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Prediction of drug release profiles using an intelligent learning system: an experimental study in transdermal iontophoresis

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Abstract

This paper investigates the use of a neural-network-based intelligent learning system for the prediction of drug release profiles. An experimental study in transdermal iontophoresis (TI) is employed to evaluate the applicability of a particular neural network (NN) model, i.e. the Gaussian mixture model (GMM), in modeling and predicting drug release profiles. A number of tests are systematically designed using the face-centered central composite design (CCD) approach to examine the effects of various process variables simultaneously during the iontophoresis process. The GMM is then applied to model and predict the drug release profiles based on the data samples collected from the experiments. The GMM results are compared with those from multiple regression models. In addition, the bootstrap method is used to assess the reliability of the network predictions by estimating confidence intervals associated with the results. The results demonstrate that the combination of the face-centered CCD and GMM can be employed as a useful intelligent tool for the prediction of time-series profiles in pharmaceutical and biomedical experiments.

Keywords: Neural network; Gaussian mixture model; Transdermal iontophoresis; Drug release profile prediction; Face-centered central composite design; Bootstrap

1. Introduction

Artificial intelligence (AI) is a multi-disciplinary field focusing on the study and creation of computing systems that exhibit some form of human intelligence. AI has been widely applied to real world problems recently because it provides a powerful tool to assist our work and also greatly to improve our ability to accomplish work. The main challenge of AI research is knowledge acquisition. This distinguishes AI computing from conventional computing. With the ability to draw inferences from a knowledge base, the computing system can be used as an intelligent

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learning system in various application domains [1,2].

AI encompasses a number of methodologies including neural networks (NNs) [2]. NNs are relatively crude mathematical models based on a physiological understanding of the nervous systems that learn from experience. They emulate a biological neural system that receives inputs from other sources, combines them in some way, performs a general nonlinear operation on the result, and then outputs the final result.

NNs, acted as an intelligent learning system, can provide useful aids in many tasks. It is a powerful tool for learning nonlinear mappings from data [3]. NNs are able to model and map a vector of input variables with a vector of output variables. Moreover, they have the ability to generalize the acquired information, i.e. once trained, the network can process previously unseen data to predict a response [4]. Therefore, NNs require minimum prior understanding of the underlying process or phenomenon before making predictions. In addition, NNs are adaptable and flexible [5] in the way they deal with new and changing environment, and, are relatively easy to maintain. These features make NNs suitable for modeling and solving nonlinear estimation and predictions problems.

Recently there has been an increasing interest in NNs for biomedicine research. In pharmaceutical and pharmacokinetic areas, NNs have been applied to model complex relationships between causal factors and response variables [6–9]. In [6], they compared the performance of a mechanistically-based model and an empirical NN model to describe the relationship between the tissue-to-unbound plasma concentration ratios (*Kpu's*) of 14 rat tissues and the lipophilicity (log *P*) of a series of nine 5-*n*-alkyl-5-ethyl barbituric acids. The results showed that the overall predictive power of the NN model is better than that of the mechanistically-based model. The reason for this is that building the NN model is equivalent to fitting an arbitrary (and, therefore, more flexible) multivariate function to the data samples. On the other hand, building the mechanistically-based model is equivalent to selecting a particular function type for the data samples, due to rigid modeling assumptions. An NN system was reported in [7] to predict peaks and troughs of gentamicin serum concentrations based on a set of empirical data, and the results were comparable with those using nonlinear mixed effect modeling. In [8,9], they demonstrated that a multi-objective simultaneous optimization technique incorporating NNs was useful in optimizing formulae of pharmaceutical responses that are non-linearly related to the process variables.

