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# Reverting Gene Expression Pattern of Cancer into Normal-Like Using Cycle-Consistent Adversarial Network

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***Abstract***

*Cancer show distinct pattern of gene expression when it is compared to normal. This difference results malignant characteristic of cancer. Many cancer drugs are targeting this difference so that it can selectively kill cancer cells. One of the recent demand for personalized treating cancer is retrieving normal tissue from a patient so that the gene expression difference between cancer and normal be assessed. However, in most clinical situation it is hard to retrieve normal tissue from a patient. This is because biopsy of normal tissues may cause damage to the organ function or a risk of infection or side effect what a patient to take. Thus, there is a challenge to estimate normal cell's gene expression where cancers are originated from without taking additional biopsy.*

*In this paper, we propose in-silico based prediction of normal cell's gene expression from gene expression data of a tumor sample. We call this challenge as reverting the cancer into normal. We divided this challenge into two parts. The first part is making a generator that is able to fool a pretrained discriminator. Pretrained discriminator is from the training of public data (9,601 cancers, 7,240 normals) which shows 0.997 of accuracy to discriminate if a given gene expression pattern is cancer or normal. Deceiving this pretrained discriminator means our method is capable of generating very normal-like gene expression data. The second part of the challenge is to address whether generated normal is similar to true reverse form of the input cancer data.*

*We used, cycle-consistent adversarial networks to approach our challenges, since this network is capable of translating one domain to the other while maintaining original domain's feature and at the same time adding the new domain's feature. We evaluated that, if we put cancer data into a cycle-consistent adversarial network, it could retain most of the information from the input (cancer) and at the same time change the data into normal. We also evaluated if this generated gene expression of normal tissue would be the biological reverse form of the gene expression of cancer used as an input,*

***Keywords:*** *cancer gene expression, deep learning, cycle-consistent adversarial network*

## 1. Introduction

Deep neural network is one of the top performing models currently in the field of machine learning.[1] This technology is being widely used in computer vision and natural language processing. In the field of life sciences, its usability is attracting the attention in the field of diagnosis and prediction of diseases.[2]

In 2014, Ian Goodfellow proposed a Generative Adversarial Network by changing the structure of the neural network and broadened the field of its application.[3] Ian Goodfellow's Generative Adversarial Network was a model that could produce fake data similar to the real ones. If the existing neural networks model were mainly

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used to solve regression and classification problems, the new model has reached the level where fake photographs and images created were hard for naked eyes to distinguish between real data.

Since then, attempts have been made to make various changes in the structure of neural network to obtain the desired outputs. Neural style transfer, which generates domain Y data while preserving the characteristics of domain X, has been developed. One of the noteworthy researches is Jun-Yan Zhu's *Unpaired Image-to-Image Translation using Cycle-Consistent Adversarial Networks*.[4] This Cycle-Consistent Adversarial Network, which is often called Cycle GAN, keeps the characteristics of domain X as much as possible while changing it to Y. In this way we can transform our own pictures into the style of Van Gogh's, while maintaining the overall characteristics of the original pictures. In particular, the advantage of this model is that, while previous models were able to be trained only when the data in the two domains were paired, this model is able to be trained even when the data are not necessarily paired. This enabled the use of unpaired data in solving the problem of domain transfer.

Thanks to these studies, a lot of research is now being carried out based on this idea. However, most studies are confined to image processing, which is the application field of the original paper, and it is hard to find applications in the field beyond this. In particular, to the best of our knowledge there are no examples of using the transcriptomic data, which is a high-order data of more than 20,000 features. If we can change the high-order gene expression data through Cycle GAN, we expect that the application range will be unlimited. Let X be the group of cells with disease and Y be the group of normal cells. If the domain transfer from X to Y occurs successfully, as in the case of images, we expect to transform the gene expression data of certain diseased cells to normal and preserve the characteristics of that particular cell. In other words, we hypothesized that the use of Cycle GAN will change the gene expression of a specific cell in a disease state to the state of gene expression before the disease takes place in the cell. If this hypothesis is proved to be correct, it is expected that the original expression of the specific cell with the disease will be identified and be analyzed to provide a customized analysis of the cause and treatment of the disease. Although we can deal with many diseases, we focused on cancer since the availability of large size of public data.

