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Application of Ant Colony Optimization for Image Segmentation

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Introduction

Swarm intelligence approach in solving complicated optimization problems is relatively new. The main advantage of swarm intelligence approach is that system of simple communicating agents is capable of solving complex problems [1, 2]. Ant Colony Optimization (ACO) being a branch of swarm intelligence [3, 4] is here considered and its use for important biomedical image processing application is investigated.

In proteomics two-dimensional electrophoresis (2DE) is used for proteins separation according to their isoelectric point and molecular mass. The result of two-dimensional electrophoresis process is a gel. Usually it is scanned with specialized high sensitivity scanner for the further computerized image analysis. Even in a laboratory environment received images of 2DE gels vary in resolution, their geometry and can be affected by various artifacts [5, 6].

A wide dynamic range of intensity magnitudes of protein spots in gel images, reaching seven or eight orders of magnitude, complicates automatic gel image analysis. Moreover, various artifacts prohibits from correct image segmentation and spot quantification. Gel staining is used in order to increase the visibility of individual proteins. Unfortunately, the application of any staining method for protein spot detection can also result in image artifacts, e. g., if staining reagent is overdosed or development time is prolonged then truncated spots with saturation effects in gel images can appear.

Correct segmentation and identification of protein spots in two-dimensional electrophoresis gel images is of vital importance as it gives information about all proteins existed in the sample under investigation. That knowledge is crucial for development of modern diagnostics and prognostics systems, intelligent and personalized drugs, etc.

In this paper an ability of Ant Colony Optimization technique [3, 7–9] for two-dimensional electrophoresis gel image segmentation [5, 6] is investigated. We start with introductory presentation of selected ACO model – describe model itself as also as modifications for its use for image segmentation. Then we present experimental setup – necessary steps of image preprocessing, synthetic and natural experimental data. Afterwards we show and comment the results of experimentation on the selection of suitable ACO model parameters. Finally we present results of Ant Colony Optimization use for segmentation of two-dimensional electrophoresis gel images and discuss possible further ways for segmentation improvement.

Model of Ant Colony Optimization

The initial model idea was proposed by Bocchi, Ballerini and Hässler [10], while population control model was proposed by Fernandes, Ramos and Rosa [11]. Let us here briefly present selected ACO model.

Ants are supposed to be moving over the grayscale image. Doing so, each ant can occupy only one cell, moreover only one ant can be in one cell. Each ant has certain associated with it probability to move to unoccupied region and to leave a pheromone trace. If all adjacent cells are occupied by other ants, the ant will stay in its cell. In a course of processing ants can die and can reproduce (that will be discussed latter).

During initialization ants are placed on grid in random way. Then ants are moved by iteration. Probability of ant to move and movement direction are chosen by evaluating earlier direction of travel, surrounding ants and pheromone level.

Ant movement direction coefficient $w(\Delta_\theta)$ is taken according to data presented in Fig. 1. There Δ_θ is an angle of ant's deflection from its earlier direction of travel (45° multiple). In the figure it is assumed that earlier direction of ant travel was North.

1/2	1	1/2
1/4		1/4
1/12	1/20	1/12

Fig. 1. Ant movement direction coefficient $w(\Delta_\theta)$

Another associated with ant is coefficient $W(\sigma)$. It is related to pheromone concentration σ and is expressed as:

$$W(\sigma)=\left(1+\frac{\sigma}{1+\delta\sigma}\right)^{\beta}. \quad (1)$$

Here $1/\delta$ is pheromone sensor capacity of ant; β is coefficient describing how strong ant is attracted by pheromone.

So the probability for an ant to move to surrounding cell i from cell k is:

$$P_{ik}=\frac{W(\sigma_i)w(\Delta_\theta)}{\sum_{j=1}^{k}W(\sigma_i)w(\Delta_\theta)}. \quad (2)$$

At each iteration ant leaves some pheromone:

$$T=\eta+p\frac{\Delta_{\text{gl}}}{255}. \quad (3)$$

Here η is a constant amount of pheromone the ant leaves at each iteration; p is a constant determining the amount of pheromone placed due to image intensity gradient Δ_{gl}.

