

# Study and Analysis of Microarray Denoising Using Systholic Boolean Orthonormalizer Network in Wavelet Domain

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## ABSTRACT

In this paper, we present a new approach to deal with the noise inherent in the microarray image processing procedure. The method is based on the following procedure: We apply 1) Bidimensional Discrete Wavelet Transform (DWT-2D) to the Noisy Microarray, 2) scaling and rounding to the coefficients of the highest subbands (to obtain integer and positive coefficients), 3) bit-slicing to the new highest subbands (to obtain bit-planes), 4) then we apply the Systholic Boolean Orthonormalizer Network (SBON) to the input bit-plane set and we obtain two orthonormal output bit-plane sets (in a Boolean sense), we project a set on the other one, by means of an AND operation, and then, 5) we apply re-assembling, and, 6) rescaling. Finally, 7) we apply Inverse DWT-2D and reconstruct a microarray from the modified wavelet coefficients. Denoising results compare favorably to the most of methods in use at the moment.

## 1. INTRODUCTION

A microarray is affected by noise in its acquisition and processing. Microarray denoising is used to remove the additive noise while retaining as much as possible the important image features. In the recent years there has been an important amount of research on wavelet thresholding and threshold selection for bioimages denoising, e.g., microarray images [1], [2], because wavelet provides an appropriate basis for separating noisy signal from the image signal. The motivation is that as the wavelet transform is good at energy compaction, the small coefficients are more likely due to noise and large coefficient due to important signal features [3]-[5]. These small coefficients can be thresholded without affecting the significant features of the image.

In general, the results of the microarray processing combine two sample images that after further image processing, gene expression data can be produced for further analysis, such as gene clustering or identification [1], [2]. These three crucial steps, experiment, image processing and data analysis, determine the success or not of the microarray analysis. Image processing plays a potentially large impact on the subsequent analysis. In recent years, a large number of commercial tools have been developed in microarray image processing [1], [2]. The tasks of all these tools mainly focus on two major targets, namely: spot segmentation and spot intensity extraction. However, the quality of the images from the experiments is not always perfect. The gene array experiments involve a large number of error-prone steps which lead to a high level of noise in the resulting images [1], [2]. Hence, the accuracy of the gene expressions derived from these images will largely be affected in the process.

In order to assure the accuracy of the gene expression, normally the replicated experiments and incorporated statistical methods are needed to estimate the errors [1], [2].

These methods deal mainly with measurement error, such as preparation of the sample, cross hybridization, and fluctuation of fluorescence value from gene to gene. But none deals particularly with the effect of the noise [1], [2].

In fact, the thresholding technique is the last approach based on wavelet theory to provide an enhanced approach for eliminating such noise source and ensure better gene expression. Thresholding is a simple non-linear technique, which operates on one wavelet coefficient at a time. In its basic form, each coefficient is thresholded by comparing against threshold, if the coefficient is smaller than threshold, set to zero; otherwise it is kept or modified. Replacing the small noisy coefficients by zero and inverse wavelet transform on the result may lead to reconstruction with the essential signal characteristics and with less noise. Since the work of Donoho & Johnstone [5], there has been much research on finding thresholds, however few are specifically designed for images [3], [4], [6].

## 2. BIDIMENSION DWT AND SBON

### 2.1 Bidimensional Discrete Wavelet Transform

The Bidimensional Discrete Wavelet Transform (DWT-2D) [8]-[17] corresponds to multiresolution approximation expressions. In practice, multiresolution analysis is carried out using 4 channel filter banks composed of a low-pass and a high-pass filter and each filter bank is then sampled at a half rate (1/2 down sampling) of the previous frequency. By repeating this procedure, it is possible to obtain wavelet transform of any order. The down sampling procedure keeps the scaling parameter constant (equal to  $\frac{1}{2}$ ) throughout successive wavelet transforms so that it benefits for simple computer implementation. In the case of an image, the filtering is implemented in a separable way by filtering the lines and columns.

### 2.2 Synthetic Boolean Orthonormalizer Network

The SBON was introduced by Mastriani [19] as a Boolean Orthonormalization Process (BOP) to convert a nonorthonormal Boolean basis, i.e., a set of non-orthonormal binary vectors (in a Boolean sense) to an orthonormal Boolean basis, i.e., a set of orthonormal binary vectors (in a Boolean sense). The BOP algorithm has a lot of fields of applications, e.g.: Steganography, Hopfield Networks, Boolean Correlation Matrix Memories, Bi-level image processing, lossy compression, iris, fingerprint and face recognition, improving edge detection and image segmentation, among others. That is to say, all those applications that need orthonormality in a Boolean sense. It is important to mention that the BOP is an extremely stable and fast algorithm.

