Smartphone-Obtained Images to Extrapolate Canine Packed Cell volume with Enhancement Techniques

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Abstract—**Clinical diagnosis requiring central facilities and site visits can be burdensome for patients in resource-limited or rural areas. Therefore, development of a lowcost test that utilizes smartphone data collection and transmission would beneficially enable disease self-management and point-of-care (POC) diagnosis. Smartphone-obtained image can be used to extrapolate the PCV of canine blood samples. Blood samples were placed on filter paper, and images were obtained with a smartphone in different environments. These results were then compared to the PCV measurements on the same samples following the World Health Organization guidelines. Many samples were assessed and identified that smartphone images, obtained in a controlled environment, were able to predict the PCV of the samples. This prediction was most accurate when assessing samples of a normal erythroid mass or polycythemic sample.**

The results of this study suggest that smartphone-obtained images do have a utility in predicting canine PCV with Image Enhancement Techniques. If this can be incorporated into a smartphone application, there would be scope to use this in low resource settings.

Index Terms—Anemia, Diagnostics, Hematocrit Estimation, PVC, Smartphone Camera, Canine Blood Samples

I. INTRODUCTION

Packed cell volume (PCV) measurement is a routinely performed diagnostic test in both primary care and referral settings and is often included as part of a minimum database when evaluating patients at initial presentation. [1] This provides an approximation of the patient's circulating erythroid mass to be readily assessed, allowing pertinent clinical decisions to be made in a short space of time. [2] Packed cell volume measurements has multiple benefits in addition to its relative rapid availability, with a result available within 5–10 minutes. Only a small volume of blood is needed, which can be taken from EDTA or lithium heparin treated blood [3]. In the emergency setting or in small patients, with neonates being the classic example, blood can also be obtained directly from an IV cannula in micro hematocrit tubes pretreated with anticoagulant. The small volume facilitates repeat sampling, allowing trends to be tracked and comparisons to be made. An additional benefit is that, after the initial outlay on equipment, it is inexpensive to perform each test, making it an affordable test that can yield a large amount of information with relatively limited input.

Packed cell volume measurement is not without its limitations. One of the most notable is operator-dependent variation. Two previous studies have identified that the mean degree of variation is 0.09 L/L [9%], when taking into account differing operator experience in veterinary medicine [4-5]. The latter study identified that this variation could be ameliorated by the introduction of a standard operating procedure. This is not in widespread use at present, and as such this variation is likely still present in a proportion of the samples being analyzed. This variation could result in inappropriate interventions or, conversely, a failure to take action when necessary.

An additional failing of this test is that it is not portable due to the equipment involved. This limits its utilization in certain settings, such as ambulatory equine or farm animal work in the veterinary field, or low resource settings in the human field. There are points of care hematocrit readers, although cost and additional equipment needed may detract from their use.

A more pertinent issue in human medicine, although still relevant in veterinary medicine, is the risk from biological material. In particular, sharp-related injuries secondary to handling of glass micro hematocrit tubes and blood aerosolization, in the case of sealant failure during centrifugation, are of potential risk to operators.

Smartphone use is becoming increasingly widespread in the medical field and has been used to augment or improve upon current diagnostic techniques. There are previous reports of smartphone-obtained images being utilized in forensic medicine to age blood spatter. [6] Additionally, there are also reports of the use of add-on equipment, which allows hemoglobin concentration to be calculated. [7-8]

The aim of this study was to assess whether images obtained with a smartphone and use Enhancement techniques reduce its noise and then widely available equipment could be used to extrapolate the PCV. A secondary aim was to establish whether a controlled environment and standardized blood volume were necessary to improve the accuracy of the results. The hypothesis was that images introduces obtained in a controlled environment with a standardized volume of blood spot would correlate best with them anual PCV.

II. PCV TEST

Despite the fact that a packed cell volume is measured dozens of times a day at the Animal Medical Center, most pet owners have never heard of packed cell volume, sometimes referred to as a hematocrit. If one of your pets has experienced a serious issue with anemia, then you might have heard your veterinarian talk about this test. Also known as PCV, packed cell volume is

one measure of the number of red blood cells in the blood. There are other methods to assess the number of red blood cells, but these take more time and much more sophisticated laboratory equipment. The laboratory can count the number of red blood cells; there are millions in a drop of blood. The oxygen-carrying protein hemoglobin contained inside of red blood cells can also be measured; like red blood cells, hemoglobin decreases when a patient is anemic.