At the end of each iteration pheromone amount left by all ants is decreased (evaporated) by a constant K.

During initialization a fixed amount of energy $e(0)$ is assigned to each ant. Afterwards the energy amount is iteratively updated according to:

$$e(t)=e(t-1)-\alpha+\alpha\frac{\Delta_{\text{gl}}}{\max\Delta_{\text{gl}}}. \quad (4)$$

Here t is an iteration or time index; α is a constant; $\max\Delta_{\text{gl}}$ is a maximum gradient found by the current ant so far. Initial energy $e(0)$ is set to $1+\alpha$.

Reproduction is triggered when there is at least one ant in the neighborhood of the parent ant. Reproduction probability is calculated by:

$$P_{\text{R}}=R(n)\left(\mu+\frac{(1-\mu)\Delta_{\text{gl}}}{\max\Delta_{\text{gl}}}\right). \quad (5)$$

Here $R(n)$ is reproduction coefficient (dependency of reproduction coefficient's values on the number of occupied cells is shown in Fig. 2); n is a number of occupied by ants cells; μ is ants reproduction constant.

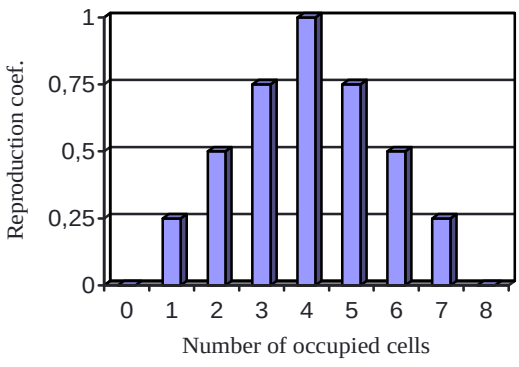


Fig. 2. Dependency of ant's reproduction coefficient on the number of occupied cells

ACO model for image segmentation

For image segmentation into multiple regions purposes each ant is assigned to a different colony. Ants from different colonies can crossover with the same probability as with ants from the same colony. New ant colony is chosen from the surrounding ants and parent ant colonies by a roulette-well method. Thus, more ants from one colony surrounds the parent ant, the greater is the probability for new ant to belong to that colony.

Main difference of ACO use for image segmentation is in pheromone deposition. Here the competition of ant colonies occurs. Each colony places a different type of pheromone. Because of the fact that in the same cell there can not be pheromone of different types, when an ant places pheromone in a cell that posses another colony pheromone, that pheromone level is decreased by amount T calculated by expression (3). When ant wants to place pheromone in the cell possessing pheromone of the same colony, then pheromone level is increased in a usual way. Specifically, pheromone level $F_{i,j}$ in a cell with coordinates i and j is presented by:

$$F_{i,j}(t)=\begin{cases}\left|F_{i,j}(t-1)-T\right|, & \text{different ant family;}\\ F_{i,j}(t-1)+T, & \text{the same ant family.}\end{cases} \quad (6)$$

When the pheromone vanishes (its level reaches zero), new ant's pheromone class is assigned to that cell.

Another difference of ACO use for image segmentation is related to expression (1) describing calculation of pheromone concentration coefficient $W(\sigma)$. Here it is expressed differently:

$$W_{\text{S}}(\sigma)=\begin{cases}\dfrac{1}{W(\sigma)}, & \text{different ant family;}\\ W(\sigma), & \text{the same ant family.}\end{cases} \quad (7)$$

From last expression becomes clear that ants from different colony will be repelled by a pheromone level and segmentation of area will occur.

Pheromone evaporation $\mathbf{F}(t)$ with K being evaporation constant is decided to be expressed by:

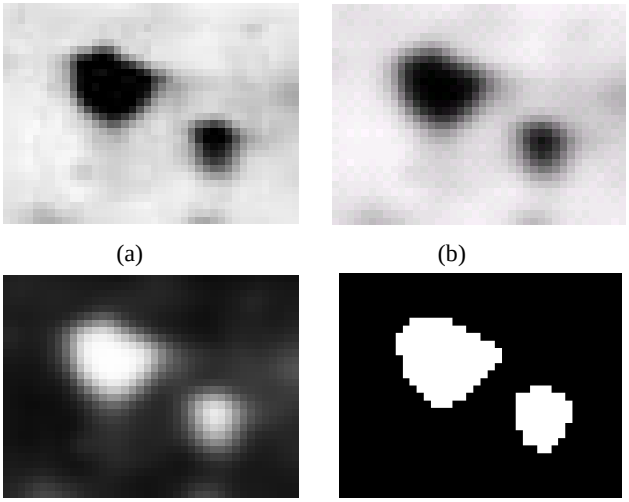
$$\mathbf{F}(t)=\frac{\mathbf{F}(t-1)}{K+1}. \quad (8)$$

Such evaporation helps to keep trace of ant colony on pheromone map even after all colony has died.

Image pre-processing

Let us describe image pre-processing steps necessary to be done before execution of ACO segmentation. The aim of image pre-processing is to remove speckle noise and to build image mask. The use of image mask in image segmentation by ACO is going to shorten processing time.

Initial image $\mathbf{S}$ is grayscale (for a fragment of natural two-dimensional electrophoresis gel image see Fig. 3 a). In



(a) (b) (c) (d)

Fig. 3. Illustration of image pre-processing steps: a) initial image; b) smoothed image; c) inverted image; d) binary image

order to remove noise and blur sharp edges, Gaussian filter is applied (Fig. 3 b). For this purpose we use kernel

$$\mathbf{G} = \frac{1}{8}\begin{bmatrix} 0 & 1 & 0 \\ 1 & 4 & 1 \\ 0 & 1 & 0 \end{bmatrix}. \quad (9)$$

In the next step image is inverted (Fig. 3 c):

$$I_{i,j} = \max(\mathbf{S}) - S_{i,j}. \quad (10)$$

The final step is preparation of binary (black and white) image mask $\mathbf{B}$. The mask will speed up calculations. Moreover, it will improve segmentation by removing ants and their pheromone traces from empty (without protein spots) image regions. Calculation of image mask is performed firstly by assigning initial value:

$$B_{i,j}(t) = I_{i,j}. \quad (11)$$

Then multiplication is applied for emphasizing protein spots:

$$B_{i,j}(t+1) = B_{i,j}(t) \cdot I_{i,j}, \quad t \in [1,4]. \quad (12)$$

Lastly mask is normalized:

$$B_{i,j} = \frac{B_{i,j}}{\max B_{i,j}} \cdot 255, \quad (13)$$

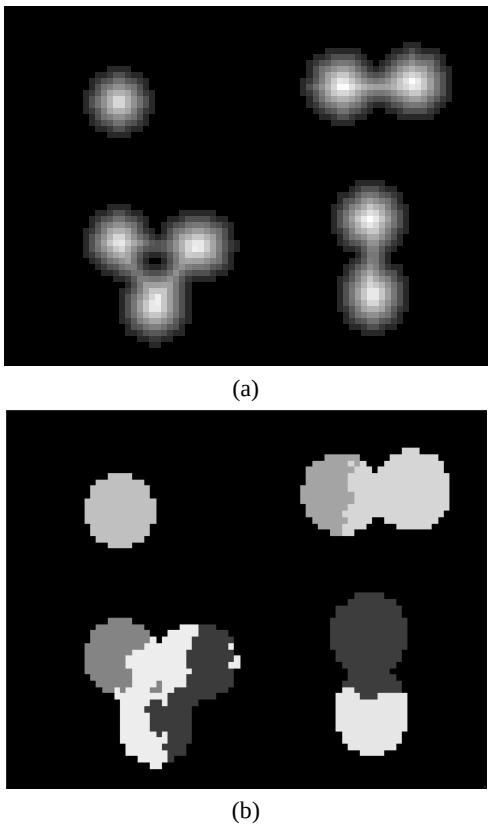
and threshold function is applied to make it binary:

$$B_{i,j} = \begin{cases} 0, & B_{i,j} < 2; \\ 1, & \text{otherwise.} \end{cases} \quad (14)$$

Final mask $\mathbf{B}$ is calculated from image $\mathbf{I}$. The result of this operation is presented in Fig. 3 d.