### 2.3 Noise Sources and Its Statistical Measurement in Microarray Imaging

It is well known microarray technology can monitor thousand of DNA sequences in a high density array on a glass. The basic procedure for a microarray experiment is simply described as follow. Two mRNA samples are reverse-transcribed into cDNA, labeled using different fluorescent dyes (e.g., the red fluorescent dye Cy5 and the green fluorescent dye Cy3), then mixed and hybridized with the arrayed DNA sequences. After this competitive hybridization, the slides are imaged using a scanner which makes fluorescence measurement for each dye. From the differential hybridization of the two samples, the relative abundance of the spotted DNA sequences can be assessed.

Exciting methods to reduce the noise source include using clean glass slide and using a higher laser power rather than higher PMT voltages. However, there are not adequate for the required image qualities and an enhanced software procedure embedded within the process in a much better alter-native. Here, we focus on the implementation of the SBON method (in wavelet domain) to the denoising on microarray images [2]. Yet there are some fundamental obstacles that need clarification before the full potential of microarrays can be explored. One of the major problems in interpretation of microarray data is that different clustering techniques produce different results.

On the other hand, the assessment parameters that are used to evaluate the performance of noise reduction [20], [21] are the following ones:

Average Absolute Difference (AAD):

$$AAD = \frac{\sum_{r,c} |I(r,c) - I_d(r,c)|}{R * C} \quad (1)$$

Peak Signal to Noise Ratio (PSNR):

$$PSNR = \frac{R * C * \max_{r,c} (I(r,c))^2}{\sum_{r,c} (|I(r,c) - I_d(r,c)|)^2} \quad (2)$$

Signal to Noise Ratio (SNR):

$$SNR = \frac{\sum_{r,c} I(r,c)^2}{\sum_{r,c} (|I(r,c) - I_d(r,c)|)^2} \quad (3)$$

Image Fidelity (IFy):

$$IFy = 1 - \frac{1}{SNR} \quad (4)$$

Correlation Quality (CQy):

$$CQy = \frac{\sum_{r,c} I(r,c) * I_d(r,c)}{\sum_{r,c} I(r,c)} \quad (5)$$

Structural Content (SCt):

$$SCt = \frac{\sum_{r,c} I(r,c)^2}{\sum_{r,c} I_d(r,c)^2} \quad (6)$$

Where for an image of  $R * C$  (rows-by-columns) pixels,  $r$  means row,  $c$  means column,  $I$  means original image (without

noise), and  $I_d$  means denoised image. Such as, a lower AAD gives a “cleaner” image as more noise is reduced; larger SNR and PSNR indicates a smaller difference between the original (without noise) and denoised image; if IFy and SCt spread at 1, we will obtain an image  $I_d$  of better quality; and a larger value of CQy usually corresponds to a better quantitative performance [20], [21].

### 3. LITERATURE SURVEY

Microarrays have become the tool of choice for the global analysis of gene expression. Powerful statistical tools are now available to analyze this expression and to gain an understanding of how changes in gene expression patterns impact biological systems. Innovations have been made since the origin of this imaging technique which dates back to the 1970s [23]. The analysis of such data has become a computationally- intensive task that requires technological developments at various stages, from the design of the array, to image analysis, database storage, data processing and clustering and information extraction. Further progress has been made in biology research and genomic clinical diagnosis. In recent years, a large number of commercial tools have been developed for microarray image processing. Microarray arrays are a scientific tool that should be viewed in a similar fashion to any other laboratory technique with careful experimental planning, replication, and proper statistical analysis. A lot of research has been conducted to examine these data with statistical techniques to help discern possible patterns in the data.

Among other approaches used for microarray image denoising and enhancement are the iterative normalization of cDNA microarray data [24] and the image reconstruction technique (IRT) introduced by O’Neill and Magoulas [25].

A Method based on fuzzy vector filtering has also been used for processing microarray images [26].

Other methods based on the wavelet transform [27] and cellular networks [28] have also been used for this purpose [29, 30].

Wang et al. [31] proposed enhancing the microarray image using the stationary wavelet transform (SWT) [32].

Another approach based on a variant of the translation-invariant wavelet transform was proposed by Adjeroh [33].