Blood, in general, is a mix of plasma as well as cells. The PCV test measures how much of the blood consists of cells. If the PCV returns a reading of 50%, it means that 50 ml of the cells are present in exactly 100 ml of blood. If the RBC number increases, then the total reading of the PCV is also up. This number can also increase due to dehydration.

Performing the PCV tests and the total solids is a pretty routine and simple test undertaken at many hospitals. All medical members can easily perform the test but interpreting them is the tricky part. The readings can provide a lot of information regarding the patient's status and also help plan the next treatment step.

PCV [25-27] is the percentage of red blood cells in circulating blood. A decreased PCV generally means red blood cell loss from any variety of reasons like cell destruction, blood loss, and failure of bone marrow production. An increased PCV generally means dehydration or an abnormal increase in red blood cell production. TS are a measurement of plasma proteins. These proteins include albumin, globulins, and fibrinogens. Decreased TS generally means the animal is suffering from protein loss from any variety of reasons like blood loss, PLE, PLN, or malnutrition. Increased TS usually means dehydration but can be present in certain chronic diseases.

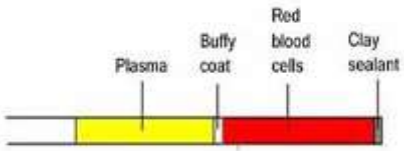


Figure 1: Packed Cell Volume

By looking at a hematocrit tube fresh out of the centrifuge, you can also get some idea for white blood cell content by examining the buffy coat. The buffy coat sits between the red cell layer and the plasma (and should not be counted as part of the PCV) and is normally 1 percent or less. A large buffy coat can signify a large increase in WBC count. The plasma layer should also be examined for hemolysis, lipemia, and icterus. These characteristics should be noted along with the PCV/TS results. Remember to look at both values together to get the entire clinical picture of your patient, and continue to monitor throughout their hospital stay.

- **↑PCV, ↑TS**: This patient is most likely suffering from dehydration. As the water portion of blood is decreased you will see an elevation in both the PCV and TS. Both of these values should decrease as fluid therapy rehydrates the patient.
- **↑PCV, normal TS**: This patient also may be dehydrated, but remember that the addition of fluids will bring both values down, so there is a loss of proteins occurring. Watch this patient for the development of hypoproteinemia and associated clinical signs (hypotension, peripheral edema) as fluids are administered. This patient could be suffering from polycythemia, a rare condition where the body overproduces red blood cells.
- **↑PCV, ↓TS** : This patient could be suffering from dehydration and a profound protein loss. More commonly, you may see this with a recent trauma. This patient may be suffering from acute blood loss and splenic contraction has temporarily increased the PCV, but the low TS point you towards blood loss. In a trauma patient where you expect blood loss it is important to recheck the PCV and TS after starting treatment.
- **Normal PCV, ↑TS**: This is a common scenario with CKD cats. This patient is most likely suffering from anemia and dehydration. The normal PCV may fool you into thinking this animal is okay, but as you rehydrate remember that both numbers will drop leaving you with an anemia to treat.
- **Normal PCV, normal TS**: Normal is good, right? Be sure to match the results to the patient. If this patient sustained recent trauma there may be blood loss that isn't apparent on blood work yet. If the results are different than what you expected, then recheck the PCV/TS as you begin treatment.
- **Normal PCV, ↓TS**: This patient is most likely suffering from a protein losing disease (PLN, PLE), chronic diarrhea, or certain liver and kidney diseases. Be prepared for clinical signs of hypoproteinemia (hypotension, peripheral edema) and their treatment.
- **↓PCV, ↑TS**: The elevated TS most often points to dehydration, and remembering that the addition of fluids will further drop the PCV, this patient is anemic and needs close monitoring of the PCV and most likely the addition of blood products.
- **↓PCV, normal TS**: This patient is suffering from RBC destruction lack of production. In blood loss we expect to see a decrease in the TS as well. With just the red cells decreased be on the lookout for IMHA.
- **↓PCV, ↓TS**: This patient is suffering from whole blood loss and needs to be monitored very closely. Blood products should be considered in treating this patient.
- Armed with this information, you now know a great deal about your patient just by looking at the PCV/TS. By using critical thinking skills and knowledge of the treatment plan for these patients you can prepare for their future needs.

