Research Article

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Energy-saving analysis of desalination equipment based on a machine-learning sequence modeling

https://doi.org/10.1515/EHS-2024-0002
received January 19, 2024; accepted May 04, 2024

Abstract: To control water quality and seawater desalination dosage, modeling the coagulation process of saltwater is crucial. With a focus on the features of seawater coagulation with a long lag, a machine-learning sequence-based modeling approach is suggested. The link between influent and effluent turbidities, flow rates, flocculant and coagulant dosages, and other parameters is modeled using structured units such as a gate recurrent unit encoder and a linear network decoder. The model's validity is confirmed by numerical experiments based on real operating data, which also offer a solid foundation for managing flocculant and coagulant assistance reduction.

Keywords: desalination, pretreatment, coagulation and sedimentation, sequential modeling, machine learning

1 Introduction

A viable technological solution widely acknowledged as one of the most effective approaches to address severe freshwater scarcity caused by the unequal distribution of water resources worldwide is desalination (Soleimanzade et al. 2022). Desalination technology reduces severe freshwater scarcity by increasing water availability, diversifying sources, and improving water quality. It benefits ecosystems by lowering the demand for freshwater sources and enhancing climate resilience. Desalination fosters social fairness by ensuring that vulnerable populations can access clean water. Sustainable techniques are vital for reducing environmental damage and ensuring long-term water security. When confronted with the issue of elevated energy usage, conventional desalination methods must look for novel approaches. This study aims to improve the economics and energy efficiency of the desalination process by optimizing the water quality control of coagulation and sedimentation processes by introducing a machine-learning sequence model. Turbidity, pH, alkalinity, temperature, coagulant dosage, and settling time are standard parameters used in water quality management during coagulation and sedimentation operations. This is done through the use of a thermal/membrane-coupled technology.

The high-energy consumption of conventional technologies limits the advancement of seawater desalination technology (Abba et al. 2023a). Conventional desalination processes use a lot of energy, which is expensive and causes environmental problems. Thermal distillation and reverse osmosis (RO) demand a lot of energy, which could burden electricity infrastructure and increase greenhouse gas emissions. This increases operating costs, making desalinated water more expensive than typical freshwater sources. Balancing water security with energy-intensive desalination's environmental and economic costs is a critical challenge for sustainable water management. While RO for membrane methods and multi-stage flash (MSF) and low-temperature multi-effect distillation (MED) for thermal methods are mature, they are nevertheless constrained by increased energy consumption (Abba et al. 2023b). Thermal energy is used in the MED system to heat seawater and produce vapor, which condenses into freshwater. This thermal energy, generally derived from waste heat or solar sources, is the primary source of freshwater generation in MED systems.

In contrast, electric energy is primarily employed for auxiliary functions in MED systems, mainly for power transfer pumps. These pumps move seawater through several stages of the distillation process without immediately contributing to freshwater production. In a thermal/membrane-coupled desalination system, the cooling water of the MED system contributes to the feed seawater for RO. This uses MED waste heat to preheat seawater for RO, which

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increases energy efficiency. With heat and membrane techniques coupling, the thermal/membrane-coupled desalination technology has emerged in recent years, providing new opportunities to increase energy consumption efficiency (Salem et al. 2022). Using a machine-learning sequence model in the desalination process improves energy efficiency and economics by optimizing operational parameters, estimating energy consumption, and discovering process optimization opportunities.

Several researchers have achieved some progress in seawater desalination, but there are still obstacles to overcome (Bonny et al. 2022). While the system's economy has been somewhat enhanced by hot-film coupling technology, more innovation and optimization are still required to help it better fit the intricate and ever-changing desalination environment (Hai et al. 2023). Hot-film coupling technology improves the economic sustainability of seawater desalination by increasing the energy efficiency and lowering operating expenses. It excels because of its excellent heat transmission, minor construction, and compatibility with various desalination processes, including RO. Overall, it increases cost-effectiveness by optimizing the energy consumption and operating efficiency. The direction of increasing system efficiency and cutting costs is the primary emphasis of current academic research in the field of seawater desalination (Mahdavi-Meymand and Sulisz 2023). Integrating thermal and membrane techniques, particularly in thermal/membrane-linked desalination technology, offers a fresh avenue for technological advancement (Zouli 2023). The difficulties are still present, though. Current research is still hot and challenging regarding energy consumption concerns, system stability, and application under various climatic situations (Habieeb et al. 2023).