As stated in the introduction, our primary goal is to give Cycle GAN the gene expression data of cancer cells and transform it into the expressions of normal cells. Secondary goal is to verify whether this normal state simulation data has been well preserving the characteristics of that specific cancer cells.

The first goal is to give the Cycle GAN model both cancer and normal cells into the input, which in turn makes it possible to make the cancer data into generated normal data and normal data into generated cancer data. Computer science used Cycle GAN mainly based on images, so it was easy to confirm visually that fake images were well produced. However, it is not easy to examine multiple gene expressions data and find out how well it has been converted from cancer to normal or vice versa. Therefore, it is also necessary to devise a proprietary measurement method that confirms that training has been successful. To do this, we decided to use pretrained discriminator model. We used the neural network model which has been pretrained with normal and cancer data. We used cycle GAN to generate fake normal data and fake cancer data, and put it into the pretrained model, which aims to confirm that this model cannot distinguish fake data from real. The goal was to make the accuracy more than 98 percent when generated data were given, when the pretrained model showed 99 percent accurate when real data were given.

The second goal is to verify that the generated fake data is preserving the characteristics of the original data well. This is also easy to identify in the image, but it is difficult to identify with gene expression data. Two methods are suggested for this purpose. The first method is to verify by using paired data. Paired sample means cancer data acquired from cancer tissues and adjacent normal data acquired by cancer tissues from the same individual. Since it is a pair, we can say that we have the correct answer data of the hypothesis we want to verify. This is because the adjacent normal tissues in which cancer has not metastasized can be considered to be similar to those in normal cancer cells around. Another method is to verify using unpaired data. Unpaired data does not have the correct label, but it has data of cancer tissues. In other words, if the generated fake normal data is closer to the original cancer tissue than the other cancer tissue data, we can verify our hypothesis.

## 2. EXPERIMENT

### Data Description

The data used to create the model was obtained from a public database. The Cancer Gene Atlas (TCGA), TARGET, and Genotype-Tissue Expression (GTEx) platforms were integrated to obtain 9,601 cancer samples and 7,240 normal sample data from a total of 25 tissues.[5-7]

**Table 1. Data Description**

| Tissue | Cancer | Normal |
|---|---|---|
| BRCA | 1410 | 2 |
| LUAD | 813 | 3 |
| OV | 723 | |
| KIRC | 612 | |
| PRAD | 582 | |
| THCA | 566 | |
| LUSC | 549 | 5 |
| HNSC | 543 | 1 |
| SKCM | 469 | |
| BLCA | 446 | |
| LIHC | 422 | |
| STAD | 399 | 5 |
| COAD | 379 | 15 |
| CESC | 309 | |
| SARC | 264 | |
| UCEC | 190 | 16 |
| PAAD | 182 | |
| GBM | 167 | 5 |
| ESCA | 167 | 3 |
| Testicular Germ Cell Tumor | 154 | |
| MESO | 86 | |
| ACC | 77 | |
| DLBC | 47 | |
| Cholangiocarcinoma | 45 | |
| Non-cancer | | 7185 |

We conducted experiments with 308 genes included in the commercial cancer mutation panel (Foundation One ™). We also included the process of normalizing the data before learning the model. Sample within standardization was performed and standard normalization was performed using the mean and standard deviation of each sample.

### Pretrained Discriminator

In order to calculate the performance of the Cycle GAN model, we constructed a pre-trained discriminator using a deep neural network. The model is a typical fully connected multi-layer perceptron. One input layer and five hidden layers, and finally an output layer. The input layer consists of 308 nodes and the hidden layer consists of 1500, 200, 200, 200 and 50 nodes in order. The final output layer consists of a single node that indicates the probability that the data will be cancer. The most widely used ReLu function is used as the activation function of the neural network and drop-out is applied at a ratio of 0.5.