The approach reported in the above works requires a priori assumption for selecting a mathematical model before applying the NN model to predict coefficients of some parametric equation that characterizes the drug dynamics. Instead of predicting parameters of models or equations that characterize certain drug dynamics, we, however, demonstrate that NNs could be used as a predictor to model and predict the drug release profile directly, in a non-linear time-series form. In this paper, an experimental study was conducted to evaluate the applicability of a NN model, i.e. the Gaussian Mixture Model (GMM) [10], to predict drug release profiles in a time-series form. In NN applications, a good set of training data is of paramount importance for the NN learning algorithm to capture the underlying dynamics of the problem domain. Therefore, in this work, we employed a design of experiment technique in Response Surface Methodology (RSM), i.e. the face-centered Central Composite Design (CCD) [11], to collect a representative set of domain data samples for training the GMM.

The RSM is a useful approach to minimize the number of experiment trials, especially for an unknown system with single or multiple responses in multi-variable systems. Several researchers [12–14] have investigated the use of RSM in drug release profile research. The RSM has been demonstrated to be a useful method to approximate the true system behavior as a function of the formulation and process variables, and to determine the apparent optimum conditions. The work in [12] demonstrated the applicability of regression models based on RSM and NNs to estimate the coefficients of a mathematical model that characterizes the in vitro drug release profile of a hydrophilic matrix capsule system. In [13], the RSM was applied to examine the effects of a

number of pharmacokinetic variables on a tablet based on vegetable extract. In [14], a study on iontophoresis of thyrotropin-releasing hormone facilitated by periodically mono-phase-pulse current across excised skin was conducted. The optimum operating conditions were achieved via the RSM by systematically adjusting the corresponding variables in experiments.

In this paper, we demonstrate the combination of RSM and NNs for the prediction of drug release profiles. The main objective of using face-centered CCD is not to search for optimum process conditions. Rather, we examine the feasibility of applying the face-centered CCD to design a set of experiments such that a representative data samples for training the GMM can be collected. The rationale is to exploit the powerful and flexible data learning properties of the GMM to model and predict the drug release profiles given a well-represented data space constructed by experimental samples collected through the face-centered CCD.

2. Experimental

2.1. Methods

2.1.1. The Gaussian mixture model

The GMM is based on the structure of the Radial Basis Function Networks. Fig. 1 depicts the network structure of the GMM. The network consists of three layers of nodes, i.e. the input layer, hidden layer (composed of radial basis function units), and output layer. Each radial basis function unit has a vector of center, c. The links between the hidden layer nodes and output layer nodes are the network weights, w. The output layer performs a linear combination of the outputs from the basis nodes. Thus the output of the network is:

$$y(\vec{x}) = \sum_{i=0}^{H} w_i \phi(a_i) \quad (1)$$

where H refers to the number of nodes in the hidden layer, and vector $\boldsymbol{a}$ refers to the distance of the input vector $\boldsymbol{x}$ to each of the centers in the hidden layer. In generally, the Euclidean norm is used to compute distance:

$$a_i = \sqrt{\sum_{j=1}^{d} (x_j - c_{ij})^2} \quad (2)$$

where d is the dimension of the input vector $\boldsymbol{x}$. In this network, the radial basis function, $\phi(a_i)$ is Gaussian function:

$$\phi(a_i) = \exp - (a_i^2/\sigma_i^2) \quad (3)$$

where σ_i is the width of the basis function.

Basically, the GMM uses a two-stage training algorithm to determine the network weights using the training data set. The advantage of this two-stage training is that the non-linear representation given by first layer of the network can be determined using a large quantity of unlabeled data, leaving a relatively small number of parameters in the second layer to be determined using the labeled data. Therefore, the training speed for this network is relatively faster than the conventional multi-layer Perceptron network trained with backpropagation [15]. Note that the labeled data samples consist of known (input–output) vector pairs sampled from an unknown model, while the