A. Reading the PCV

A lower number of the PCV means that the RBC count loss is due to reasons such as blood loss, cell destruction and less bone marrow production. Increased PCV can generally mean that a person is dehydrated and there is a higher number of RBC productions. By looking at the tube out of the centrifuge, you can get an idea of the WBC content as well. This buffy coat normally lies between the plasma and red cell layer. (This shouldn't be counted as a part of the PCV test). The layer of plasma should also be examined for lipemia, hemolysis in addition to icterus.

B. Preparation for the PCV Test

There is not any special preparation required for the PCV test. If you are anxious about the test, it is better to talk to the doctor and let him/her know. Also, any medications that you've been taking have to be relayed to the doctor. If there are any medical problems which are underlying too, you need to fast before taking a test.

C. Uses of Packed Cell Volume Test

A low PCV implies that the patient has a low number of red blood cells and is suffering from anaemia. The doctor may ask the patient to undergo further tests to determine the underlying causes of anaemia. Treatment will be given accordingly.

D. Measuring the PCV Test

The PCV test is calculated with the help of an automated analyzer which means that it isn't directly measured. By multiplying the red cell count with the mean cell volume, doctors get the final amount. PCV is slightly less accurate than the hematocrit as they include small amounts of the plasma from the blood that is generally trapped in between two red cells. By tripling the haemoglobin concentration and dropping the units, an estimated hematocrit can be determined in percentage.

The PCV can also be determined with the help of the capillary tube and the centrifuging heparinised blood in it at around 10000 RPM for roughly five minutes. This process helped in separating the blood into different layers, and the volume of the total packed RBC divided by the blood sample's total volume gives the final amount of the PCV. Since a tube is also used, it can be used to measure the lengths lying between certain layers.

There is also another way to measure the levels of hematocrit, and this is through optical methods such as spectrophotometry. With the help of differentials, the differences between the optical densities of the sample flowing through the glass tubes at isosbestic wavelengths and the product containing the luminal diameter along with the hematocrit can create a linear relationship.

E. When Does A Low PCV Reading Occur?

There are certain conditions that contribute to the low reading in the PCV. These include:

- Nutritional deficiencies of iron or vitamin (B12 or folate) and mineral deficiencies
- Bleeding
- Inflammatory conditions, for example, rheumatoid arthritis
- Kidney diseases
- Haemolysis, which is the situation where the RBCs are destroyed prematurely by the immune system. This occurs due to certain organ damages and inherited abnormalities of the RBCs
- Liver cirrhosis
- Medicines - including that of chemotherapy
- Abnormalities of RBCs or haemoglobin containing disorders such as myelodysplastic syndrome, lymphoma, bone marrow disorders and myeloma
- One of the most common causes of heightened PCV readings is that of dehydration. With adequate fluid intake, the levels return to normal, but it can also create a condition known as polycythaemia where there are more RBCs

III. Image Enhancement

Image enhancement includes sharpening, contrast manipulation, filtering, interpolation and magnification, pseudo coloring, and so on. The greatest difficulty in image enhancement is quantifying the criterion for enhancement. Therefore, a large number of image enhancement techniques are empirical and require interactive procedures to obtain satisfactory results. However, image enhancement remains very important because of its usefulness in virtually all image processing applications. Color image enhancement may require improvement of color balance or color contrast in a color image. Enhancement of color images becomes a more difficult task not only because of the added dimension of the data but also due to the added complexity of color perception [11].