With a focus on controlling water quality during coagulation and sedimentation, this study aims to investigate the possible applications of machine learning in the desalination process. One innovative approach to modeling the seawater coagulation process is machine-learning sequence models, particularly the gated recurrent unit (GRU) structure (He et al. 2022). We anticipate that by carefully examining the crucial elements of coagulation and precipitation processes, we will be able to optimize the control approach, cut down on resource waste, and accomplish more sustainable development – all of which will contribute to an even better desalination system (Rashidi et al. 2022). GRU structural modeling is an excellent way to manage water quality during desalination procedures. GRU models use a neural network architecture to anticipate water quality changes, optimize process parameters, detect anomalies, assist adaptive management strategies, and provide data-driven decision support. This allows operators to proactively maintain desired water quality levels, reduce energy usage, and assure regulatory compliance, eventually increasing the operating efficiency and producing high-quality desalinated water.

In this work, we incorporate machine-learning sequence modeling and thermal/membrane-linked desalination technology to address several issues in the desalination process. Incorporating energy-using aspects from thermal and membrane systems into energy-saving assessments improves the energy efficiency and responsiveness to changing climates. Hybrid solutions can be constructed by combining each system's capabilities, such as thermal systems' high freshwater production and membrane systems' energy efficiency. This approach optimizes desalination plants to run efficiently independent of external circumstances, delivering consistent freshwater production. Combining thermal and membrane technologies in seawater desalination improves the energy efficiency and climatic adaption. In particular, we manage the water quality using GRU structural modeling to optimize the coagulation and sedimentation processes (Ray et al. 2022). The benefit of this approach is that it enables us to increase the stability and efficiency of the system by using deep learning to more precisely understand the intricate relationships involved in the coagulation and sedimentation processes. To more effectively utilize energy year-round and better adapt to varying climatic conditions, we integrate the energy-using features of both the thermal and membrane systems in the energy-saving analysis and suggest two coupling strategies that match the temperature of the feed seawater (Shim et al. 2023, Hai et al. 2023). Each coupling approach has distinct advantages and disadvantages, such as improved energy usage, but with possible issues like membrane fouling or high-pressure pumping requirements. Evaluating trade-offs is critical for implementing sustainable desalination procedures.

This thesis will begin with an introduction, then go into the history of development and background of desalination technology, and then provide a thorough explanation of the potential applications of machine learning in desalination. The research technique, which includes data pretreatment, the use of the Seq2Seq model, and the GRU structure, will next be covered. Then, the outcomes of the experiments confirm the model's validity. Subsequently, we will examine the energy-saving analysis and the optimization approach of dosage control in the seawater coagulation process. We will summarize the findings and offer a prediction for future lines of inquiry. We hope the study presented in this thesis will provide fresh perspectives and methodologies for advancing seawater desalination technology.

2 Methodology of this paper

2.1 Data preprocessing

Data pretreatment is a crucial phase in the machine-learning-based modeling process, which can enhance data quality and facilitate modeling (Yin and Lei 2022, Liang et al. 2023). Preprocessing the data can increase model accuracy and decrease training difficulties by considering the errors of different kinds of equipment and the effect of random noise in the data sampling process. Preprocessing data to account for random noise and equipment flaws improves machine-learning models' overall accuracy and reliability. The models may focus on essential patterns by cleaning and normalizing the data before training, resulting in more trustworthy predictions and less sensitivity to noise or erroneous inputs. Feature engineering preprocesses data by converting raw input variables into relevant features better suited for machine-learning algorithms. It helps to find and choose important features by extracting meaningful information, producing new features, and selecting the most relevant ones to improve the model performance. This technique enhances the model accuracy, reduces the overfitting, and increases the interpretability by concentrating on the most significant features of the data. Data preparation can be implemented through the use of the following methods:

(1) *Processing of outliers: Outliers will appear in the data because of the instrument's measurement mistake. To select the outliers based on real circumstances, a threshold may be set, and the outliers that fall below the threshold can be eliminated.*

(2) *Average processing of slides: Sliding average processing is applied to the original data to minimize the random noise overlay on the initial data.* Equation (1) *displays the sliding average's mathematical expression* (Ali et al. 2023*). A sliding average is used in data processing to smooth out oscillations and variability, minimizing noise. By taking an average value over a sliding window of consecutive data points, this method provides a more consistent representation of the underlying signal, making it easier to discern essential trends or patterns. Sliding average processing for noise reduction offers simple and effective smoothing of data fluctuations, revealing trends while retaining the data structure. However, it may result in a loss of detail, latency in response to changes, and a reduction in the influence of outliers, lowering analysis precision.*

$$y(t) = \frac{1}{w} \sum_{k=t-\lfloor w/2 \rfloor}^{k=t+\lfloor w/2 \rfloor} x(k). \tag{1}$$

When the sliding window size is denoted by w, the instant is indicated by t, the original data is represented by $x(k)$, and the smoothed processed data is denoted by $y(t)$.

(3) *Processing for normalization: The convergence speed of the model can be improved by data normalization, which ensures that all variables are calculated on the same scale* (Ali et al. 2023, Ren et al. 2023). *Min-max normalization is applied to normalize the original data*; equation (2) *provides the mathematical expression for this technique* (Nazeer et al. 2023, Zeng and Chu 2024). *Min-max normalization reduces numerical data to a specific range, usually 0 to 1, for simplicity and interpretability. However, it may not handle outliers well and is computationally expensive for large datasets compared to alternative normalization techniques such as Z-score normalization or decimal scaling.*

$$y = \frac{(x - x_{\min})}{(x_{\max} - x_{\min})}, \tag{2}$$

where y represents the normalized data, $x_{\max}$, $x_{\min}$ represents the minimum and maximum values of the original data, respectively, and x represents the original data.

2.2 Sequence-to-sequence model

The sequence-to-sequence model (Seq2Seq model) can extract and parse complicated features from sequences and is mainly used to describe sequence-to-sequence form challenges. Tasks involving natural language processing frequently use this model (Jiao et al. 2024). In a Seq2Seq model, the encoder gathers input sequence information and encodes it into a fixed-length vector, while the decoder constructs an output sequence using this context vector. The encoder summarizes the input's content and context, which the decoder then uses to construct output tokens step-by-step. Together, they allow Seq2Seq models to process and create sequences for tasks such as translation and prediction. Seq2Seq models excel at handling issues in NLP tasks such as machine translation and text summarization because they capture complicated links between input and output sequences, can handle variable-length inputs, and produce coherent outputs. Seq2Seq models may not be suitable for sequence-to-sequence tasks involving radically varied sequence lengths, large sequences, complicated linguistic patterns, or long-range dependencies in the data.

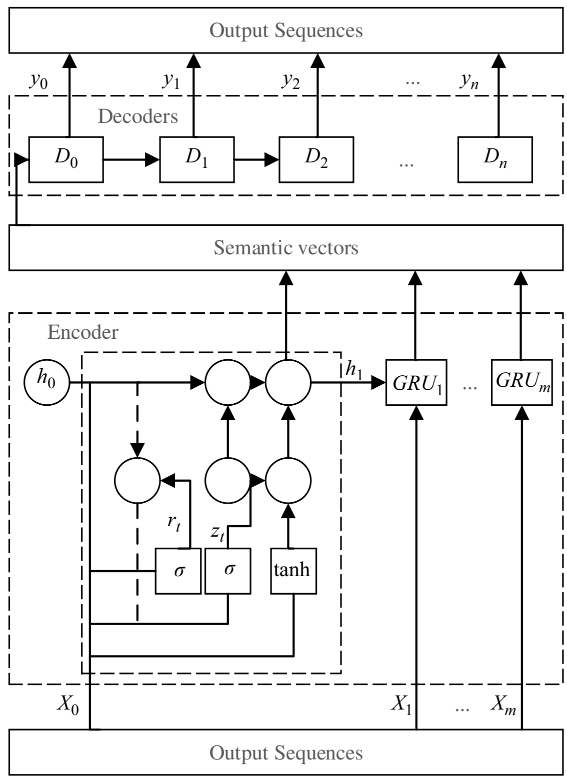

Figure 1: Structure of the widely used GRU encoder Seq2Seq model.