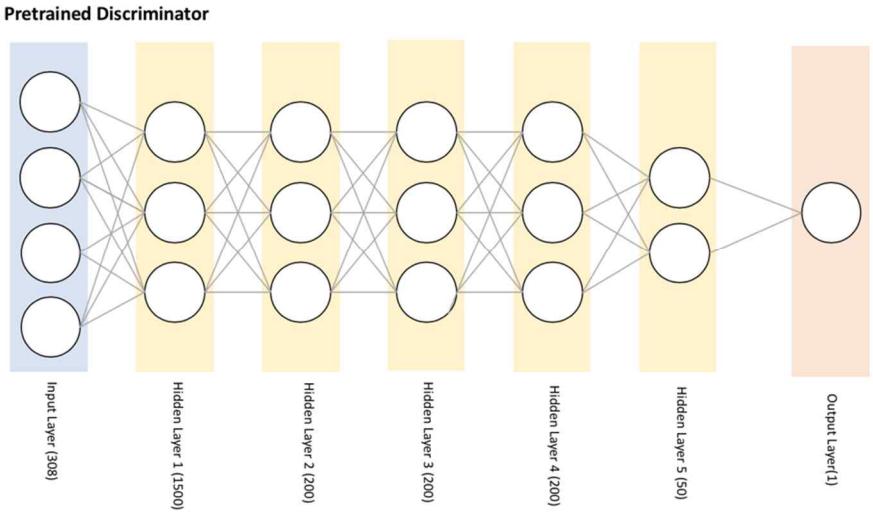


**Figure 1. Structure of Pretrained Discriminator**

The loss function used to study the model was a binary cross-entropy, which is mainly used for the binary classification problem, and the gradient was updated after backpropagation using the Adam optimizer. At this time, in order to prevent over-fitting, 1/9 of the training data was separated and the training was stopped by applying early stopping if the validation-loss did not decrease even after 10 loops. The data used in the training the model are 13,530 samples as training data, 1,692 validation data and 1,720 test data. As a result of measuring the performance of the model, we showed the accuracy of 0.998 for the test data.

**Table 2. Data Description**

| Data | Cancer | Normal |
|---|---|---|
| Train & Validation | 8648 | 6574 |
| Test | 953 | 666 |

### Cycle-Consistent Adversarial Network

Cycle GAN consists of two Generators and two Discriminators. Generator G is a generator that converts normal data into fake cancer data. Generator F is a function that converts cancer data into fake normal data. To study this network, Cycle GAN adds two Discriminator X and Discriminator Y and adds a loss of cycle loss.

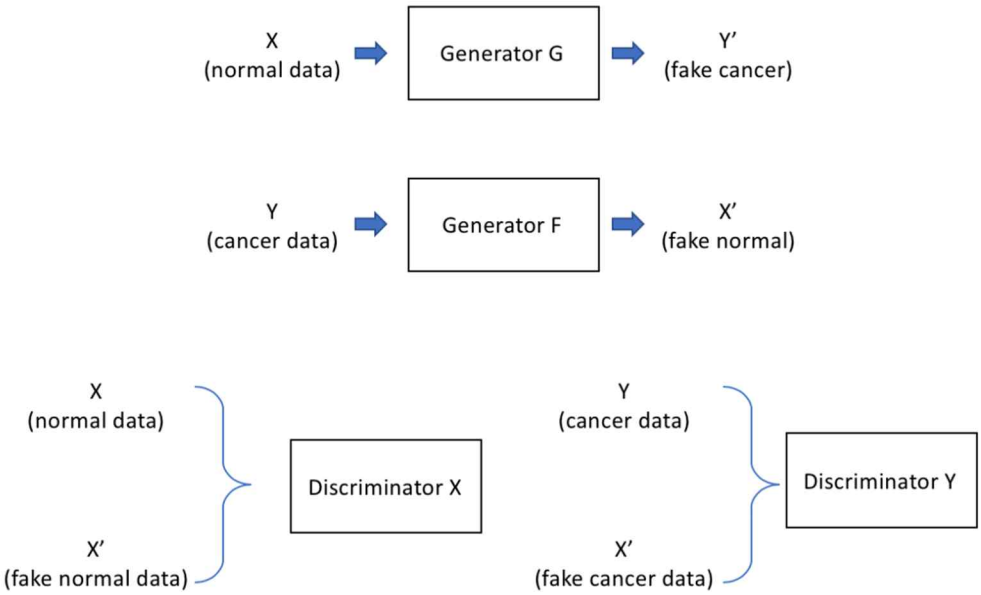


**Figure 2. Structure of Cycle GAN Network**

Generator G and F learn the following objective functions in a minimax game. We learned the following objective function for 10 epochs and 100 for batch size. $L_{GAN}$ is an objective function of the basic GAN Network, and $L_{cyc}$ is a function that maintains the characteristic of X and turns it into Y. The value of λ, which determines the importance of cycle loss, is set to 0.001. The larger the λ, the stronger the tendency to preserve the properties of the original domain.