Image enhancement techniques are used to improve the appearance of the image or to extract the finer details in the degraded images. The principal objective of image enhancement is to process an image so that the result is more suitable than the original image for a specific application. A method that is quite useful for enhancing one category of images may not be necessarily be the best approach for enhancing other category of images. Color image enhancement using RGB color space is found to be inappropriate as it destroys the color composition in the original image. Due to this reason, most of the image enhancement techniques, especially contrast enhancement techniques, use HSV color space [12].

Image enhancement methods may be categorized into two broad classes: transform domain methods and spatial domain methods. The techniques in the first category are based on modifying the frequency transform of an image, whereas techniques in the second category directly operate on the pixels. However, computing a two dimensional (2-D) transform for a large array (image) is a very time consuming task even with fast transformation techniques and is not suitable for real time processing.

Image enhancement is basically improving the interpretability or perception of information in images for human viewers and providing `better' input for other automated image processing techniques. The principal objective of image enhancement is to modify attributes of an image to make it more suitable for a given task and a specific observer. During this process, one or more attributes of the image are modified. The choice of attributes and the way they are modified are specific to a given task.

IV. MATERIALSAND METHODS

Blood samples were placed on filter paper, and images were obtained with a smartphone in different environments. These results were then compared to the PCV measurements on the same samples following the World Health Organization guidelines.

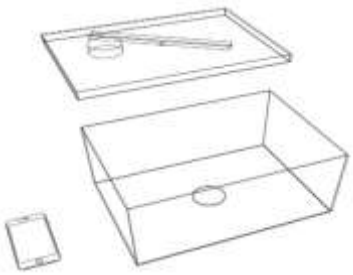
- **Collection of Samples**

Canine samples were obtained from a selection of cases presenting to the Hospital for Small Animals. These samples were surplus to clinical requirement and would have otherwise been discarded as clinical waste. They were solely based on availability, and no samples were excluded from analysis .All samples were analyzed within 12 hours of being obtained and were refrigerated between collection and analysis.

- **Equipment**

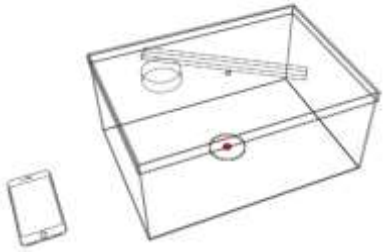
To minimize external sources of variation, particularly lighting, a controlled environment was created. To achieve this, a commercial, white, paper-based box with the dimensions 27 cm × 35 cm × 20 cm was used. This box was modified with a 1 cm diameter hole created in the lid, to provide a position for the camera lens (Figure 2), without allowing external light entry. Handle spaces were sealed with the same paper-based material. A single sheet of bright white, 80 g/m2 A4 paper† was placed at the bottom of the box and was replaced throughout to prevent any blood contamination, which may interfere with color analysis of subsequent Samples .A commercial 35 cm, 30 lumen, battery-powered lighting strip was secured to the underside of the lid. A fresh piece of 90 mm, grade 1 filter paper§ was used for each sample. A circle, the size of the filter paper, was traced onto the bottom of the box, to allow consistent positioning.

Step-1



Step-2

Step-3



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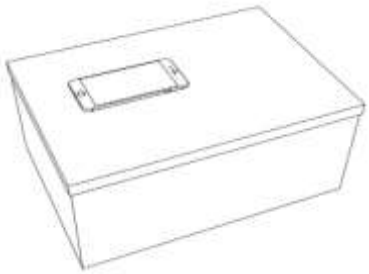


Step-5

Figure 2: Schematic representation of the controlled environment created for analysis

The position of the smartphone on the outside of the lid was outlined to ensure it was placed in the same position each time. Representative images obtained with this technique are shown in Figure 3.

Representative images obtained are shown in Figure 4, and a schematic diagram of this appearance from a horizontal plane is depicted in Figure 4. All images were obtained using a smartphone.

- **Image Acquisition**

Volume of blood and the environment in which images were acquired were the 2 variables that needed to be standardized. The potentially confounding variables ameliorated by the controlled environment include the distance from the camera to the blood spot, the angle of the camera in relation to the filter paper, degree of movement while image was acquired, and ambient lighting.