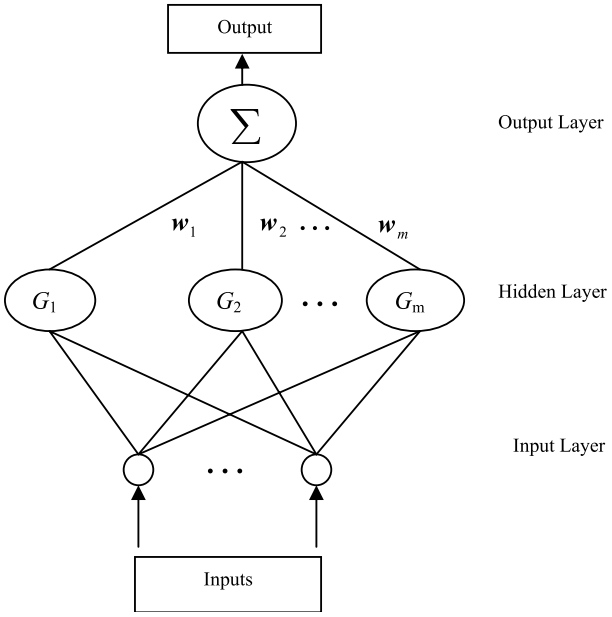

Fig. 1. The architecture of the GMM.

unlabeled data samples consist of observed input samples exclusively. Therefore, the first stage of training is termed unsupervised as only the input vector is provided for training process, while the second stage is considered as supervised training as the desired output vectors are specified for the input vectors.

In the first stage of training, the K-means clustering algorithm [16] is used to determine the initial center points. The center points are then optimized using the expectation maximation (EM) algorithm [17]. The basis function widths, σ_i, are tuned by setting to the maximum inter-center square distance after the network is trained by EM-algorithm. In the second stage, the generalized cross-validation (GCV) [10] method is used to determine the weights between the hidden and output layers. Note that the above description is a concise account on the network structure and learning strategy of the GMM; a detailed explanation with mathematical equations of GMM can be found in [10].

2.1.2. The bootstrap method

Confidence intervals are regarded as the range of values that are likely to contain the true value of a population parameter. In this study, the bootstrap method [18] is used to estimate the confidence intervals of the network predictions. Bootstrap is a form of randomization test—one of the alternatives to exhaustive re-randomization. Bootstrap involves generating subsets of data on the basis of random sampling with replacements as data are sampled. This method has no constraint upon the number of times that a datum may be represented in producing a re-sampled subset. The size of re-sampled subsets may be fixed arbitrarily which is independent on the parameter of the experimental design, and may even exceed the total number of data samples. Therefore, the bootstrap method can be used for estimating confidence interval of parameters when the underlying distribution function of the parameters is unknown.

The produce of bootstrap in estimating confidence intervals is as follows.

1) Collect a sample (x_1, x_2, …, x_n) with mean $\hat{\alpha}$ that defines a discrete distribution function $\hat{G}$ having mass ($1/n$) at each of the n sample points.
2) Draw, with replacement, a sample randomly from $\hat{G}$. The distribution of each x^{*i} in the bootstrap sample is $\hat{G}^{*i}$, x_1^{*i}, x_2^{*i}, …, $x_n^{*i} \sim \hat{G}^{*i}$.
3) Calculate the new mean, $\hat{\alpha}^{*i}$.
4) Repeat steps 2 and 3 m times to obtain $\hat{\alpha}^{*1}$, $\hat{\alpha}^{*2}$, …, $\hat{\alpha}^{*m}$.
5) Sort the bootstrap mean values in ascending order, $\hat{\alpha}^{*1} < \hat{\alpha}^{*2} < \cdots < \hat{\alpha}^{*m}$.
6) Calculate the confidence intervals from the sorted list. The $100(1-a)\%$ 100 confidence interval is ($\hat{\alpha}^{*c_1}$, $\hat{\alpha}^{*c_2}$), where $c_1 = ma/2$ (upper limit), and $c_2 = m - c_1 + 1$ (lower limit) (e.g. $a = 0.05$ for 95% confidence interval).

