**Szereted csinálni azt amit csinálsz, és élvezed a kutató munkát!!!! ☺**

**Célkitűzések:**

* Mikrofluidikai rendszerek, szűrések bemutatása (általános + parazita + eszközök ismertetése)
* Irodalomkutatás bemutatása
* Algoritmusláncolat létrehozása:
  1. Sutaration algoritmus (mi az alapja, miben különbözik a többi detekciós eljárástól)
  2. Canny edge detector: (élkeresésről általánosan is írni + Cannyt bemutatni)
     + Sima + skálázott összefűzése (összehasonlítások is persze)
  3. Színinformációs eljárás: (általános bevezetés, hogy mi az alapja, miben más, mint a többi)
     + Lokális színkülönbségeken alapuló élkeresés
  4. Alapszerkezet kiszűrése a képről: (hol szokták használni, miért hasznos [általánosan])
     + Adaptive thresholdolások
     + SIFT alkalmazása
* Matlab program bemutatása

Abstract (2 page)

Contents

* Big numbers for chapters, X.Y.Z number format for subchapters (where X,Y,Z nature numbers)

List of Figures

List of Tables

List of the Used Abbreviations

From here, you count the pages! (30 and 50k character)

Abstract (In english and hungarian too)

Introduction

Main theme, work

Conclusion

Until this point!

References

Appendices 🡪 here you can write the code 🡪 enough to give it on CD

Acknowledgements

**Image processing algorithms in microfluidic systems**

**Abstract** (0.5 page) – akit nem érdekel az is tudja miről szól

The main goal of the thesis is to collect and merge different kind of algorithms giving an effective program for *Trichinella spiralis* detection and counting its based on videos about biomicrofluidic device measurements. Biomicrofluidic device research projects are a part of ‘Lab on a chip’ program, in that the researches and engineers strive to design a small piece of device for some – nowadays one or two – specific detection or segmentation (van-e más felhasználási területe?) problem of biologic samples. One of the leading project in our laboratory is *Trichinella spiralis* detection procedure. *Trichinella spiralis* is a dangerous parasite for animals as well as humans, which most dangerous source for humans is the pork. Avoiding the sales of infected meat products is really important the continuous and reliable controlling at the slaughterhouses, which nine times out of ten does not have a complete and equipped laboratory. For these purposes, our research team tries to give an alternative detection mode, which requires less laboratory instruments, only a microfluidic device, a syringe pump, some plastic tubes, a light microscope with a suitable camera, and a user-friendly detector program, which alerts the controlling person when some pathogen in the sample is. My task was to research the further possible developments of a latter implemented algorithm by Péter Zsíros and combine its results with some other detection methods taking the algorithm more robustness and effectiveness for real-life application. In the interest of reaching these aims, I worked with Canny edge detector, an adaptive thresholding method, a hole-filler, and a new . After determining the meaningful objects, the program measures all of its extents, and divides this number with the average size of the parasites, what was calculated earlier from the training set videos. The method is called as “Divided approach”.

**Absztrakt** (magyarul)

**Introduction** (1 + ¼ page)

**Literature**

1. Microfluidic (**intro**, **history**, its mechanic + some function, manufacture methods, laboratory applications + our specific device’s functionphysic + useless + outlook)
   1. Microfluidic systems are those kind of systems, what actually do not need a huge volume from the sample just some microliter. Generally, it has a specific structure, which is designed for its function, and thus one device usually fits only for one or two problem. More exactly it is “The science and engineering of systems in which fluid behaviour differs from conventional flow theory primarily due to small length scale of the system.” [1]. These systems functions could be varied ranging from detection to segmentation onto executing some complex biochemistry technique for example Loop-mediated isothermal amplification – LAMP – or Quantitative Polymerase Chain Reaction – qPCR. Engineers, who works on this field has to study a lot of and various kind of subjects as nanophysics, mathematics, biochemistry and biotechnology, because microfluidic bases on these basic disciplines. Whereas nanophysics and biotechnology just in the last few decades has been growing up as high level as microfluidic needs, this is an absolutely new field of the science. The first devices were made in the 1980s, and through the mid-1990 years the developments of these apparatuses including the microvalves, micropumps and microflow sensors actively continued. At this time, the engineers discovered that if they would like to minimize the size, they will need to use external actuators for microvalves and micropumps. This was came from two basic statement: first is that, if we scale down the size, this will indicate the power decreasing of the device by a length scale cubed, so we don’t anticipate as high level activity from microvalves and micropumps as conventional devices have. The second one is the fact, that “The surface-to-volume ratio varies as the inverse of the length scale” [1]. It means that, a large surface has large viscous forces, and often the integrated microactuators are not able to give enough power to micropumps, which should move the fluid in microfluidic systems. First devices manufactured from silicon, but nowadays, remaining this direction for example Polydimethylsiloxane – PDMS, the industry are focusing some more advanced materials as different type of plastics. Nonetheless, the most important physical parameters have not changed since the beginnings, which are the followings:
   * Small volume [μL, nL, pL, fL]
   * Small size [μm, mm]
   * Low volumetric flow rate [μL/s, μL/h, ml/s, ml/h]
   * …