*I.* $$G^*, F^* = arg\ min_{G,F} max_{D_X,D_Y} L(G, F, D_X, D_Y)$$

*II.* $$L(G, F, D_X, D_Y) = L_{GAN}(G, D_Y, X, Y) + L_{GAN}(F, D_X, Y, X) + \lambda L_{cyc}(G, F)$$

*III.* $$L_{cyc}(G, F) = E_{x\sim p(x)}[||F(G(x) - x||_1] + E_{y\sim p(y)}[||G(F(y) - y||_1]$$

*IV.* $$L_{GAN}(G, D_Y, X, Y) = E_{y\sim p(y)}[log\ D_Y(y)] + E_{x\sim p(x)}[log\ (1 - D_Y(G(x))]$$

*V.* $$L_{GAN}(F, D_X, X, Y) = E_{x\sim p(x)}[log\ D_X(x)] + E_{y\sim p(y)}[log\ (1 - D_X(F(y))]$$

The structure of Generator G, F consists of one input layer (308 nodes), four hidden layers (100, 50, 50, 100 nodes) and the last output layer (308 nodes). At this time, ReLu activation function is applied to the hidden layer. The structure of Discriminator X and Y is as follows. One input layer (308 nodes) consists of three hidden layers (150, 100, 50) and the last output layer (1 node). Similarly, the ReLu activation function is applied to the hidden layer.

The data used in the study consisted of 6,300 normal data and 6,300 cancer data among the non-paired data of the platform data of The Cancer Gene Atlas (TCGA), TARGET and Genotype-Tissue Expression (GTEx). Here, the meaning of not being paired means that there is no normal data paired with cancer. In the case of TCGA, the patient data of the cancer patients and the normal data adjacent to the tissues are also included. This case is called paired data. There are two test data used to validate the training. First, the experimental results were verified using 700 cancer data and 700 normal data among the unpaired data not used for learning, and the results were verified by using 600 pairs of paired data.

## 3. RESULTS

First, we tried to deceive the Pretrained Discriminator using Cycle GAN which was the first research objective. The results are as follows. The Pretrained Discriminator is a well-trained model with an accuracy of 0.998 in the test data mentioned in the Experiment.

**Table 3. Unpaired Test Data (700 normal samples, 700 cancer samples)**

| Cancer Code | Numbers of Generated Cancer | Numbers of Generated Normal | Total Accuracy |
|---|---|---|---|
| HNSC | 0 | 44 | 1 |
| BRCA | 0 | 76 | 1 |
| BLCA | 0 | 23 | 1 |
| COAD | 0 | 21 | 1 |
| LUAD | 0 | 63 | 1 |
| SARC | 0 | 17 | 1 |
| THCA | 0 | 44 | 1 |
| STAD | 1 | 32 | 1 |
| KIRC | 0 | 40 | 1 |
| LUSC | 0 | 44 | 1 |
| LIHC | 0 | 27 | 1 |
| PRAD | 0 | 36 | 1 |
| ESCA | 0 | 7 | 1 |
| Cholangiocarcinoma | 0 | 1 | 1 |
| PAAD | 0 | 14 | 1 |
| CESC | 0 | 28 | 1 |
| UCEC | 3 | 13 | 1 |
| SKCM | 0 | 48 | 1 |
| ACC | 0 | 12 | 1 |
| Testicular Germ Cell | 0 | 16 | 1 |
| DLBC | 0 | 3 | 1 |
| OV | 0 | 71 | 1 |
| GBM | 1 | 10 | 1 |
| MESO | 0 | 10 | 1 |
| NON-CANCER | 695 | 0 | 1 |