2.2. Procedures

In the process of developing a new form of drug, selecting an appropriate formulation of various drug compositions and the associated process variables to meet the required release profile is a very time-consuming task. The usual way is to conduct a series of physical experiments, based on trial-and-error and experience of the drug formulator, with different process variables in order to obtain a desired profile. This is indeed a difficult and laborious task. In this paper, we propose that an intelligent prediction tool, such as a NN trained using existing profile data from different experiments, could be used to predict a new drug release profile of unknown process variables. Once a satisfactory profile is found, confirmation tests can then be conducted to ascertain the profile experimentally. This could significantly reduce the time and effort required in drug formulation tasks. To demonstrate the idea, an experimental study in Transdermal Iontophoresis (TI) has been conducted.

Iontophoresis is an alternative drug delivery method based on electrically induced transport of drug molecules. It has been found to be an effective and painless method of delivering medication. TI is the facilitated transport of drug molecules through skin with the application of

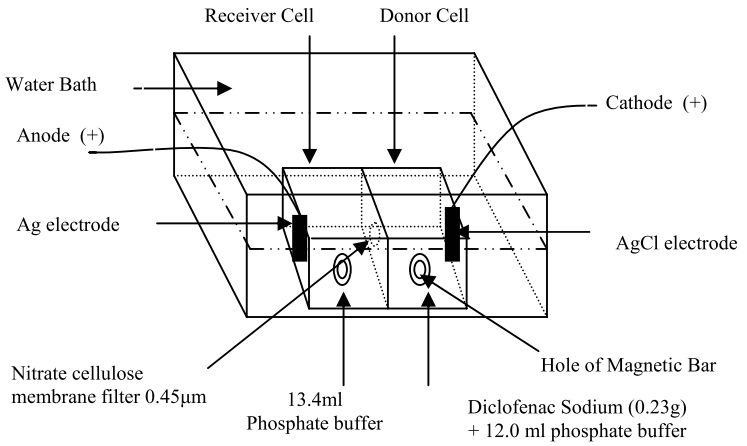

Fig. 2. A diffusion cell with water bath constructed for the TI experiment.

an external electrical field [14,19]. It is a new, non-invasive technique for delivering drugs to patients. By using electrical forces, it is able to increase the penetration rate of drug molecules through the stratum corneum—the main barrier of drug transport. Research works on TI have been focused on the area of developing and testing of chemicals or drugs transporting across the skin/membrane [20–22]. In this paper, we examined the delivery rate of Diclofenac Sodium ($C_{14}H_{10}Cl_2NO_2Na$, MW = 318.15) [23] facilitated by direct current across a synthesis membrane in the first 7 h. Diclofenac Sodium was selected in this study because of its anti-inflammatory, analgesic, and antipyretic properties. It is normally recommended for chronic inflammatory conditions such as rheumatoid arthritis, and osteoarthrities, and for the treatment of acute musculo-skeletal pain.

A two-compartment (donor and receptor) diffusion cell was designed and fabricated for the study, as shown in Fig. 2. The drug was prepared in the form of saturated solution. The phosphate buffer consisted of 0.178 M of disodium hydrogen orthophosphate, 0.016 M sodium dihydrophosphate and 0.12 M sodium chloride adjusted to desired pH with 1 M NaOH. Three independent variables, i.e. pH of the buffer, the ionic strength of the buffer, and the magnitude of electrical current, were systematically changed during the experiment to investigate the effects of the variables towards the transdermal delivery rate of the drug. The TI experiment was conducted according to the following procedures:

1) The diffusion cell with a nitrate cellulose membrane filter was assembled together by using silicone sealant, and left overnight to dry off.
2) The negatively charged permeant solution was placed in the cathode (negative) compartment, and the electrolyte surrounded the anode (positive).
3) The water level of the water bath was set above the solution in the diffusion cell in order to ensure that the temperature was maintained constant.
4) Electrodes were connected to the power supply, with the current adjusted to the desired value for each experiment.
5) The experiment commenced by pouring in the donor and receptor solutions.
6) Samples (0.5 ml) of the receptor solution were collected hourly by using Eppendorff micropipet (volume range = 100–1000 μm), for 7 h.
7) Each sample (0.5 ml) was diluted to 10 ml, in the same phosphate buffer by using 10 ml volumetric flask.
8) The 1 ml solution, after the first dilution, was diluted for the second time to 10 ml, in the same phosphate buffer by using 10 ml volumetric flask.