### 4. DISCUSSION

The simulations demonstrate that the SBON technique improves the noise reduction performance to the maximum, for bioimages. Here, we present a set of experimental results using two bioimages. Such images were converted to bitmap file format for their treatment [22].

For statistical filters employed, i.e., Median, Lee, Kuan, Gamma-Map, Enhanced Lee, Frost, Enhanced Frost, Wiener, DS, and EDS, we use a reduction scheme [22]. Figure 2 shows the noisy (30 %) and filtered microarray images used in the first experiment of [1], with a 274-by-274 (pixels) by 65536 (gray levels) bitmap matrix. Table 1 summarizes the assessment parameters vs. 19 filters for Fig. 2, where En-Lee means Enhanced Lee Filter, En-Frost means Enhanced Frost Filter, ST means Soft-Thresholding, HT means Hard-Thresholding and SST means Semi-Soft-Thresholding.

The assessment parameters were applied to the whole image. Figure 3 shows the noisy (10 %) and filtered microarray images used in the second experiment of [1], with a 256-by-256 (pixels) by 65536 (gray levels) bitmap matrix. Table 2

summa-rizes the assessment parameters vs. 19 filters for Figure 2. In both cases, the bioimages were processed by using 10 statistical filters, VisuShrink with Daubechies 4 wavelet basis and 1 level of decomposition (improvements were not noticed with other basis of wavelets) [2], [3], [5], [6], [22], SureShrink, Oracle-Shrink, BayesShrink, NormalShrink, TNN [5]-[7], [22], and SBON respectively. Figures 2 and 3 summarize the edge preservation performance of the SBON technique vs. the rest of the filters with a considerably acceptable computational complexity. A 3-by-3 kernel was employed for all statistic noise filters. For *TNN* [7] the empirical function parameter value  $\lambda = 0.01$ .

For Lee, Enhanced Lee, Kuan, Gamma, Frost and Enhanced Frost filters the damping factor is set to 1, see [3], [22]. The quantitative results of Table 1 and 2 shows that the *SBON* technique can eliminate noise without distorting useful image information and without destroying the important image edges.

Also, in the experiment, the *SBON* outperformed the conventional and no conventional noise reducing filters in terms of edge preservation measured by Pratt figure of merit [21]. In nearly every case in every homogeneous region, the *SBON* produced the lowest standard deviation and was able to preserve the mean value of the region.

The numerical results are further supported by qualitative examination, as shown in Fig. 2 and 3.

On the other hand, all filters was applied to complete image, for Figure 2 (274-by-274) pixels and Figure 3 (256- by-256) pixels, and all the filters were implemented in MATLAB® (Mathworks, Natick, MA) on a PC with an Athlon (2.4 GHz) processor.

## 5. CONCLUSION

In this paper we have developed a *SBON* technique based tools for removing additive noise in microarrays. The simulations show that the *SBON* have better performance than the most commonly used filters for microarrays (for the studied benchmark parameters) which include statistical filters, wavelets, and a version of *TNN*. The *SBON* exploits the local coefficient of variations in reducing noise. The performance figures obtained by means of computer simulations reveal that the *SBON* technique provides superior performance in comparison to the above mentioned filters in terms of smoothing uniform regions and preserving edges and features. The effectiveness of the technique encourages the possibility of using the approach in a number of ultrasound and radar applications. Besides, the method is computationally efficient and can significantly reduce the noise while preserving the resolution of the original microarray image. Considerably increased Pratt’s figure of merit strongly indicates improvement in detection performance. Also, cleaner images suggest potential improvements for classification and recognition. On the other hand, the drawback of applying the developed *SBON* technique for removing additive noise in microarrays is the increase in the computational complexity, for blame of the slicing process.

Finally, the natural extension of this work is in Synthetic Aperture Radar (SAR) images, as well as in multimedial applications.