**Table 4. Paired Test Data (600 normal samples, 600 cancer samples)**

| Cancer Code | Numbers of Generated | Numbers of Generated | Total Accuracy |
|---|---|---|---|

| | Cancer | Normal | |
|---|---|---|---|
| HNSC | 43 | 43 | 1 |
| BRCA | 111 | 111 | 1 |
| BLCA | 19 | 19 | 1 |
| COAD | 26 | 26 | 1 |
| LUAD | 62 | 62 | 1 |
| SARC | 2 | 2 | 1 |
| THCA | 57 | 57 | 1 |
| STAD | 26 | 26 | 1 |
| KIRC | 72 | 72 | 1 |
| LUSC | 48 | 48 | 1 |
| LIHC | 50 | 50 | 1 |
| PRAD | 52 | 52 | 1 |
| ESCA | 8 | 8 | 1 |
| Cholangiocarcinoma | 9 | 9 | 1 |
| PAAD | 4 | 4 | 1 |
| CESC | 3 | 3 | 1 |
| UCEC | 7 | 7 | 1 |
| SKCM | 1 | 1 | 1 |

Surprisingly, rather, the fake samples generated by the generator were perfectly classified in the pretrained discriminator. Completely classified means that all generated fake normal data are predicted as normal, and all generated fake cancer are predicted as cancer. In other words, the generator changed completely to normal in the case of cancer and completely changed to cancer in the case of normal. In this way, we can confirm the first research objective, that the network actually makes the cancer data to fake normal data and makes the normal data into fake cancer data successfully.

To verify the second goal, we used a dimensional reduction technique called t-SNE. t-SNE is used to confirm whether the generated normal maintains the characteristics of the original cancer.

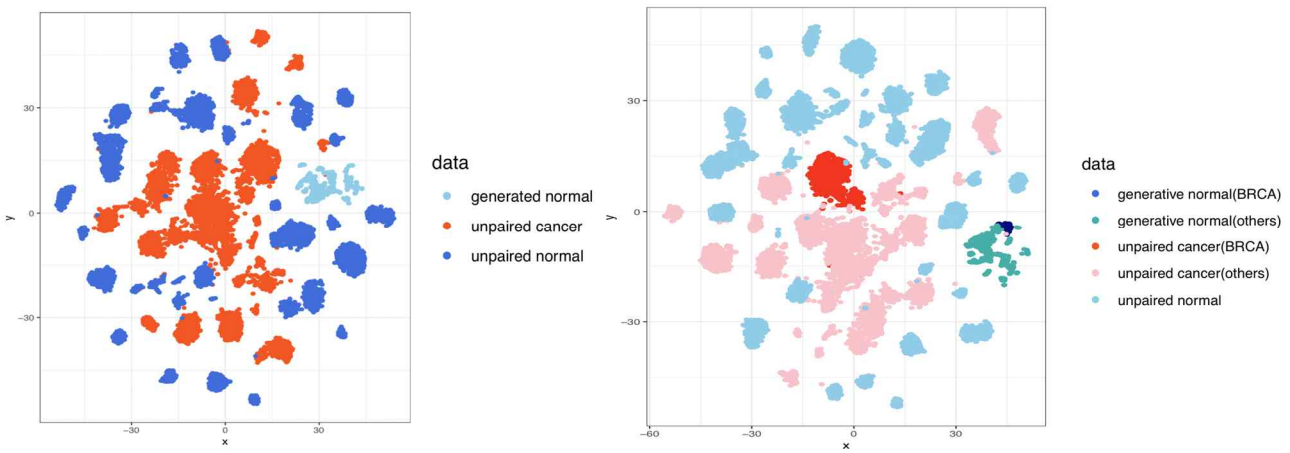


**Figure 3. t-SNE Unpaired Test Data (700 normal samples, 700 cancer samples)**

First, in the case of the generated normal, we assumed that normal data preserve the characteristics of the unpaired cancer data. However, we could see that the data were transformed in a way that the generated normal data were gathered together and were not formed differently according to the characteristics of the cancer

tissues. This is confirmed by calculating t-SNE for the large number of samples.

According to our expectation, the generated fake normal (generated from BRCA cancer) of the navy should be close to the distribution of BRCA cancer, and the result shows that it is not closely distributed.

