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# Detection of follicles in ultrasound videos of bovine ovaries

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**Abstract.** Ultrasound imaging is a veterinarian standard procedure for the monitoring of ovarian structures in cattle. Recent studies, suggest that the number of antral follicles can give a cue of the future fertility of a specimen. Therefore, there has been a growing interest in counting the number of antral follicles at early stages in life.

In the most typical procedure, the operator performs a trans-rectal ultrasound scan and counts the follicles on the live video that is seen in the ultrasound machine. This is a challenging task and requires highly trained experts that can reliably detect and count the follicles in a quick sweep of a few seconds.

This work presents the integration of several signal processing techniques to the problem of automatically detecting follicles in ultrasound videos of bovine cattle ovaries. The approach starts from an ultrasound video that traverses the ovary from end to end. Putative follicle regions are detected on each frame with a cascade of boosted classifiers. In order to impose temporal coherence, the detections are tracked across the frames with multiple Kalman filters. The tracks are analyzed to separate follicle detections from other false detections.

The method is tested on a phantom dataset of ovaries in gelatin with dissection ground truth. Results are promising and encourage further extension to in-vivo ultrasound videos.

 ${\bf Keywords:} \ {\rm follicle} \ {\rm detection}, \ {\rm cascade} \ {\rm classifier}, \ {\rm multitracking}$ 

# 1 Introduction

Recently, there has been an increasing interest in studies concerning antral follicle count (AFC) and its influence on the reproductive performance in cattle, as well as its applications in reproductive biotechnologies [6, 10]. AFC is highly variable in different species, but in cattle, there is a high repeatability in the same individual, regardless of race, age, breeding season, lactation or pregnancy conditions [1]. Also, AFC is consistent throughout the estrous cycle of individual cows; therefore, a single routine ultrasound examination is enough to identify females with low, intermediate or high AFC. More interestingly, it has been reported that cows with a lower AFC have lower fertility [4, 9]. In order to manually count the antral follicles, veterinarians perform a transrectal ultrasound (TRUS) where they first locate the ovarian region and then scan the ovary while rotating the probe. The procedure must be done in a few seconds and requires highly trained experts in order to reliably detect and count the follicles. Results are accurate when the ovary has few and big follicles, but accuracy drops when the ovary has a large number of follicles and/or follicles are small in size.

Automatically counting the number of antral follicles in the ovary remains an open challenge for state of the art methods [11, 5]. Borders of small follicles are weak and irregular due to the intrinsic characteristics of ultrasound (US) images. The presence of follicle-like structures, such as vessels, makes the problem even harder. However, counting the number of follicles has the advantage that no precise segmentation of the follicles is required. The problem has been addressed before in different ways with 2D and 3D US data but no conclusive results have been achieved in the academy or commercial developments.

A method for fully automated ovary and follicle detection in 3D ultrasound is presented in [2]. The approach proposes a probabilistic framework to estimate the size and location of each individual ovarian follicle by fusing the information from both global and local context.

The commercial product SonoAVC software which is integrated into the Voluson E8 ultrasound machine (GE Medical Systems) performs semiautomatic follicle segmentation and volume measuring on 3D ultrasound data [16].

Methods that detect follicles in 3D ultrasound are probably the most successful ones. Additional information present in 3D ultrasound can be effectively used to discriminate between follicles and other common falsely detected structures [11]. However, veterinarians of our research group, in agreement with observations made in [11], prefer 2D ultrasound scans because they are easier and faster to perform. In addition, 3D US machines are still too expensive for veterinarians in underdeveloped countries. For this reason, our goal is to investigate if it is possible to count the number of follicles using 2D ultrasound videos.

Regarding the detection of follicles in single US images, a research group introduced several fully automated approaches to ovarian follicle detection in single US images based on region-growing [12, 13]. Another group proposed several variants based on a feature extraction and classification scheme [5].

To count all the follicles of the ovary, more than one image is required. One of the possibilities is to segment the images in a frame by frame basis and then group all the detections corresponding to a single follicle as a unique detection. This kind of approach with temporal sequences is presented in [14] and improved in [15] where detected boundaries of the follicles are tracked using a combination of three mutually dependent Kalman filters.