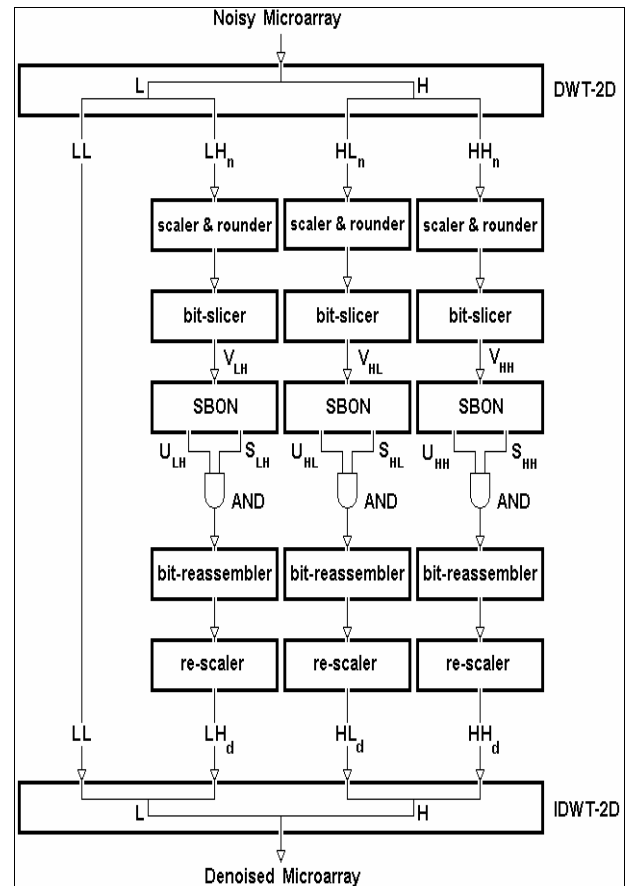
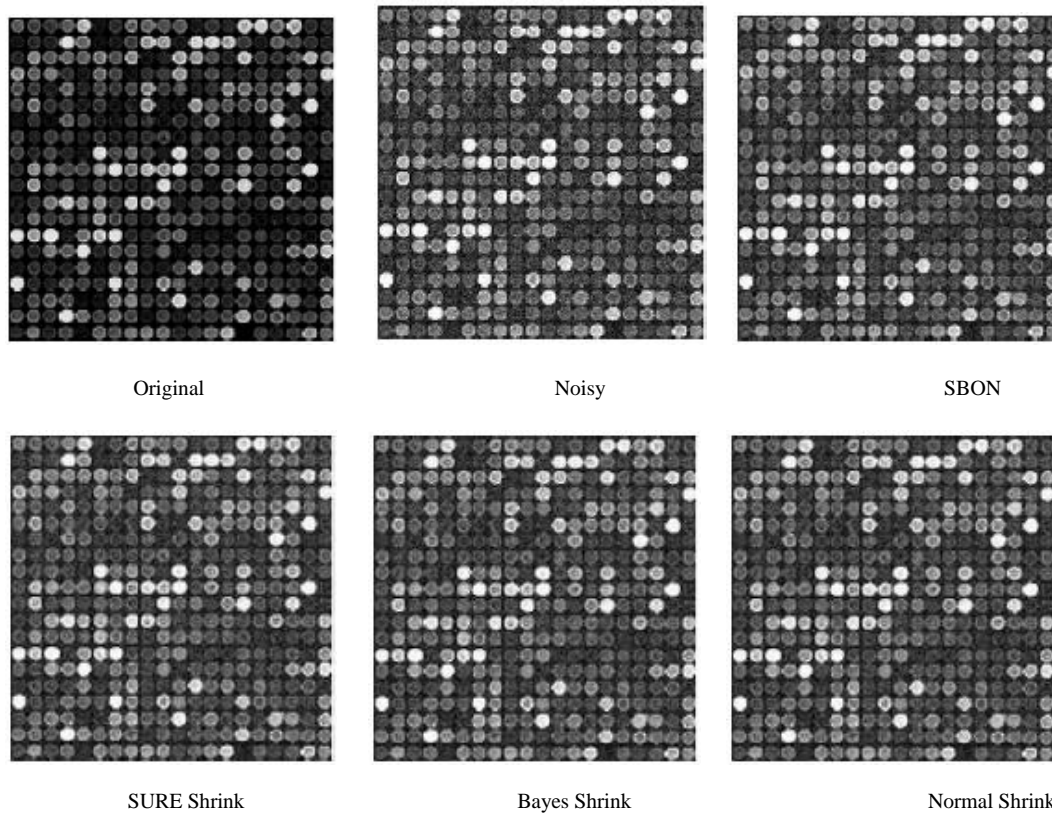