Figure 1 depicts a typical model of this kind, which primarily uses two separate networks, an encoder and a decoder. The encoder compresses the input sequences and converts them into feature vectors, which are then semanticized to produce semantic vectors. After parsing the semantic vector, the decoder produces a sequence with a given length.

Because the seawater coagulation process takes a long time, the past inputs impact the effluent's turbidity. The mathematical description of the coagulation process gives the effluent's turbidity an essential parameter for process optimization and control in seawater desalination plants. Using mathematical models to forecast turbidity levels correctly, operators may modify coagulant dosages and treatment operations in real time, ensuring ideal water quality while minimizing energy use. This procedure can be represented mathematically, with the turbidity of the effluent serving as the output and a series of past inputs as the input.

2.3 GRU

A gated recurrent unit (GRU) is a form of long short-term memory (LSTM) network, which is primarily utilized in the challenge of modeling sequence models (Yoon et al. 2022). GRU is roughly equal to LSTM in terms of model-fitting capabilities but has a more straightforward structure. In recurrent neural networks (RNNs), GRU can help with lengthy dependence issues and prevent computation-related problems like gradient vanishing. GRUs employ gated mechanisms and skip connections to mitigate gradient vanishing during training for sequential data processing. The model's training efficiency can be increased, and the LSTM's sluggish training speed issue can be resolved using GRU.

The internal structure of the GRU is shown in the encoder in Figure 1. *rt* is the update gate and *zt* is the reset gate. The internal architecture of a gated recurrent unit (GRU) allows the model to capture long-range dependencies more efficiently than typical RNNs. GRUs accomplish this by implementing gated mechanisms that control the flow of information within the network, allowing them to retain important details over longer sequences without experiencing vanishing gradient difficulties. The update and reset gates accept the current sequence input *xt* and the previous temporal hidden state input h_{t-1}. The reset gate is used to delete memories and control short-term memories. The update gate is used to prevent long-term memories and finally outputs a semantic vector containing sequence features.

3 Testing of models

The model is trained with the Adam (Drogkoula et al. 2023) optimizer with the loss function L_1, and the training and test sets are split using the random sampling technique. Figures 2 and 3 display the model's outcomes for the training and test set data, respectively. With a coefficient of determination (R_2) of 0.98 on the test set, the model demonstrates its ability to suit the coagulation and sedimentation processes.

4 Enhancement of dosage regulation for seawater coagulation

Upon examining the statistical data from the original data, it is evident that during the real manufacturing process, the flocculant dosing frequency change trend and the flow rate of the uncontrollable variable change trend are relatively close (Gollangi and Nagamalleswara Rao 2023). Understanding the relationship between the flocculant dosage frequency and the flow rate of uncontrollable factors in the manufacturing process is critical. It aids in the optimization of the dosing

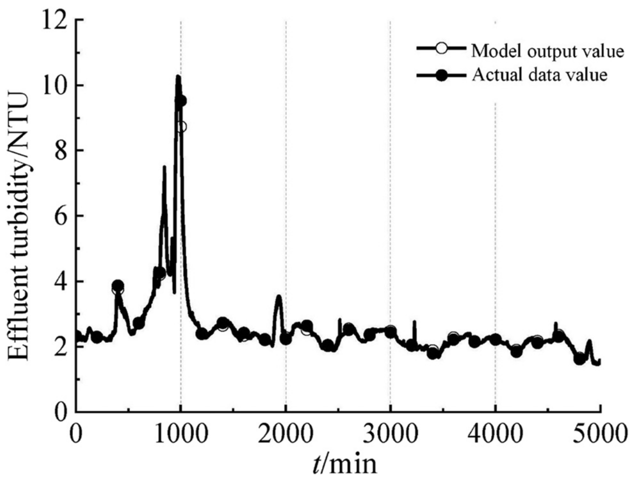

Figure 2: Model performance using training set data.