A drop of mixed blood was applied on to the center of the filter paper. To standardize volume, a pipette and tip were used to aliquot exactly 20 μL of blood. For non-standardized volume, disposable 2mL plastic droppers, which have a drop volume stated to be 22 μL according to the manufacturer, was used to deliver 1 drop. For each sample, the sample conditions were analyzed in the same order that is, starting with pipette in uncontrolled environment and finishing with dropper in controlled environment. After 30 seconds, the filter paper was either left in place or transferred to the box if the sample was to be assessed in a controlled environment. Five images were acquired in rapid succession with the smartphone, taking approximately 1 second to obtain, in order to provide a selection of samples to evaluate for unaccounted variation between images.

This was to provide an adequate amount of time for the blood spot to wick into the paper, while allowing the technique to still be performed more rapidly than the standard PCV measurement method. In uncontrolled conditions, the photograph was

obtained at a height so that the filter paper filled the screen. This distance was not measured. There were no adjustments made to the lighting of the room, although images were taken in the absence of daylight and with only ambient overhead fluorescent strip bulbs. The images were evaluated grossly for any evidence of movement artifact and would have been reacquired had this been evident.

For a standardized environment, the filter paper with the blood spot was placed within the box. Thus, images were obtained from 4 different experimental conditions: dropper used out, with a standardized environment; pipette used out, with a standardized environment; dropper used inside a standardized environment; pipette used inside a standardized environment.

For the alternative technique, the coverslips were placed at either end of one of the microscope slides, with the other microscope slide then overlaid. A dropper was then used to fill the created aperture between the slides via capillary action. Enough blood was instilled to fill the area between the 2 coverslips. This was then transferred into the box and images acquired.

- **Image Processing**

Images were transferred from the smartphone to a computer and imported into Image for analysis. An example of the images obtained is shown in Figure 3 for the filter paper method and Figure 4.3 for the microscope slide method.

Figure 3: Representative images from 2 cases, with the dropper method in a controlled environment, with the image on the left having a PCV of 0.05 L/L [5%], and on the right a PCV of 0.45 L/L [45%]

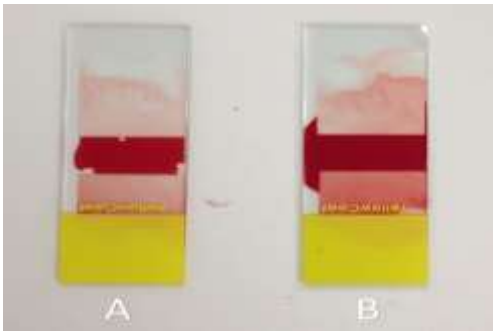


Figure 4: Representative image from the microscope slide method

Using a stylus, the largest, subjectively homogenous area within the blood spot was traced. This was to avoid the inclusion of blood that had not fully absorbed into the filter paper, as this led to light reflection and would have artifactually altered the mean color intensity of the sample.

From the area encircled, the program assigned each pixel a numerical value, with the value increasing with an increase in the whiteness of the pixel. The higher the mean color intensity (MCI) is the paler and brighter the pixels, which would be expected to be concordant with a lower PC. An MCI of the pixels within the encircled area is then generated. This process was repeated for all the images obtained for each sample, and a mean of the 5 images was then calculated. There was no statistically significant difference between a single image and the mean of the 5 images.To assess whether obtaining a mean from 5 images was necessary, 20 samples were reassessed and 1 of the 5 images was chosen at random and analyzed. The single image was then compared against the mean.

- **Work Statistics**

The mean relative color intensity was recorded against the manually measured packed PCV in Microsoft Excel. The Bland–Altman plots were generated using MATLAB. Data were tested for normality using a Kolmogorov–Smirnoff test.

A scatter plot was generated by plotting the relative color intensity of the blood spots against measured PCV. A trend line was then generated based on the data points, with an associated R^2 value. A Pearson correlation coefficient was performed to test whether the relationship was statistically significant. Following the classification by Evans, [20] this correlation was then designated as very weak (0–0.19), weak (0.2–0.39), moderate (0.4–0.59), strong (0.6–0.79), and very strong (0.8–1.0).