Unfortunatelly, the lack of publicly-accessible datasets of follicles on ovarian ultrasound images (2D or 3D) prevents from an objective comparison between different methods.

The rest of this paper is organized as follows. Next section introduces the datasets used for developing and evaluating the system. Section 3 presents the

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signal processing techniques applied to the ultrasound videos in order to detect the follicles. Experiments and results are presented in sections 4. Finally, the paper ends with some concluding remarks.

# 2 Dataset

With the purpose of developing and evaluating the approach, two phantoms were prepared with nine ovaries each immersed in gelatin. The ovaries were collected from the slaughterhouse and conditioned removing tissue debris. Each phantom was built by placing the ovaries at an approximate depth of 1 cm in a box filled with gelatin.

For each ovary three acquisitions with a TRUS probe were performed by an expert. The acquisitions were made by rotating the probe about its axis. The videos were acquired at 30 frames per second and 640x480 format with the following criteria: (a) scan from right to left from end to end of the ovary, (b) scan from left to right from end to end of the ovary, (c) scan back and forth from end to end of the ovary while the expert performs a count of the follicles.

Following the US scans, the phantoms were disassembled and the ovaries dissected. For each ovary, all the follicles and corpora lutea were measured. Follicle size ranges from 2 to 20 mm.

Figure 1 shows one of the phantoms and the US acquisition procedure.



(a) One of the phantoms.



(b) Ultrasound scanning of the phantom

Fig. 1: Phantom with ovaries immersed in gelatin.

## 3 Signal processing for follicle detection

## 3.1 Follicle-like regions detection in each frame

The first step in the proposed system is to automatically detect in each frame of the video the regions that are likely to be a follicle. Follicles are roughly spherical structures with hard walls and filled with liquid. Echogenicity is high in the walls while the internal fluid is almost anechoic. This gives in US a typical circular pattern brighter in the borders and darker in the middle. A cascade of boosted classifiers [17] based on local binary pattern features is a good and fast alternative to detect this kind of structure. The classifier was trained with a set of follicle regions and negative samples (samples were scaled to 24x24 pixels or equivalently 2.4x2.4 mm for the spatial resolution of the videos). Figure 2a shows an example of follicle like detected regions in a frame.



(a) Example of putative follicle regions detected with the cascade classifier in one of the frames of the video.

(b) Detected tracks in red. The blue dots are the centers of the regions detected with the cascade classifier.

Fig. 2: Follicle-like regions are detected with a cascade classifier. In order to impose temporal coherence, the detections are tracked with multiple Kalman filters.

## 3.2 Temporal coherence by multiple tracking

The main movement of the probe during an US scan of an ovary is a rotation but there may also be translations. These translations may be involuntary (e.g. trembling of the operator or movement of the animal during the procedure) or due to a necessary panning if the ovary is wider than the size of the US probe. In any case, if these movements are not too brusque, each follicle is normally detected during several frames with the 2D positions of the detections describing a soft movement along the frames. The objective of the tracking step is to group temporal coherent detections into tracks where each track is presumably related to a single follicle.

In order to impose temporal coherence, the detections are tracked with multiple Kalman filters [7] across the video frames with independent constant acceleration models. Detections are assigned to tracks in each frame based on the Hungarian Algorithm [8] using as cost matrix the distance between predicted positions of each track and actual detections (a maximum distance of 1mm is tolerated in the current implementation). When a detection cannot be assigned to any active track, a new track is created.

Figure 2b shows an example of detected tracks.

#### 3.3 Identification of the ovarian region

In the phantoms, the ovary is surrounded by gelatin and that may ease the identification of the ovarian region. In vivo, the ovary is surrounded by other tissues and the boundary of the ovary is usually not easily discernible. Moreover, the surrounding tissues may lead to spurious detections showing follicle-like regions. According to expert veterinarians, the boundary of the ovary is usually hard to identify and the ovarian region is recognized by the grouping of follicles that can be more easily identified in the US video. With this in mind, the approach in this work is to identify the ovarian region as the most important cluster of detected tracks. To cluster the tracks, a probability map is constructed by convolving the track position (weighted by track length) with a Gaussian kernel. The ovary region is detected as the main mode of the probability map using the Mean Shift algorithm [3] considering a typical ovary size. Figure 3 shows the identification of the region of an ovary.



