the f1-score value. As a result of the study, the European Union in terms of reducing drug residue in the products and shortening the application period by using artificial intelligence methods in the use of organic products, hazelnut and chicken vinegar, instead of chemical drugs, reducing the time and product residue problems in the fight and integrated combat against storage damages. It is thought to provide an important gain in terms of pessist residue values. It is thought to be a pioneering study on the application of machine learning methods in pessist licensing studies in terms of both organic agriculture-based combat methods and conventional combat methods against other storage and storage pests that are difficult to combat. In addition, it is anticipated that by combining different methods other than the spraying method used in this study, using machine learning methods in both laboratory and field and semi-field trials, pessist impact assessments can be made using machine learning methods.

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