To assess the agreement between the predicted PCV and actual PCV, a Bland–Altman plot was generated. The percent error was calculated by dividing the difference between the predicted PCV and actual PCV, divided by the actual PCV and multiplied by 100. The standard deviation of the error percentage was calculated as a whole. The samples were then split into 3 groups based on measured PCV and classified based upon a single laboratory's reference intervals: high (PCV >0.55 L/L [>55%]), normal (0.38–0.55/L [38–55%]), and anemic (<0.38 L/L [<38%]). The anemic group was further divided into marginal (0.20–0.37 L/L [20–37%]) and marked (<0.20 L/L [<20%]) for further analysis. The standard deviation of the error percentage was calculated for each. A cut-off of 0.2 L/L (20%) was used based on this being a transfusion trigger used in human medicine, as a set cut-off has not been chosen in canine patients. [21] To assess whether there was any significant difference between the mean of 5 samples or 1 sample at random, a 1-sample *t*-test was performed. A *P*-value <.05 was deemed significant.

V. Result and Discussions

Firstly, I take image samples for smartphone then remove the noises of images and analyze the all results. Twenty-four samples were analyzed in the 4 combinations. Eight of these samples were from anemic patients (PCV <0.39 L/L [<39%]), had erythrocytosis (PCV >0.55 L/L [>55%]), and the remaining 15 were within reference interval (0.39–0.55 L/L [39–55%]). Of this set, 1 sample was hemolyzed and another lipemic. All the data were normally distributed (Kolmogorov–Smirnoff, 0.21). When a plastic dropper was used out with a controlled environment, there was a mild positive correlation with an R^2 of 0.4049 (P <.0001), as demonstrated in Figure 5.

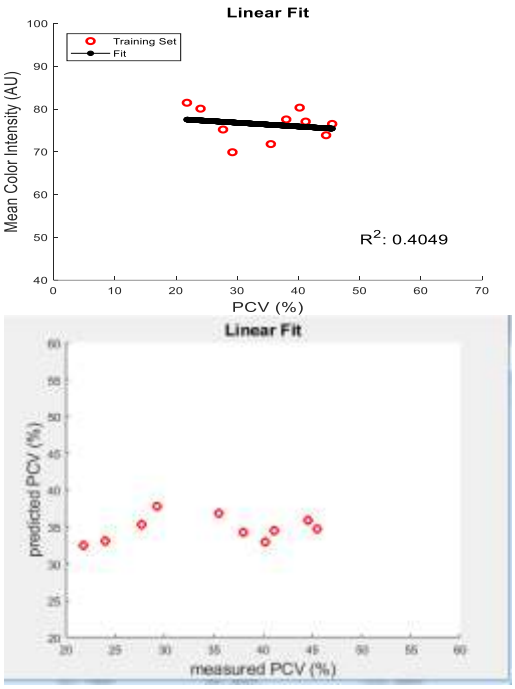
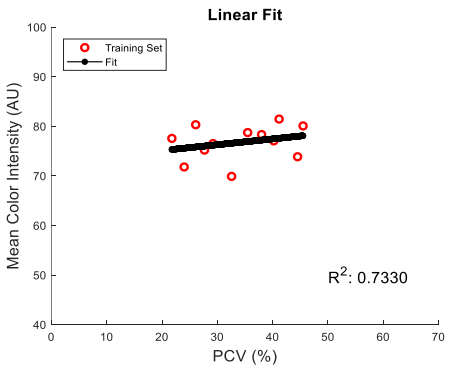


Figure 5: The liner correlation achieved between the image analyses when obtained with a dropper in an uncontrolled environment (mean color intensity) and the measured PCV (n = 10). Line of best fit has an R^2 of 0.4049 (P ≤ .0001). AU, arbitrary unit