Results on the search of acceptable ACO parameters

Application of ACO for image segmentation (1–8) is controlled by 8 parameters. Final results of segmentation heavily depend on the selected values of ACO parameters.



(a) (b)

Fig. 4. Results of synthetic test image segmentation by ACO: a) pre-processed image; b) segmented image

Let us perform search for acceptable values of ACO parameters based on synthetic images.

Initial parameters of ACO are chosen according to [11]: $\alpha = 0.025$; $\beta = 3.5$; $\delta = 0.2$; $\eta = 0.07$; $p = 1.5$; $K = 0.01$; $\mu = 0.1$. Population size S is 30 % of the total image size. Simulation is performed in MATLAB environment.

For estimation of ACO model parameters ants behavior is simulated on synthetic test image presented in Fig. 4 a. Test image consists of 8 spots: one separate spot, two spots joined horizontally, two spots joined vertically, and three spots joined together.

A total of 3200 iterations are performed. After each 25's iteration a snapshot of a pheromone map is taken. During revision of snapshot maps it is easy to see tendency of ants' behavior.

After each second step the mask $\mathbf{B}$ is applied to the ants $\mathbf{A}$ and pheromone $\mathbf{F}$ matrixes:

$$A_{i,j} = A_{i,j} \cdot B_{i,j}, \quad (15a)$$

$$F_{i,j} = F_{i,j} \cdot B_{i,j}. \quad (15b)$$

During each experiment different parameters are tried in order to improve segmentation. Results are summarized in Table 1. Here N is the total number of segmented spots while acceptable values of parameters are outlined presenting them in bold font. Using these parameter values afterwards experiment on real world data is done and its result is presented in Fig. 5.

The main problem segmenting protein spots in two-dimensional electrophoresis gel images is that protein spots are often overlapped. Thus ACO model is also tested on images with overlapped protein spots.

Table 1. Results of estimation of acceptable values of ACO model parameters

Population size		Ant's reproduction		Ant's energy		Pheromone attraction		Ant's sensor limiter		Ant's pheromone		Gradient's sensitivity		Pheromone evaporation	
S %	N	μ	N	α	N	β	N	δ	N	η	N	p	N	K	N
10	0	0.05	4	0.0125	4	2	4	0.1	5	0.05	5	0.5	4	0.0005	4
20	2	**0.06**	**6**	**0.025**	**6**	2.5	5	**0.2**	**6**	0.06	5	1	5	**0.001**	**6**
30	**5**	0.07	6	0.035	4	3	4	0.3	4	**0.07**	**6**	**1.5**	**6**	0.0015	5
40	5	0.08	4	0.05	3	**3.5**	**6**	0.4	4	0.08	5	2	5	0.002	4
50	3	0.1	5	–	–	4	4	–	–	0.09	4	3	4	0.0005	4
–	–	0.2	4	–	–	5	4	–	–	–	–	–	–	–	–

Estimation of ACO parameters is an optimization process. Parameters are estimated using steepest descent method. Let us discuss how ants behave when different ACO parameter values are used.

Initial population size is one of the main factors that determine time of iteration. The more ants have populations the longer it takes to compute final result. When S is selected to be more than 30 % from image size, it causes a lot of ant colonies situated on the same spot and spot is over-segmented. It takes a long time until single ant colony overtakes one spot. When S is smaller than 30 % – calculations are performed quicker, however image covering is not dense enough and some protein spots may be missed by ants.

Increase of reproduction parameter μ causes ants to reproduce everywhere on image and population size remains very high for a long time. Often the same colony dominates on several spots. Decrease of μ results in quicker death of ants, because without reproduction ant colony eventually gets older and dies. Moreover protein spots are not fully covered by colony and often there are several ant colonies on the same protein spot.

With high values of ants' energy coefficient α ants can travel longer distance prior death at the same time through reproduction increasing the total number of ants. Ants of the same colony tend to occupy multiple spots. With low α values, ants die quickly and segmentation of image does not occur.

Greater sensitivity to pheromone β causes ants to spread around protein spots with greater probability to miss a top of a spot. With lower values of β ants just move according to gradient, competition decreases and protein spots are not fully covered by pheromone.