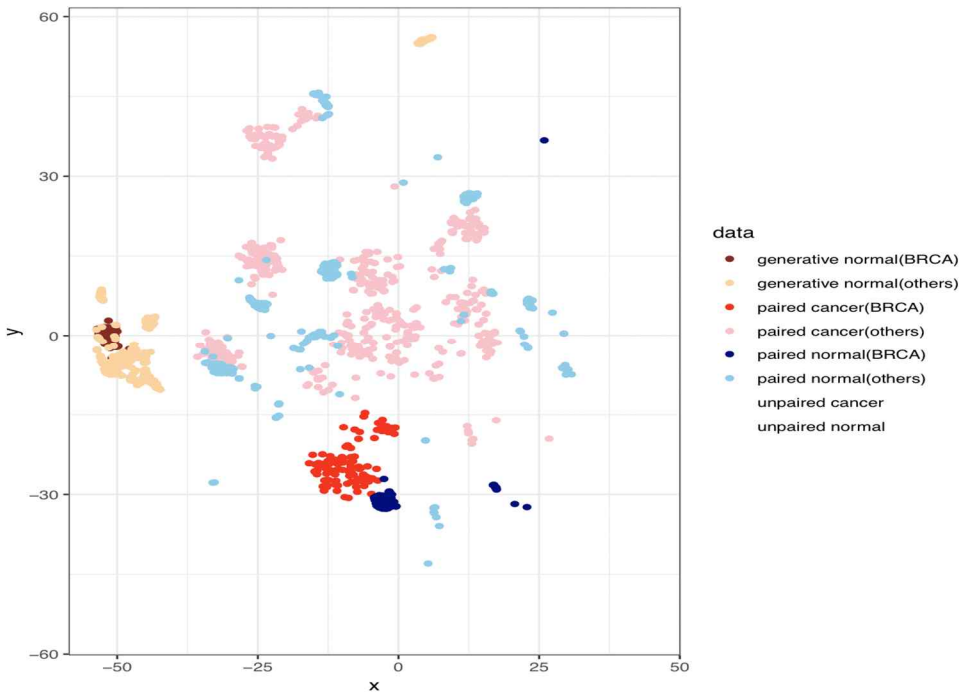


**Figure 4. t-SNE Paired Test Data (600 normal samples, 600 cancer samples)**

Similar results were obtained in the paired samples. The expecting results were that the dark brown, generated normal (generated from BRCA cancer) should have a similar distribution to the dark blue paired normal (BRCA), but not so. Therefore, we failed to achieve the second objective we aimed at, the generated data preserves the characteristics of the original data well.

We think that this is because the cycle loss of the cycle GAN is not learning the network in the direction we want. To solve this problem, we got the idea from the relationship of paired data. The data distribution shows that the distance between the paired samples is very close. In view of this, we propose a new objective function that modified $L_{cyc}$.

$$G^*, F^* = arg\ min_{G,F} max_{D_X,D_Y} L(G, F, D_X, D_Y) \quad VI.$$

$$L(G, F, D_X, D_Y) = L_{GAN}(G, D_Y, X, Y) + L_{GAN}(F, D_X, Y, X) + \lambda L_{cyc*}(G, F) \quad VII.$$

$$L_{cyc*}(G, F) = E_{x\sim p(x)}[||G(x) - x||_1] + E_{y\sim p(y)}[||F(y) - y||_1] \quad VIII.$$

$$L_{GAN}(G, D_Y, X, Y) = E_{y\sim p(y)}[log\ D_Y(y)] + E_{x\sim p(x)}[log\ (1 - D_Y(G(x))] \quad IX.$$

$$L_{GAN}(F, D_X, X, Y) = E_{x\sim p(x)}[log\ D_X(x)] + E_{y\sim p(y)}[log\ (1 - D_X(F(y))] \quad X.$$

## 4. Conclusion

Cancer is still one of the most life-threatening disease as of today. Understanding molecular profile of cancer is the essential clue for personalized treatment. In this paper, we build a method that generate normal gene

expression when a gene expression of cancer is given using Cycle GAN technology. Our method was able to generate gene expression data of normal which deceives the discriminator that judges 'cancer' or 'normal' from gene expression data. However, in our experiments, generated gene expressions classified as normal are not similar to the gene expressions of true normal samples. This shows more effort should be made to deceive discriminator with truly normal like gene expression pattern. To achieve this goal, we suggested a new cost function which hopefully generate more true normal like gene expression patterns from a given cancer sample.

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