Table 1
Factors and levels of the experiments

Factors	Levels		
	Low (−1)	Centre (0)	High (+1)
pH	7	8	9
Ionic strength	0.100	0.145	0.19
Current	0.5	1.0	1.5

9) Concentration of each sample was determined by using ultraviolet spectrophotometer.
10) Steps 1–9 were repeated for each experiment condition as shown in Table 1. Each experiment condition was repeated four times.

2.2.1. Design of the experiment

For any unknown system, the underlying system dynamics or the relationships among the process variables are often difficult to deduce. The RSM is an experimental design procedure that can be employed to determine the optimum conditions of various process variables simultaneously. The first step in RSM is to search for the optimum condition based on factorial design or factional factorial design [11]. The method of the steepest ascent is used in this step to move sequentially along the steepest path, i.e. in the direction of the maximum increase in the response. Of course, if minimization is desired, then it is considered as the method of the steepest descent. After determining the optimum region of the process condition, a suitable experimental design method such as face-centered CCD will be selected to test the optimum condition.

In the present study, the face-centered CCD approach has been employed to determine the interaction between the process variables and the process response. One of the reasons of choosing the face-centered CCD is that we know the properties of the drug candidate (Diclofenac Sodium) and can ensure the region of operability that is used to cover the region of interest in experimental design. In addition, the face-centered CCD is simpler and requires fewer level settings of each variable when compared with other types of CCD, e.g. rotatable CCD [11]. Note that one can

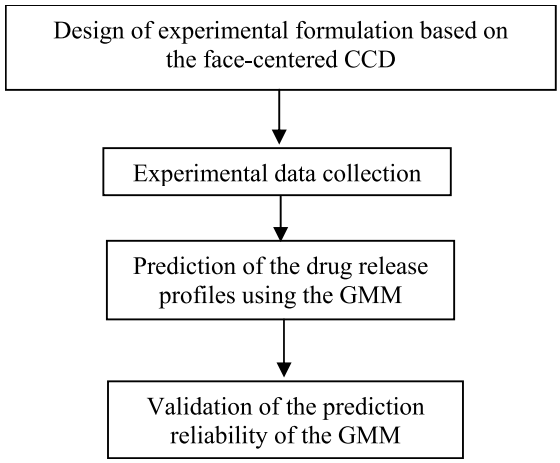

Fig. 3. Procedure of the experimental study using the face-centered CCD coupled with the GMM.

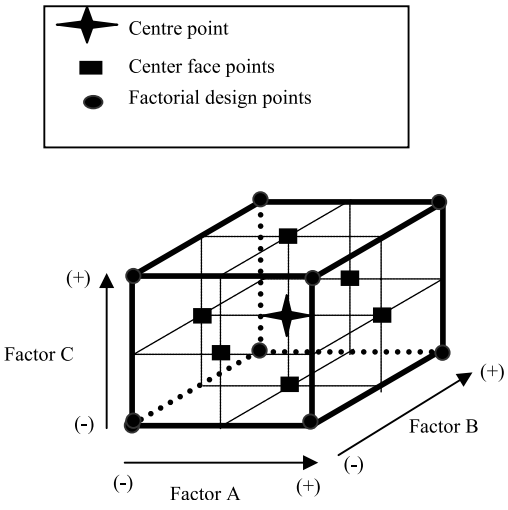

Fig. 4. The face-centered CCD with three variables.

find the optimum settings for the process variables by using the face-centered CCD approach. In the present study, however, the main objective is not to search for the optimum condition subject to various process variables, but to utilize the face-centered CCD to devise a set of experiments systematically such that the process variables can be investigated simultaneously. These experimental data samples constitute a representative data set for training the GMM. The GMM is then utilized to model the drug release process and to

Table 2
Process conditions of the test sets

Test set	Operation conditions		
	pH	Ionic strength	Current
Number 1	9	0.150	1.0
Number 2	7	0.170	0.8
Number 3	9	0.145	1.5