With low sensor capacity $1/\delta$ ants tends to spread and the same ant colony occupies nearest protein spots. With high sensor capacity, ants attracted by pheromone group heavily, protein spots are not segmented correctly as gradient influence is too weak.

With high η pheromone traces stay for a longer time and other ants can easier find spots. However protein spots are not segmented correctly as ants from a different colony are influenced by pheromone that is preventing ants to move according to the gradient of image intensity. As a result protein spots are not segmented correctly. When η is low, ant colony may be separated by another one and incorrect segmentation may occur.

High p emphasizes places near protein spots letting to place there more pheromone, but sensor capacity $1/\delta$ limits ants' sensitivity in high concentrations of pheromone. Low p causes spots to be unnoticed by ants. Ratio of η/p is important because low ratio let ants to be attracted by pheromone near protein spots, but very low or high η/p ratio will lead to lack or excess of pheromone so ants will be misled.

High evaporation of pheromone K leads to lower pheromone level and one colony can easily take over few spots. Low evaporation leads to high level of pheromone and ants can become confused and even attracted by smaller pheromone levels because of $1/\delta$.

Taking into account discussed ACO parameter influence factors, becomes evident that selection of ACO parameter values is a complex optimization process, thus mainly sub-optimal solutions are found.

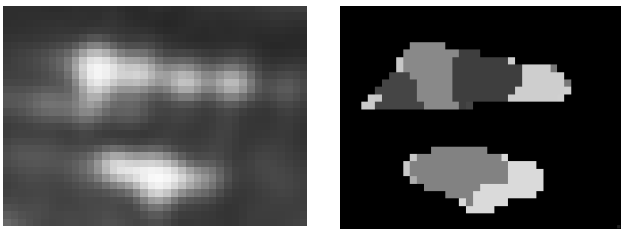
Results on 2DE gel image segmentation by ACO

Opposed to synthetic test image created by the use of Euclidian distance transform [12] from given synthetic mask, real images differs a lot. In two-dimensional electrophoresis gel images protein spots are often overlapped, have different intensity values and their intensity histogram can be quite narrow.

The results of segmentation of seven 2DE gel images by ACO are presented in Table 2. First six experiments (No. 1–6) were done with images having overlapped protein spots. Results of last – experiment No. 7 confirm that 2DE gel images without overlapped protein spots can be completely segmented by the use of ACO. The worst experimental result is in case of experiment No. 5, when protein spots where overlapping, image histogram was narrow and shifted to the brightest side.

Table 2. 2DE gel image segmentation results

Experiment no.	Nmb. of iterations	Segmented spots, %
1	1200	55.5
2	200	62.5
3	350	75.0
4	200	75.0
5	1000	44.4
6	500	50.0
7	**325**	**100.0**



(a) (b)

Fig. 5. Results of 2DE gel image fragment segmentation by ACO: a) pre-processed image; b) segmented image

Based on one of the best experiments with overlapping protein spots – experiment No. 4 – that gives 2DE gel image segmentation results presented in Fig. 5, let us examine population size dynamics. Because of ants' competition, death and reproduction, population graph presented in Fig. 6 has peaks and valleys.

First peak occurs when ants are meeting each other right after they have been placed on the image. Then number of ants decreases instantly as excess of ants is removed by the mask **B** that is applied every other iteration. First valley (at approx. 45 iteration) occurs because of the ants, which were not so close to high gradient level and died traveling without food. The second peak (at approx. 200 iteration) is due to reproduction of ants who reach the required gradient level, i.e., the protein spot. Here simulation can be stopped, because if continued, it can result in overtaking of one colony by another and lead to an over segmentation.

The visualization of segmentation procedure is presented on pheromone class map shown in Fig. 7. Here it can be clearly seen that after two iterations (Fig. 7 a) there are a lot of pheromone spots with different intensity each belonging to different ants' colony and pheromone placement picture is rather chaotic. On Fig. 7 b pheromone map is more ordered as ants are following gradient and reproduce. After 100 iterations (Fig. 7 c) pheromone map is ordered, as ant colonies reached spots and reproduced there. In a case of prolonged iterating ant colonies tend to take over neighbour spots and thus over segmentation occurs (Fig. 7 d).