Principles of function and behaviour (gen.) 🡪 Library **REF**

As microfluidic channels are in micro- and nanoscale range, the physic in this dimension varies from that Newton physic, what we know very well. The usual gravity force and the buoyancy has less influence to the flowing liquid in the systems, in turn the surface forces like surface tension or van der Walls force between particles are more important. Besides this, because we work with fluids, the viscous force is also important, moreover in laminar flowing this is the dominating force. [2] We can describe these flows whit the Reynolds number, which had been declared in 1883 by Osborne Reynolds as the ratio of inertial forces to viscous forces. It is defined as “[3]”

If Re is much less 2000, viscous forces dominate the flow, so it is laminar, but on the other hand if this number is higher than 2000, the flow turns to be more turbulent. In our laboratory every devices have a low Reynolds number, so in all of these the flow is laminar.

Manufacture technologies (silicon waffer + PDMS device) 🡪 Misi

Fabricate a microfluidic chip is not as difficult thing as for first hearing it seems to. First of all, we need to order a silicon wafer, which contains our previously designed device structure.

Így készültek a mi eszközeink is.

Our device description (function goal + principle) 🡪 Misi

Outlook (what other devices in a lab too, developments) 🡪 Sci-hub **REF**

1. Sample introduction (trichinella spiralis + preparation)
   1. *Trichinella spiralis* 🡪 Net + library **REF**
   2. Preparation 🡪 Ádám
2. Describe the measurement (main steps + equipment, evaluation earlier + researches + connection to image processing)
   1. Main steps + equipment 🡪 Ádám + net **REF**
3. Microfluidic + image processing (hypothetical + real applications generally + in laboratory – Peti)
   1. Hypothetical + real applications generally 🡪Sci-hub **REF**
   2. In laboratory 🡪 Peti **REF**
   3. Evaluation earlier + researches 🡪 Ádám + Alexandra + own thoughts

**Algorithm:**

1. Microscopic setting probe measurements with uEye camera

Improving the incoming images for Saturation algorithm

* 1. The best recording equipment is not enough for taking good quality images. We have to know very well the software and hardware too, what we use under recording, that means we need to understand the most important features and setting parameters. My research in the theme began at this point. To understand, how does the above-mentioned uEye Cockpit software and microscope work together, I had to take some test measurements with its. Based on my tests the following parameters were the most interesting:
  2. Microscope
  + Aperture levels
  + Light intensity levels
  1. Software
  + Auto-contrast on/off mode
  + Auto-white balance on/off mode

Our plan with these measurements was to improve the results of a previously implemented parasite detection algorithm – the so-called Saturation algorithm, which have been made by Péter Zsíros [6]. The algorithm was super and functioned well, but it was not enough for us. We would have like to develop it to reach higher efficiency in parasite detection than it have been until that time. In order to achieve our goals, I had to make a uniformly illuminated image composition by modifying the light intensity level and the aperture level at the same time. Auto contrast and auto white balance switches could have been turned on/off also for achieving that hypothetical light intensity equilibrium state.

1. Saturation algorithm – detection algorithm 1.
   1. As I mentioned above, the algorithm is not my intellectual product. I just worked with it and tried to find a solution for its weaknesses. The main idea of the algorithm is to use the distinct saturation level of the parasites from other artefacts on the image. First step of it is the noise reduction, especially Salt&Pepper noise reduction. The input image is filtered with a 5x5 median filer on every channel of it eliminating this kind of noise. After converting the smoothed image from RGB to HSV, the saturation level was taken out and converted to grayscale anticipating the following operation, the Difference of Gaussian. DOG uses two distinct Gaussian filter and thereafter subtracting/substracts one of them from the other convolves the previous image with this itself.
2. Canny edge detector – detection algorithm 2. + modifications

Canny edge detector is one of the most known object detector in image processing. It had been developed by John F. Canny in 1986 to satisfy the following requirements and make a better a solution than previous edge detection methods:

* Catch as many real edges as possible
* Do not detect false edges, which might arise from image noise
* Detected edges be as close to real edges as possible
* The detection be separate from edge direction - isotropic

Four different part builds it up, but via linking these parts it could get pretty good outcome for well-defined images. In addition of its attribution, it is not too costly and easy to implement. The first two advantages were the reason why I turned to this and build it to my code/algoritmus láncolat, because of our images is made by constant lighting with fixed static camera, and in the near future I would like to extend it to lower teljesítményű architectures. The four main step of the algorithm are the followings:

1. Noise reduction

The incoming image is convolved with a Gaussian kernel reducing the noise of the image, which may come from different sources. e.g.:

1. Gradient intensity and direction calculation

The algorithm calculates the gradient intensity and its direction to emphasize the edges on the input image. KÉPLET

1. Non-maximum suppression

All edges are categorized into for different group – 0o, 45o, 90o, 135o – based on their directions. The selection method puts every edge to the appropriate numbered group, if its direction is between the two boundaries of the group e.g: 67o goes to the 2 group, because it value is more than 45o, but less than 90o. The groups does not finish at 180o, but goes further continuously for second half of the circle until 0o by corresponding way with the same differentiations between the limits. [kör számos kép]. The goal of the categorization is to supress the edges forming the skeleton of the objects. Namely, if the magnitude of an edge is not greater than magnitude of its two neighbours of the gradient direction, its value will set to zero. [9 pontos kép]

1. Hysteresis thresholding

In this step the algorithm solves the “one threshold” problem, which claims that if the threshold level is too low so many invalid edges will appear, however if its level is too high, true edges will disappear. This two threshold system is working by the following way:  
[képletek az önlabból]

1. + modifications

Adaptive thresholding

Eliminate small objects

Eliminate basic structure from background

Correction of luminesce changes

Back loading

1. Hole filler
2. Counting – how, why does Peti and I do it
3. Training set – how do I select, collect images
   1. Categories
4. GUI – introduction, utilities, benefits

**Evaluation:**

1. Microscopic setting probe measurements with uEye camera
   1. Improving the incoming images (only the results)
2. Saturation algorithm – detection algorithm 1.
3. Canny edge detector – detection algorithm 2. + modifications
   1. Adaptive thresholding
      1. Cleaning small objects from image
      2. Base removal
      3. Luminosity robustness (rea)
   2. Back loading
4. Counting – evaluate the results, explanation + future outlook (development)
5. Training set
   1. How do I select, collect images 🡪 ennek sikeressége
6. Comparison the 2 algorithm
   1. Based on results
7. GUI – introduction, utilities, benefits (based on 50 person’s feedback)

**Results:**

**References:**

[1] Fundamentals and application of microfluidics, zöld-vörös borítós

[2] Fainman article

[3] Reynold number: <https://en.wikipedia.org/wiki/Reynolds_number#History>

[4] Parazitás képre 🡪 D. Van den Eden, "Illustrated Lecture Notes on Tropical Medicine - Tissue nematodes", *Itg.author-e.eu*, 2013. [Online]. Available: http://itg.author-e.eu/Generated/pubx/173/helminthiasis/tissue\_nematodes.htm. [Accessed: Apr- 2013].

[5] Peti Bsc-s szakdogája

[6] Zsíros Péter Attila, „Optikai úton történő részecskedetektálás mikrofluidikai eszközökben”, Diplomaterv, ITK, PPKE, Budapest, Magyarország, 2016

[7] Dániel Szolgay. Class Lecture, Topic: “2D Convolution” Neumann Lecture Hall, Faculty of Information Technology and Bionics, Pázmány Péter Catholic University, Budapest, Sept. 20, 2016.

**Piszkozat:**

**Abstract:**

* This program is implemented by me, but one part of it is developed by a graduated student, Péter Zsíros.
* The detector program could also help the researches of the Biomicrofluidic laboratory to improve the devices. was built up from different type of shape or edge detection algorithms, which gives a good and plausible solution enhanced each other benefit parts and reduced their disadvantages.
* could be substituted simply by a parasite filtering microfluidic device, a light microscope with a, and an attached user friendly detector program, what is able to detect the parasite.
* This program could help the development of biomicrofluidic device, and in the future will may attach to device as a friendly user program. The plan of device + program combination is to detect/manifest these pathogen parasites more rapid and simply way in the slaughterhouses avoiding to market some infected pork.
* The detection and counting helps device development and could be improved
* Merging algorithms allows us to configure a

**Introduction:**