Table 3
The SSEs from three multiple regression models

Test set	First-order model	Second-order model	Third-order model
Number 1	1.6045	0.4234	0.2614
Number 2	2.5964	0.2983	0.6257
Number 3	5.0531	0.8305	0.4357
Mean	3.0847	0.5174	0.4409

predict the drug release profiles. In addition, outputs from the GMM can be further processed to provide useful information concerning accuracy as well as reliability of the predictions, as shown in a previous study [25]. Fig. 3 shows the flow chart of the experimental study using the face-centered CCD with the GMM as a prediction model.

With a three-variable face-centered CCD, 15 experiments (i.e. eight factorial points, one center point, and six face center points as depicted in Fig. 4) in accordance with the conditions in Table 1 were conducted. Each experiment was repeated four times. The order of the experiments were randomized to avoid influence by uncontrolled variables such as changes in environment, changes in raw materials, and changes in the conditions of device. These changes, which often are time-related, could influence the response. The samples data collected constituted the training data set for the GMM. On the other hand, three different process conditions as shown in Table 2, each with four repetitions, were used as the test set to evaluate the performance of the GMM. The sum of squared errors (SSEs) between the predicted values from the GMM and the experimental values was used as the performance indicator between the predicted and actual profiles, as follows:

$$\mathrm{SSE} = \sum_{i=1}^{n} |t_i - a_i|^2 \quad (4)$$

where t_i predicted drug release rate, a_i, drug release rate from the experiments, and n, number of time points. Note that $n = 7$ in this study with one measurement in each hour of the experiment. In addition, three multiple regression models, i.e. first-order, second-order model, and third-order models, were constructed to predict the drug release profiles. The results were used as performance comparison with the GMM.

3. Results and discussion

Table 3 shows the average SSEs (from four repetitions) for each test set using three multiple regression models, as well as the overall mean results. Based on the results in Table 3, the third-order regression model yielded the best-predicted results, i.e. with a mean SSE of 0.4409, when compared with those from the first-order and second-order models.

In ANN applications, one of the problems often encountered is the determination of suitable number of hidden nodes. Although is time-consuming and laborious, the trial-and-error method is normally used to obtain a good network structure that can produce satisfactory performance. The same approach was adopted in the present study. An initial experiment to determine the number of hidden nodes using only the first experiment of each test set was conducted. There were four inputs to the GMM, the first three inputs were the process conditions of pH, ionic strength, and current, and the fourth input was the time point. The output was the predicted permeation rate of Diclofenac Sodium.

Table 4 summarizes the SSEs from a set of experiments using different numbers of hidden neurons. Note that 32 hidden neurons yielded, on average, the best performance (SSE = 0.0562). As a result, the GMM network with 32 hidden neurons was selected for further experimentation.

Table 4
The SSEs from different number of hidden neurons

Test sets	Number of neurons						
	24	26	28	30	32	34	36
Number 1	0.4170	0.3137	0.1421	0.0321	0.0466	0.0537	0.0276
Number 2	0.1350	0.0821	0.0476	0.0323	0.0383	0.0337	0.1124
Number 3	0.6301	0.4857	0.2026	0.1287	0.0837	0.1698	0.0586
Mean	0.3940	0.2939	0.1308	0.0644	0.0562	0.0858	0.0662

Table 5
The SSEs from four tests with 32 hidden nodes

Test sets	Replications				
	1	2	3	4	Mean
Number 1	0.0466	0.0509	0.0427	0.0216	0.0405
Number 2	0.0383	0.0320	0.0325	0.1036	0.0516
Number 3	0.0837	0.1496	0.1273	0.0577	0.1045
					Overall mean 0.0655