(a) Scatter plot of the xy positions of (b) Probability map based on the dethe tracks weighted by track length on tected tracks. sample frame.

Fig. 3: Ovary identification as the main cluster of detected tracks. The white rectangle depicts the main mode.

#### 3.4 Follicle identification and measurement

To identify a detected track as a follicle, the track must be active for at least a minimum number of frames. This can allow to differentiate a follicle that is consistently detected across several frames from tracks originated by short term spurious detections of the cascade classifier. With this approach, it is necessary to determine as operating point the best threshold for the minimum number of frames. In this work, the threshold is selected as the one to give the best results in terms of mean square error against the ground truth given by dissection.

Upon detection, the diameter of a follicle can be estimated as the largest detection in the track which corresponds to the frame when the US plane cuts the follicle (roughly a sphere) in its great circle.

## 4 Experiments and Results

The phantom dataset was used to evaluate the approach. As mentioned in section 2, in the phantom dataset there are two US scans (right to left and left to right) where each ovary is scanned from end to end. In the experiments, a 50 times 6-fold cross validation was performed on the right-to-left sweep to determine the operating point (the threshold for the minimum number of frames to consider a track as a follicle detection). Figure 4 presents the results for the cross validation.

The computed operating point concentrates around a minimum track length of 17 frames (figure 4b). Since the videos have 30 frames per second, this means that the tracks must be active for more than half a second to be considered a follicle. Around the operating point, results against dissection can be considered comparable to the expert with a lower correlation factor but better centering (figure 4 a,c). It is known to experts that the accuracy of follicle count drops when the number of follicles in the ovary is high (the experts tend to underestimate the count). The results on this dataset are consistent with this issue . The expert outperforms the automatic counting in the ovaries with few follicles (eg. less that 18 in this dataset). For the ovaries with more follicles, the automatic approach also underestimates the counts but is in general closer to the ground truth than the expert count.

## 5 Concluding remarks

An automatic algorithm for the problem of detecting follicles in ovarian US videos of bovine cattle ovaries is presented. The proposed approach can work directly on the 2D US videos generated in the typical procedures done by the veterinarians with the only restriction that the US scan must be done in a single sweep from end to end of the ovary. The lightweight processing enables to have the results immediately after the US scan. The method can give also the size of the detected follicles which is an information usually disregarded in the routine follicle count procedure.

Although the phantoms constitute a small dataset, results can be considered promising with count results comparable to an expert in a controlled environment. This encourages future work to robustly extend the approach to in-vivo scans. Future work should include for example the evaluation of other strategies to identify the true follicles from the detections, as well as alternative forms to





(a) Results of the cross validation on the right-to-left scan

(b) Histogram of the operating points determined by the cross validation

	Expert	Right-to-left scan					Left-to-right scan					
		15	16	17	18	19	15	16	17	18	19	
Pearson correlation	0.90	0.82	0.83	0.85	0.85	0.86	0.81	0.78	0.81	0.84	0.85	
Max. overestimation	2	11	8	6	5	3	9	8	8	5	3	
Min. underestimation	-11	-5	-5	-6	-8	-11	-8	-9	-9	-9	-9	
Average of differences	-3.33	2.22	0.78	-0.56	-1.33	-2.78	1.22	-0.33	-1.50	-2.56	-3.44	
Median of differences	-2	1.50	0.50	0.00	-0.50	-3.00	1.00	-0.50	-1.50	-2.50	-2.50	
Differences std	3.93	4.37	4.18	3.97	3.93	4.05	4.44	4.70	4.37	4.10	4.10	

Count results around the operating point (minimum length of track between 15 and 19 frames)

(c) Results for the expert and around the computed operating point for the right-to-left and left-to-right scans

Fig. 4: Results for the 50 times 6-fold cross validation on the right-to-left scan.

identify the ovarian region on the video. Also, a method to detect the corpora lutea in the ovary may help to differentiate cavities from real follicles.