Fig.1 Microarray Denoising

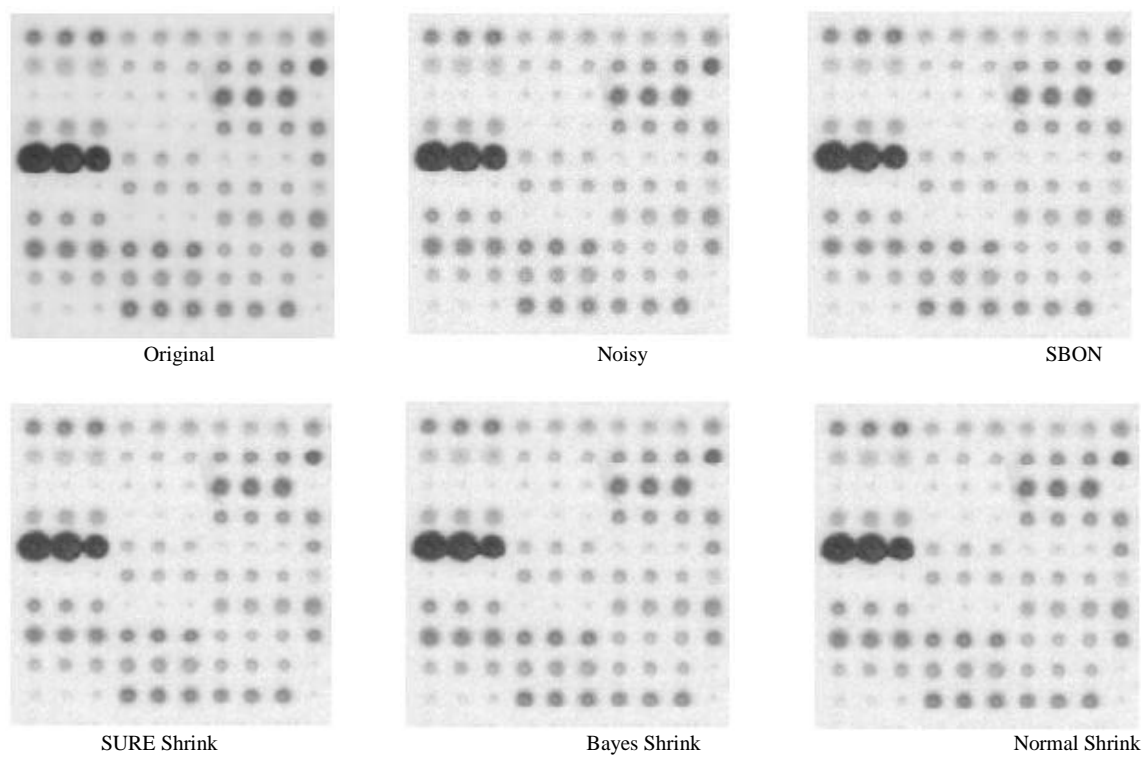
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**Fig. 2 Original, noisy and filtered images**



**Fig. 3 Original, noisy and filtered images**  
**TABLE I ASSESSMENT PARAMETERS VS. FILTERS FOR FIGURE 2**

| Filter           | Assessment Parameter |        |         |        |          |        |         |
|------------------|----------------------|--------|---------|--------|----------|--------|---------|
|                  | AAD                  | SNR    | PSNR    | IF     | CQ       | SC     | FOM     |
| En-Frost         | 38.2653              | 3.4464 | 33.7364 | 0.7109 | 150.7467 | 0.5663 | 0.39857 |
| En-Lee           | 39.7437              | 3.3363 | 33.8373 | 0.7112 | 150.7472 | 0.5632 | 0.49876 |
| Frost            | 38.4374              | 3.2423 | 33.7033 | 0.7106 | 150.5244 | 0.5689 | 0.48756 |
| Lee              | 39.2427              | 3.4242 | 32.6363 | 0.7015 | 150.4141 | 0.5924 | 0.43447 |
| Gamma            | 39.6252              | 3.1112 | 33.2703 | 0.7063 | 150.1918 | 0.5751 | 0.44235 |
| Kuan             | 39.8224              | 3.1243 | 31.8272 | 0.7041 | 149.3121 | 0.5715 | 0.45342 |
| Median           | 39.5252              | 3.1131 | 32.7916 | 0.6852 | 148.9172 | 0.5896 | 0.40704 |
| Wiener           | 39.1829              | 3.4557 | 33.7033 | 0.7106 | 150.5244 | 0.5689 | 0.44236 |
| DS               | 38.7332              | 3.4657 | 33.9997 | 0.7169 | 150.9898 | 0.5599 | 0.64111 |
| EDS              | 38.1484              | 3.6969 | 34.1315 | 0.7182 | 151.5252 | 0.5612 | 0.64324 |
| VisuShrink (ST)  | 39.1450              | 3.4596 | 33.7412 | 0.7109 | 151.1527 | 0.5657 | 0.44382 |
| VisuShrink (HT)  | 38.8612              | 3.5283 | 34.4115 | 0.7166 | 151.3316 | 0.5666 | 0.44324 |
| VisuShrink (SST) | 38.1829              | 3.5557 | 34.7033 | 0.7196 | 151.9202 | 0.5612 | 0.46432 |
| SureShrink       | 38.1612              | 3.5751 | 34.7193 | 0.7198 | 151.9244 | 0.5611 | 0.43322 |
| OracleShrink     | 38.1189              | 3.6957 | 34.7233 | 0.7198 | 151.9844 | 0.5619 | 0.45534 |
| BayesShrink      | 38.1145              | 3.6968 | 34.7237 | 0.7199 | 151.9953 | 0.5612 | 0.46329 |
| NormalShrink     | 38.1098              | 3.6998 | 34.8734 | 0.7199 | 151.9983 | 0.5609 | 0.59333 |
| TNN              | 38.1008              | 3.7157 | 34.8833 | 0.7199 | 151.9992 | 0.5600 | 0.65432 |
| SBON             | 37.7155              | 3.7772 | 36.8388 | 0.7353 | 155.4613 | 0.5513 | 0.69123 |