Table 6
Bootstrap confidence interval estimates of the SSEs

Test sets	Number of resamplings	Bootstrap mean	Confidence intervals	
			Lower	Upper
Number 1	100	0.0396	0.0279	0.0489
	200	0.0393	0.0269	0.0489
	400	0.0399	0.0269	0.0489
	500	0.0397	0.0269	0.0489
	1000	0.0396	0.0269	0.0489
Number 2	100	0.0536	0.0321	0.0873
	200	0.0545	0.0321	0.0873
	400	0.0531	0.0322	0.0873
	500	0.0532	0.0322	0.0873
	1000	0.0535	0.0322	0.0873
Number 3	100	0.0985	0.0625	0.1393
	200	0.0985	0.0625	0.1393
	400	0.0988	0.0625	0.1353
	500	0.0989	0.0625	0.1353
	1000	0.0982	0.0625	0.1353

Since each test set was repeated four times, the GMM with 32 hidden nodes was then used to predict the permeation rate of Diclofenac Sodium in other replications (other than those used in Table 4). Table 5 depicts the overall SSEs and the average results. Based on the results obtained, on average, the GMM achieved almost 7-fold improvement in performance when compared with the third-order multiple regression model, with the average SSEs of 0.0655 and 0.4409, respectively.

To ascertain the reliability of the predicted results, bootstrap was then used to estimate the

95% confidence intervals of the results from the four replications. Table 6 depicts the 95% confidence intervals of the SSEs with different number of bootstrap re-samplings, i.e. 100, 200, 400, 500, and 1000 re-samplings. One can see that 400 re-samplings were enough to estimate the associated confidence intervals as there was no variation in the estimated confidence intervals after 400 re-samplings. Generally, the bootstrap means of the GMM are better than those from the third-order multiple regression model. Notice that even the upper confidence interval limits of test sets 1, 2, and 3 (0.0489, 0.0873, and 0.1353) are significantly lower than those from the third-order multiple regression model (0.2614, 0.6257, and 0.4357). Therefore, from the results and analysis the GMM appeared to be a useful network for the prediction of drug release profiles in this experimental study.

4. Conclusion

The feasibility of combining face-centered CCD and the GMM as an intelligent learning system for drug release profile prediction has been examined. An experimental study in TI was conducted to investigate the applicability of the proposed system. The face-centered CCD was first employed to investigate the relationship between various process variables simultaneously. The data samples collected from the experiments were then used as a representative training set for the GMM. The bootstrap method was applied to assess the reliability of the network predictions statistically. From the experiments, we have demonstrated that by using RSM, particularly the face-centered CCD, as a design-of-experiment tool for data collection and NNs, particularly the GMM, as a data modeling tool for profile prediction, a lengthy and time-consuming experimentation process to determine the appropriate conditions that produce a satisfactory drug release profile can be reduced.

To further validate and verify the effectiveness of the proposed system, more experiments using data samples from other benchmark problems should be conducted. Other types of NN models could also be applied and compared with the results from the GMM. In addition to NNs, other data analysis techniques, such as multiple adaptive regression splines and kriging approximation, could be combined with the RSM and used as a prediction tool. Comparative studies to ascertain the superiority of each technique in data modeling and prediction should also be performed. These are the areas for future work.

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References

[1] L.E. Frenzel, Jr, Crash Course in Artificial Intelligence and Expert Systems, Howard W. Sams, Indianapolis, 1987.
[2] D.W. Patterson, Introduction to Artificial Intelligence and Expert Systems, Prentice Hall, New Jersey, 1990.
[3] D.E. Rumelhart, J.L. McClelland, Parallel Distributed Processing I, MIT Press, Cambridge, MA, 1986.
[4] H. White, Connectionist nonpararnetric regression: multilayer feedforward networks can learn arbitrary mappings, Neural Netw. 3 (1990) 535–549.
[5] S. Haykin, Neural Network: A Comprehensive Foundation, Macmillan, New York, 1994.
[6] I. Nestorov, S.T. Hadjitodorov, I. Petrov, M. Rowland, Empirical versus mechanistic modelling: comparison of an artificial neural network to a mechanistically-based model for quantitative structure pharmacokinetic relationships of a homologous series of barbiturates, AAPS Pharm. Sci. 1 (4) (1999) 55–71.
[7] E. Brier, J.M. Zurada, G.R. Aronoff, Neural network predicted peak and trough of gentamicin concentrations, Pharm. Res. 12 (1995) 406–412.
[8] J. Takahara, K. Takayama, T. Nagai, Multi-objective simultaneous optimization based on artificial neural network in sustained released formulations, J. Control. Release 49 (1997) 11–20.
[9] J. Takahara, K. Takayama, K. Isowa, T. Nagai, Multi-objective simultaneous optimization based on artificial neural network in a ketoprofen hydrogel formula containing *O*-ethylmenthol as a percutaneous absorption enhancer, Int. J. Pharm. 158 (1997) 203–210.
[10] C.M. Bishop, Neural Networks for Pattern Recognition, Clarendon Press, Oxford, UK, 1995.