Obtained final segmentation results from experiments No. 1–6 were compared with the threshold function segmentation method where threshold was selected individually for each image to segment as much spots as

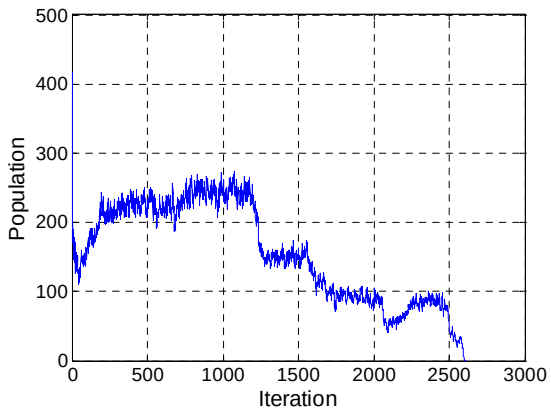
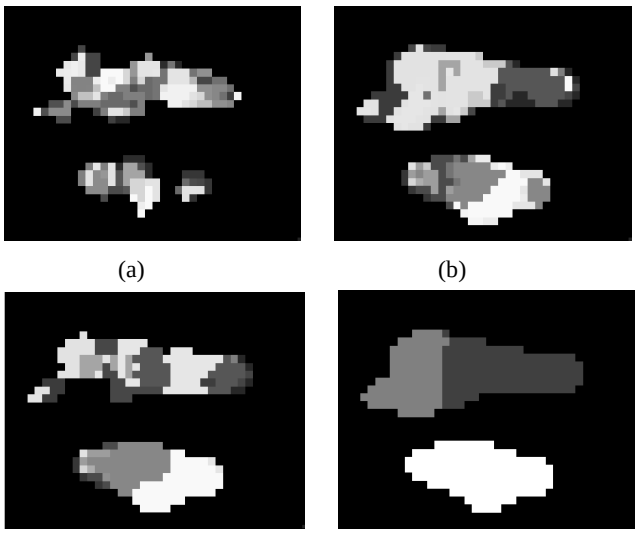


Fig. 6. ACO population change segmenting natural image



(a) (b) (c) (d)

Fig. 7. Dynamics of segmentation by ACO: a) iteration 2; b) iteration 25; c) iteration 100; d) iteration 2500

possible. ACO segmentation results were up to 67 % better than simple threshold function method on images with overlapped spots and different spots' intensity levels.

Use of the mask **B** not only improved segmentation, but also drastically decreased processing time. For 40×30 pixels image (see Fig. 5 a.) it took 108 s to reach 200 iterations with a mask and 408 s without mask. Total processing time (2600 iterations) took 1 440 s with mask and 5 400 s without mask. Processing time was measured on computer with Intel® Mobile 1.6 GHz processor, Centrino® platform.

Conclusions

The ability of Ant Colony Optimization technique for two-dimensional electrophoresis gel image segmentation was investigated:

1. Selected ACO model was adjusted for image segmentation and its parameter acceptable values where estimated using synthetic test image.

2. Image pre-processing using image mask was proposed that shortened processing time more than three times.

3. ACO model proved to be suitable for 2DE gel image overlapped protein spot segmentation with 66 % of correctly segmented spots.

2DE gel image segmentation by ACO results can be improved, e.g., trying to estimate ACO model parameters using 2DE gel images or performing more thorough image pre-processing. Possibilities, making some ACO parameters change dynamically in time or introducing other stopping criteria, i.e., based on colonies number ratio with population size, need further investigation.

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Ant colony optimization application for two-dimensional electrophoresis gel image segmentation is investigated. Standard ACO model with one ants' colony is presented also provided ants' colony size control method. Presented modified ACO model for two-dimensional electrophoresis gel image segmentation. Created synthetic image and experimentally estimated model parameters for segmentation are provided. Image pre-processing operations and processing results for decrease of processing time and increase segmentation accuracy are presented. Obtained results show that model is suitable for overlapped protein spots segmentation. Was reached average segmentation result that depends on number of overlapped spots and how close they centers are to each other. Model parameters and their influence on ant's behaviour and segmentation results are provided. Segmentation dynamics analyzed and intermediate segmentation results are provided. Population changes analyzed and population size change is proposed as a stopping parameter for segmentation process. Segmentation accuracy was about 67% improve over simple threshold function segmentation. Processing times are commented. Segmentation result can be improved by the use of real electrophoresis images for parameters estimation, also by the use of more effective optimal model parameters estimation algorithm. Ill. 7, tabl. 2, bibl. 12. (in English; summaries in English, Russian and Lithuanian).