**TABLE II ASSESSMENT PARAMETERS VS. FILTERS FOR FIGURE 3**

| Filter           | Assessment Parameter |          |          |        |          |        |         |
|------------------|----------------------|----------|----------|--------|----------|--------|---------|
|                  | AAD                  | SNR      | PSNR     | IF     | CQ       | SC     | FOM     |
| En-Frost         | 12.4747              | 290.1324 | 363.6712 | 0.9830 | 226.4744 | 0.8972 | 0.41265 |
| En-Lee           | 12.8474              | 290.2522 | 363.9321 | 0.9883 | 226.8373 | 0.8932 | 0.51986 |
| Frost            | 12.1847              | 290.2772 | 363.0233 | 0.9828 | 226.3272 | 0.8923 | 0.55312 |
| Lee              | 12.3733              | 290.2333 | 363.0238 | 0.9838 | 226.2822 | 0.8943 | 0.44421 |
| Gamma            | 12.3830              | 290.8331 | 363.3433 | 0.9882 | 226.8383 | 0.8934 | 0.51235 |
| Kuan             | 12.3833              | 290.8272 | 363.4923 | 0.9887 | 226.8381 | 0.8934 | 0.54129 |
| Median           | 12.9973              | 289.1212 | 361.8374 | 0.9673 | 225.9287 | 0.8734 | 0.51286 |
| Wiener           | 11.9042              | 290.8635 | 363.5568 | 0.9866 | 226.8901 | 0.8954 | 0.56413 |
| DS               | 11.4572              | 290.9950 | 363.9393 | 0.9898 | 226.9723 | 0.8993 | 0.64213 |
| EDS              | 11.5792              | 290.9998 | 363.9865 | 0.9899 | 226.9975 | 0.8993 | 0.64449 |
| VisuShrink (ST)  | 11.9055              | 289.2367 | 361.5523 | 0.9761 | 222.7564 | 0.8872 | 0.51228 |
| VisuShrink (HT)  | 11.9042              | 290.8673 | 363.5615 | 0.9966 | 226.8909 | 0.8976 | 0.56424 |
| VisuShrink (SST) | 11.7864              | 290.9546 | 363.9822 | 0.9975 | 226.8937 | 0.8984 | 0.56389 |
| SureShrink       | 11.7074              | 291.0753 | 363.8343 | 0.9991 | 226.8942 | 0.8991 | 0.57432 |
| OracleShrink     | 11.8436              | 290.9332 | 363.7363 | 0.9968 | 226.8963 | 0.8983 | 0.55234 |
| BayesShrink      | 11.9353              | 290.9363 | 363.7361 | 0.9923 | 226.8942 | 0.8962 | 0.56328 |
| NormalShrink     | 11.6875              | 290.9992 | 363.9353 | 0.9992 | 226.9021 | 0.8999 | 0.59611 |
| TNN              | 11.4447              | 291.7243 | 363.9991 | 0.9994 | 226.9732 | 0.9002 | 0.62900 |
| SBON             | 10.9071              | 294.9237 | 383.1090 | 0.9992 | 229.8972 | 0.9173 | 0.69322 |