[11] D.C. Montgomery, Design and Analysis of Experiments, Wiley, Arizona State University, 1997.
[12] A.S. Hussain, X. Yu, R.D. Johnson, Application of neural computing in pharmaceutical product development, Pharmacol. Res. 8 (1991) 1248–1252.
[13] R. Linden, O.G. González, P.R. Petrovick, V.L. Bassani, Response surface analysis applied to the preparation of tablets containing a high concentration of vegetable spray-dried extract, Drug Dev. Ind. Pharm. 26 (4) (2000) 441–446.
[14] Y.Y. Huang, S.M. Wu, C.Y. Wang, Response surface method: a novel strategy to optimise Iontophoretic transdermal delivery of thyrotropin-releasing hormone, Pharm. Res. 13 (4) (1996) 547–552.
[15] D.E. Rumelhart, G.E. Hinton, R.J. Williams, Learning internal representation by error propagation, in: D.E. Rumelhart, J.L. McLelland (Eds.), Parallel Distributed Processing, vol. I, MIT Press, Cambridge, MA, 1986, pp. 318–362.
[16] S.P. Lloyd, Least squares quantization in PCM, IEEE Trans. Inf. Theory 28 (2) (1982) 129–137.
[17] A.P. Dempster, N.M. Laird, D.B. Rubin, Maximum-likelihood from incomplete data via the EM algorithm, J. R. Stat. Soc., B 39 (1) (1977) 1–38.
[18] R. Efron, Tibshirani, An Introduction to the Bootstrap, Chapman & Hall, New York, 1993.
[19] P. Panus, A.K. Banga, Iontophoresis device: clinical applications for topical develiry, Int. J. Pharm. Compounding 1 (6) (1997) 420–424.
[20] T. Masada, W.I. Higuchi, V. Srinivasan, U. Rohr, J. Fox, C. Behl, S. Pons, Examination of iontophoretic transport of ionic drugs across skin: baseline studies with the four-electrode system, Int. J. Pharm. 49 (1989) 57–62.
[21] W.H.M. Craane-van Hinsberg, L. Bax, N.H.M. Flinterman, J. Verhoef, H.E. Junginger, H.E. Boddě, Iontophoresis of a model peptide across human skin in vitro: effect of iontophoresis protocol, pH, and ionic strength on peptide flux and skin impedance, Pharm. Res. 11 (9) (1994) 1298–1300.
[22] W.L. Chou, C.H. Cheng, S.C. Yen, T.S. Jiang, The enhanced iontophoretic transport of TRH and its impedance study, Drug Dev. Ind. Pharm. 22 (9 and 10) (1996) 943–950.
[23] L. Liauw, J.D. Moscaritola, J. Burcher, Nonsteroidal Anti Inflammatory Drugs: Mechanisms and Clinical Use (Chapter 19), Marcel Dekker, New York, 1987, pp. 329–347.
[24] S.S. Quek, C.P. Lim, K.K. Peh, Reliability of artificial neural network predictions—a case study in drug release profile predictions, in: P. Sincak, J. Vascak, V. Kvasnicka, R. Mesiar (Eds.), The State of the Art in Computational Intelligence, Physica-Verlag, 2000, pp. 337–343.