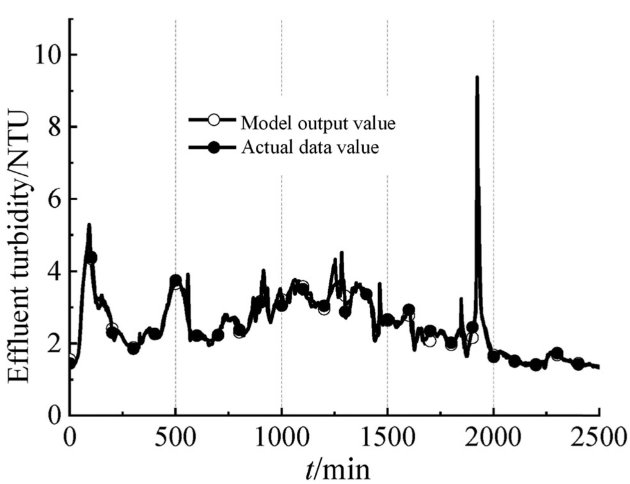

Figure 3: Model outcomes utilizing test set data.

strategy for effective water treatment, ensuring that flocculants are given at a proper frequency to compensate for flow rate variances and maintain constant water quality standards. Strategies for mitigating the impacts of flow rate on the flocculant dosing frequency include continuous monitoring, automated dosing systems, flow rate thresholds, adaptive algorithms, and dynamic dosage control. These provide appropriate dosage despite fluctuations, which improves the water treatment efficiency. A positive correlation has been seen between the flocculant dosage and the flow rate; as the flow rate increases, so does the flocculant dosing frequency. Maintaining the flocculant dosage frequency when the flow rate surpasses the threshold guarantees that the water treatment efficiency remains stable. Reducing the dosing frequency at lower flow rates conserves the flocculant while maintaining quality standards. The dosage can be adjusted based on the flow size; when the flow is large, the initial dosing frequency is maintained; when the flow is small, the dosage frequency is decreased. Reduced dosage based on proposed criteria may result in ineffective treatment, insufficient pathogen elimination, and impaired water quality standards. Insufficient dosage can result in microbial regrowth, increased turbidity, and significant health risks, particularly in essential applications such as water treatment. This way, you can guarantee that water's turbidity meets production standards while lowering the dosage to save production costs (Chen et al. 2023, Ba-Alawi et al. 2023). Variations in flow size can impact the effectiveness of flocculant dosage adjustment in maintaining appropriate water turbidity levels. Smaller flow sizes may result in flocculant overdose, causing excessive treatment and potential water quality issues. In contrast, larger flow sizes may result in underdosing, failing to treat the water effectively, and potentially enabling turbidity to exceed permitted limits. To achieve the best results in water treatment, flocculant dosage must be balanced with flow size fluctuations (Tables 1 and 2).

The analysis presented above was used to establish the following dose control guidelines. The flow rate was measured using the 75% quantile (6196.40) as the threshold value. If the flow rate was higher than this value, the flocculant dosing frequency was maintained; if not, it was appropriately reduced, and the coagulant aid dosing frequency was maintained. Managing the reduction of flocculants and coagulants is critical for managing effluent turbidity since these chemicals play an important role in aggregating suspended particles in water and assisting in their removal via sedimentation or filtration processes. Failure to maintain proper amounts of flocculant and coagulant can result in poor particle removal, increasing the effluent turbidity and potentially causing environmental and regulatory compliance difficulties. An interval of 5,000 points from the original data set was chosen for testing, and

Table 1: Model performance using training set data

Time (*t*/min)	Effluent turbidity/NTU	
	Model output value	**Actual data value**
0	2.40	2.40
500	3.83	3.74
1,000	9.40	8.59
1,500	2.43	2.31
2,000	2.31	2.31
2,500	2.10	2.10
3,000	2.49	2.49
3,500	1.89	2.08
4,000	2.31	2.31
4,500	2.22	2.22
5,000	1.60	1.60

Table 2: Model outcomes utilizing test set data

Time (t/min)	Effluent turbidity/NTU	
	Model output value	Actual data value
0	1.53	1.84
250	2.40	2.40
500	4.10	3.89
750	2.91	2.91
1,000	3.41	3.62
1,250	3.93	3.91
1,500	2.91	2.84
1,750	2.46	2.31
2,000	10.20	10.20
2,250	1.81	1.72
2,500	1.48	1.48

the reduction multiplier for the test was taken as 0.75, 0.85, and 0.95, respectively. Figure 4 displays the predicted effluent turbidity time series curve. This approach can reduce the dosage by around 20% overall if a reduction multiplier of 0.75 is applied.