# References

- David S Burns, Fermin Jimenez-Krassel, Janet LH Ireland, Phil G Knight, and James J Ireland. Numbers of antral follicles during follicular waves in cattle: evidence for high variation among animals, very high repeatability in individuals, and an inverse association with serum follicle-stimulating hormone concentrations. *Biology of Reproduction*, 73(1):54–62, 2005.
- Terrence Chen, Wei Zhang, Sara Good, Kevin S Zhou, and Dorin Comaniciu. Automatic ovarian follicle quantification from 3d ultrasound data using global/local context with database guided segmentation. In *Computer Vision, 2009 IEEE 12th International Conference on*, pages 795–802. IEEE, 2009.
- Yizong Cheng. Mean shift, mode seeking, and clustering. Pattern Analysis and Machine Intelligence, IEEE Transactions on, 17(8):790–799, 1995.
- 4. RA Cushman, MF Allan, LA Kuehn, WM Snelling, Andrea S Cupp, and HC Freetly. Evaluation of antral follicle count and ovarian morphology in crossbred

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beef cows: Investigation of influence of stage of the estrous cycle, age, and birth weight. *Journal of animal science*, 87(6):1971–1980, 2009.

- PS Hiremath and Jyothi R Tegnoor. Follicle detection and ovarian classification in digital ultrasound images of ovaries. Advancements and Breakthroughs in Ultrasound Imaging, InTechOpen, UK, pages 167–199, 2013.
- 6. JJ Ireland, GW Smith, D Scheetz, F Jimenez-Krassel, JK Folger, JLH Ireland, F Mossa, P Lonergan, and ACO Evans. Does size matter in females? an overview of the impact of the high variation in the ovarian reserve on ovarian function and fertility, utility of anti-müllerian hormone as a diagnostic marker for fertility and causes of variation in the ovarian reserve in cattle. *Reproduction, Fertility and Development*, 23(1):1–14, 2010.
- Rudolph Emil Kalman. A new approach to linear filtering and prediction problems. Journal of basic Engineering, 82(1):35–45, 1960.
- 8. Harold W Kuhn. The hungarian method for the assignment problem. Naval research logistics quarterly, 2(1-2):83–97, 1955.
- Marcelo F Martinez, Neil Sanderson, Laurel D Quirke, Stephen B Lawrence, and Jennifer L Juengel. Association between antral follicle count and reproductive measures in new zealand lactating dairy cows maintained in a pasture-based production system. *Theriogenology*, 85(3):466–475, 2016.
- F Morotti, TRR Barreiros, FZ Machado, SM González, LSR Marinho, and MM Seneda. Is the number of antral follicles an interesting selection criterium for fertility in cattle? *Animal Reproduction*, 12(3):479–486, 2015.
- Božidar Potočnik, Boris Cigale, and Damjan Zazula. Computerized detection and recognition of follicles in ovarian ultrasound images: a review. *Medical & biological* engineering & computing, 50(12):1201–1212, 2012.
- Božidar Potočnik and Damjan Zazula. Automated ovarian follicle segmentation using region growing. In *Image and Signal Processing and Analysis, 2000. IWISPA* 2000. Proceedings of the First International Workshop on, pages 157–162. IEEE, 2000.
- Božidar Potočnik and Damjan Zazula. Automated analysis of a sequence of ovarian ultrasound images. part i: segmentation of single 2d images. *Image and Vision* Computing, 20(3):217–225, 2002.
- Božidar Potočnik and Damjan Zazula. Automated analysis of a sequence of ovarian ultrasound images. part ii: prediction-based object recognition from a sequence of images. *Image and Vision Computing*, 20(3):227–235, 2002.
- Božidar Potočnik and Damjan Zazula. Improved prediction-based ovarian follicle detection from a sequence of ultrasound images. *Computer methods and programs* in biomedicine, 70(3):199–213, 2003.
- Nick Raine-Fenning, Kannamannadiar Jayaprakasan, J Clewes, I Joergner, S Dehghani Bonaki, S Chamberlain, L Devlin, H Priddle, and I Johnson. Sonoavc: a novel method of automatic volume calculation. Ultrasound in Obstetrics & Gynecology, 31(6):691–696, 2008.
- Paul Viola and Michael Jones. Rapid object detection using a boosted cascade of simple features. In Computer Vision and Pattern Recognition, 2001. CVPR 2001. Proceedings of the 2001 IEEE Computer Society Conference on, volume 1, pages I-511. IEEE, 2001.