Р. Лаптик, Д. Навакаускас. Применение оптимизации муравьинной колонией для сегментации изображений // Электроника и электротехника. – Каунас: Технология, 2007. – № 8(80). – С. 13–18.

Исследуется применимость оптимизации муравьинной колонией для сегментирования двухмерных изображений электрофореза белка. Представлена стандартная модель оптимизации муравьинной колонией и метод контроля популяции. Предложены модификации к стандартной модели для сегментирования изображений электрофореза белка. Создано искусственное изображение и с помощью экспериментов определены параметры модифицированной модели. Представлена очередность операций предварительной обработки изображения для сокращения времени обработки и улучшения качества сегментирования. Полученные результаты показывают пригодность модели для сегментирования слившихся пятен. Был получен средний результат сегментирования, зависящий от количества слившихся пятен и расстояния их центров друг от друга. Описаны параметры модели и их влияние на поведение муравьев и сегментирование изображения. Проанализирована динамика сегментирования и представлены предварительные результаты. Проанализировано изменение популяции, и как условие остановки сегментирования, предложено изменение ее размера. При сравнении с простой пороговой функцией результат сегментирования оказался улучшен на 67 %. Также было представлено время обработки изображения. Результат может быть улучшен, используя реальные изображения электрофореза белка при определении параметров модели, а также применяя более эффективный метод для определения оптимальных параметров модели. Ил. 7, таб. 2, библ. 12 (на английском языке; рефераты на английском, русском и литовском яз.).

R. Laptik, D. Navakauskas. Optimizavimo skruzdėlių kolonijomis taikymas vaizdams segmentuoti // Elektronika ir elektrotechnika. – Kaunas: Technologija, 2007. – Nr. 8(80). – P. 13–18.

Tiriamas optimizavimo skruzdėlių kolonijomis taikymas dvimatės elektroforezės gelių vaizdams segmentuoti. Pateiktas standartinis optimizavimo su viena skruzdėlių kolonija modelis bei skruzdėlių kolonijos populiacijos kontrolės metodas. Pasiūlytos vaizdams segmentuoti tinkamos standartinio modelio modifikacijos. Sukurtas dirbtinis vaizdas ir eksperimentiškai nustatyti modifikuoti skruzdėlių kolonijos modelio parametrai, tinkami dvimatės elektroforezės vaizdams segmentuoti. Pateiktas pirminio vaizdo apdorojimo operacijų nuoseklumas ir apdorojimo pavyzdžių, siekiant sumažinti segmentavimo trukmę ir pagerinti jo tikslumą. Taikant pasiūlytą modelį gauti eksperimentų rezultatai, kurie rodo modelio tinkamumą susiliejusių dvimatės elektroforezės dėmių vaizdams segmentuoti. Gautas segmentavimo rezultatas priklausė nuo susiliejusių dėmių skaičiaus ir jų sanklotos. Atskirai aptarti modelio parametrai ir jų įtaka skruzdėlių elgesiui bei segmentavimo rezultatams. Aptarta segmentavimo dinamika ir pateikti tarpiniai segmentavimo vaizdai. Išanalizuoti populiacijos pokyčiai. Populiacijos dydžio svyravimas pasiūlytas kaip tinkamas kriterijus segmentavimui stabdyti. Gauti 67 % geresni segmentavimo rezultatai nei taikant slenksčio funkciją. Segmentavimo rezultatai gali būti pagerinti, parenkant parametrus pagal realius vaizdus ir naudojant efektyvesnį optimalių parametrų parinkimo algoritmą. Il. 7, lent. 2, bibl. 12. (anglų kalba; santraukos anglų, rusų ir lietuvių k.).