It should be noted that the range of variation of the obtained operation data is minimal and impacted by the actual production seasonal factors. This limits the model's validity, and additional temporal and seasonal operation data are required to improve it. Seasonal variations can impact the performance of dosage reduction control models by changing the water quality and demand. To mitigate this, seasonal trends are incorporated into the model, data are updated regularly, and adaptive management mechanisms are used. External influences such as weather and agricultural cycles should be integrated to improve model robustness. However, the dose reduction control approach presented above is merely an initial attempt, and more research is necessary to determine the complete optimum control technique (Ullah et al. 2023, Yoon et al. 2023, Xie et al. 2024).

5 Energy-saving evaluations

Two coupling methods that match the temperature of the feed seawater are developed to reduce consumption and save energy. Reducing energy consumption in saltwater desalination is critical due to its sizeable environmental impact and high operating expenses. Energy-efficient operations help to combat climate change, promote sustainable water management, and make desalinated water more accessible and inexpensive to populations experiencing freshwater scarcity. These methods combine the energy-using characteristics of both systems, namely thermal and membrane approaches.

Mode 1: Cooling water with the residual temperature of MED is combined with feed seawater during the summer months when seawater temperatures are high (Tcw > 15°C). This keeps the temperature of RO feed seawater consistently at 30°C.

Mode 2: Using a hot film-linked heat exchanger, feed seawater and MED-concentrated brine are heated to approximately 10°C using heat exchange during winter when seawater temperatures are low (Tcw ≤ 15°C).

5.1 Comparison between thermal/membrane-coupled desalination and thermal-/membrane-independent operation modes for water withdrawal

The total water intake of the thermal/membrane-coupled desalination system will be lower than that of the independently operated RO/MED system during summer because part of the feed seawater of the RO system comes from the cooling water of the MED system; however, during winter, the water intake of the two modes of operation will be

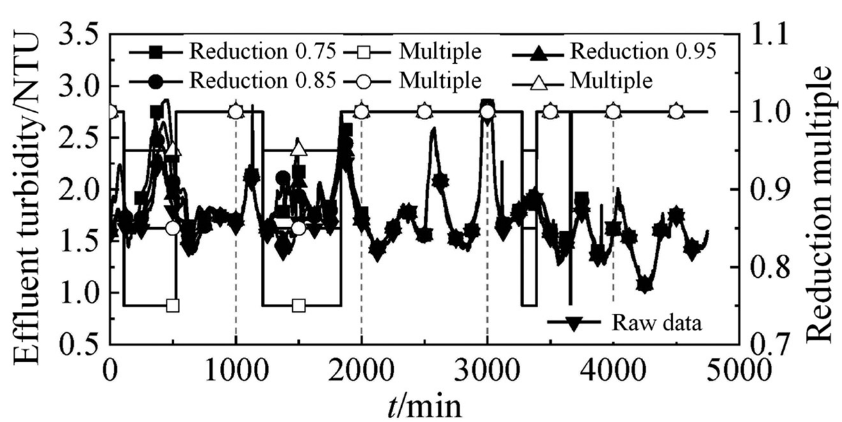

Figure 4: Dose rate optimization test curve.

equal. Desalination technology and system design advancements can improve water intake management in coupled desalination systems by incorporating innovative intake methods, such as subsurface intakes or seawater wells, that reduce the environmental impact and energy consumption during water intake. Taking the demonstration project as an example, the independently operated 521 t/h RO system + 520 t/h MED system is compared with the 1,041 t/h thermal/film-linked system (Priya et al. 2022, Shu et al. 2022, Jiang et al. 2021), and the yearly water intake of the independently operated RO/MED system is determined to be 33,303,500 t. The thermal/film-coupled seawater desalination plant takes in 31,268,800 t of water annually. A 203.3 million t/year reduction in the annual water intake is possible with the thermal/film-coupled desalination system. Thermal processes such as MSF distillation, energy-intensive high-pressure pumping in RO systems, the need for contaminant pretreatment, and energy-intensive brine disposal and post-treatment processes all contribute to conventional seawater desalination technologies' high-energy consumption. RO membrane design innovations aim to lower the energy consumption. These include high-performance thin-film composite membranes, enhanced surface modifications, innovative materials such as graphene oxide and carbon nanotubes, and optimal membrane architectures and configurations. Using a thermal/film-coupled desalination system can result in a 203.3 million t reduction in yearly water consumption. It is possible to cut the annual water input by 2,034,700 t. Figure 5 compares the water intake in the thermal/membrane-coupled desalination mode and the thermal/membrane-independent operation mode.

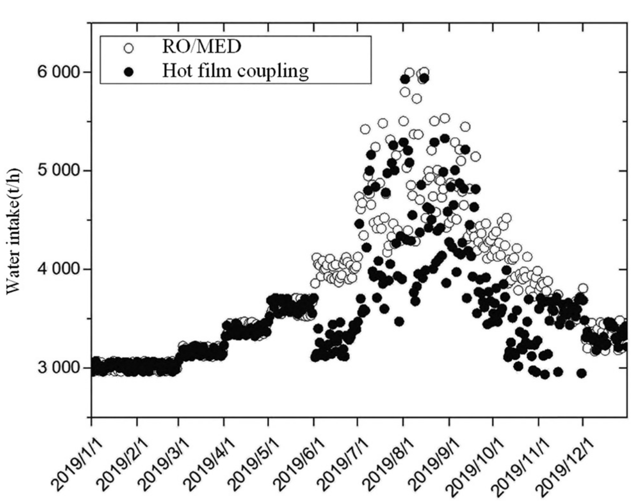

Figure 5: Comparison between the thermal/membrane-connected desalination mode and the independent operating mode of the thermal membrane method for water extraction.

5.2 Comparison of thermal/membrane-linked desalination mode and thermal-/membrane-independent operation mode electricity consumption

The primary forms of energy used in MED are thermal energy and electric energy, which are used as auxiliary energy to provide energy for the operation of the transfer pump. The power consumption of the independently operated MED/RO system is equal to that of the MED part of the thermal/membrane coupling system. Factors influencing power consumption in independently operated MED/RO systems include the energy necessary to heat saltwater in the MED stage and high-pressure pumping in the RO stage. Independently operated systems may use more energy than thermal/membrane coupling systems due to the separate energy-intensive distillation and membrane filtration operations. Feed water salinity and temperature variations can also affect power usage in both systems. The primary energy source for RO is electric energy from the high-pressure pump. The primary distinction in energy sources and consumption patterns between RO and MED systems is based on their operational mechanisms. RO systems use electric energy for high-pressure pumping to force seawater through membranes.

In contrast, MED systems use thermal energy for distillation operations, frequently sourced from waste heat or solar sources. RO systems use less energy per cubic meter of water generated than MED systems, which use more energy due to thermal distillation. As the membrane flux rises with influent temperature, so does the high-pressure pump's power consumption, which falls as the flux increases. The power consumption of high-pressure pumps in RO systems varies with membrane flow and influent temperature variations. Increased membrane flux increases the pushing force for water transport, requiring more pump energy, whereas higher influent temperatures lower water viscosity, potentially reducing pump energy consumption. However, the precise effects depend on the system. Energy recovery devices, variable frequency motors, enhanced membrane materials, and pretreatment processes are among the strategies used to optimize power usage in RO systems, mainly when accounting for fluctuations in influent water temperature. These strategies improve the energy efficiency by recapturing energy, changing pump speeds, increasing membrane efficiency, and decreasing fouling effects. The rise in influent water temperature causes an increase in the membrane flow. Figure 6 illustrates how thermal/membrane coupling technology significantly reduces the amount of electrical energy consumed in winter since seawater is colder. Thermal/

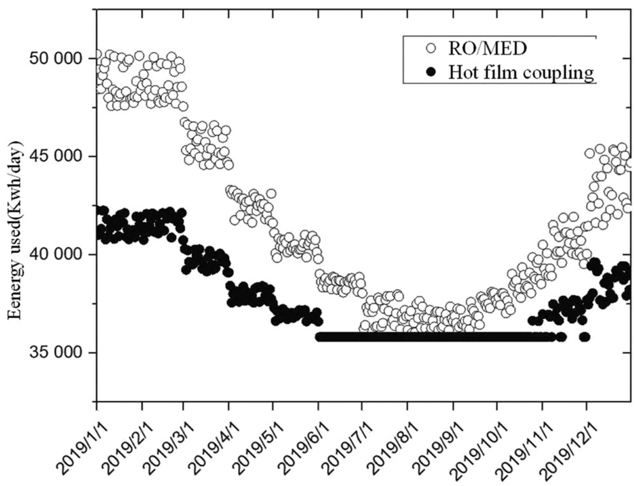

Figure 6: Power consumption comparison of thermal-/membrane-independent and thermal/membrane-coupled desalination modes.

membrane coupling technology improves energy efficiency by using waste heat to preheat seawater, reducing energy requirements during more frigid conditions.

After calculation, the annual power consumption of the high-pressure pump of the independently operated RO/membrane desalination is 15.141 million kWh. In comparison, the annual power consumption of the high-pressure pump of the thermal/membrane-coupled seawater desalination system is 13.747 million kWh. Adopting the thermal/membrane-coupled seawater desalination technology can reduce the annual power consumption of the high-pressure pump by 1.39 million kWh.

The yearly water extraction volume of the thermal/membrane-coupled desalination process can be decreased by 2.03 million tonnes annually based on the extraction pump's energy consumption ratio, computed using 0.067 2 kWh/m^3. The extraction pump's energy consumption ratio (0.0672 kWh/m^3) indicates the energy needed to extract one cubic meter of water in desalination procedures. This ratio represents the pump's efficiency in energy consumption per unit of water output, providing information about the operational expenses and sustainability of desalination operations. Lower ratios indicate more energy efficiency, reducing the overall environmental effect and operational costs of desalination. The computation above shows that it is possible to save 0.13 million kWh in yearly power consumption for the water intake pump and to lower the annual water intake of the thermal/film-coupled desalination by 2.03 million t. In conclusion, the water production of the thermal/film-coupled desalination technology in Bohai Bay can save 1.53 million kWh annually on the electricity consumption of the high-pressure pump and power intake pump. The non-hourly electricity price of 0.785 3 yuan/kWh is used to calculate the electricity price, which results in an annual savings of 1,650,116.5 million yuan on electricity costs.

6 Conclusions

This work used a desalination plant as the research object to present an efficient machine-learning sequence model-based modeling approach for controlling coagulation and sedimentation processes in water quality. Using a gated recurrent unit (GRU) encoder and a linear network decoder, a relationship model was built about the relationship between effluent turbidity and other parameters. The model's validity was confirmed through numerical experiments utilizing real-world operation data, and it served as a foundation for managing the decrease of flocculant and coagulant. Through energy-saving analysis, two coupling approaches matching the temperature of feed seawater are proposed by combining the energy consumption characteristics of each system of thermal and membrane methods. The notable annual reductions in power consumption and water input achieved with the thermal/membrane coupling desalination method provide strong support for practical applications.

Funding information: No funds or grants were received by any of the authors.

Author contributions: Xiaodong Zhang, Yuepeng Jiang and Ke Li conceptualized the study. Xiaodong Zhang, Yu Sun and Ke Li participated in the methodology procedures. Xiaodong Zhang, Yu Sun, Yuepeng Jiang and Ke Li participated in the formal analysis. Xiaodong Zhang and Ke Li wrote the original draft. All authors have read and agreed to the published version of the manuscript.

Conflict of interest: There is no conflict of interest among the authors.

Code availability: Not applicable.

Data availability statement: All data generated or analyzed during this study are included in the manuscript.

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