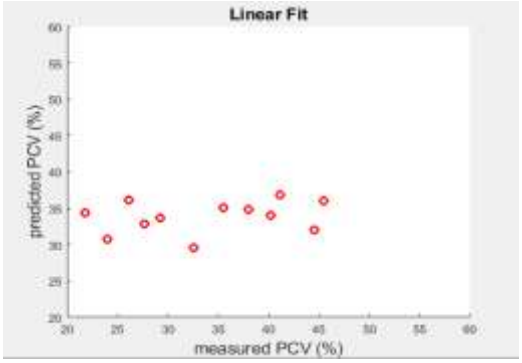
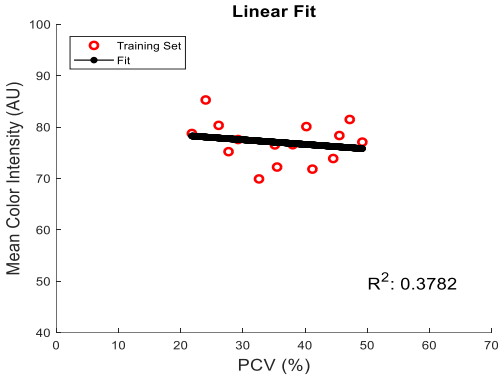


Figure 6: The linear correlation achieved between the image analysis when obtained using a standardized pipette in an uncontrolled environment (mean color intensity) and the measured PCV (n = 12). Line of best fit has an R^2 of 0.7330 (P < .0001)



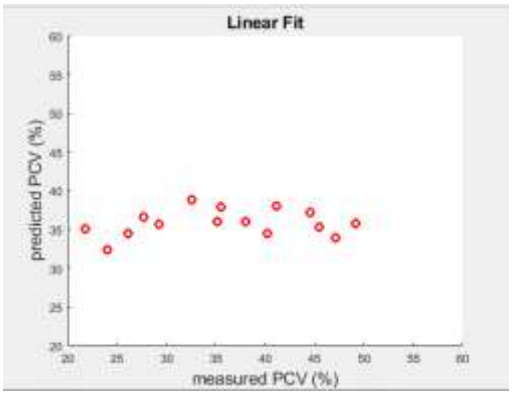
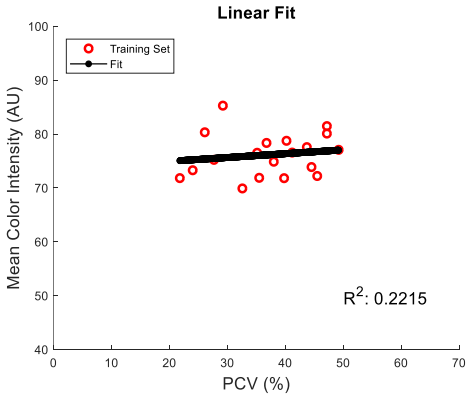


Figure 7: The linear correlation achieved between the image analysis when obtained with a controlled environment and a standardized pipette (mean color intensity) and measured PCV (n = 15). Line of best fit has an R^2 of 0.3782 (P < .0001)



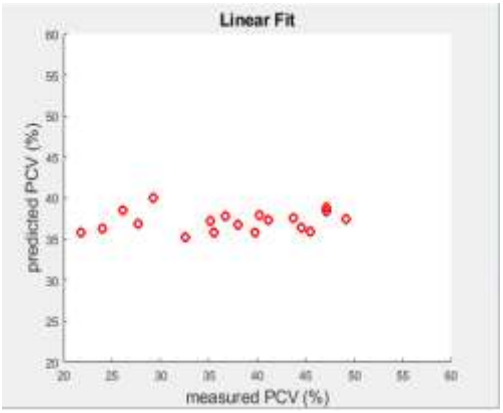


Figure 8: The linear correlation achieved between the image analysis when obtained with a controlled environment and the use of a dropper (mean color intensity) and the measured PCV (n = 19). Line of best fit has an R^2 of 0.2215 (P >.0001)

When a standardized drop from a pipette without environmental control was assessed, there was a strong correlation as demonstrated in Figure 6. When a 20 μL standardized drop from a pipette with a controlled environment was evaluated, there was a strong correlation with an as demonstrated in Figure 7. When the plastic pipette with a controlled environment was used, the correlation was identified to be strong with an R^2 of 0.2215 (P <.0001), as demonstrated in Figure 8.

In view of these results, the combination with the strongest correlation, the plastic pipette within the controlled environment, was used for additional samples. A further 45 samples were added to the analysis, yielding a total of 69 samples for this method. It was from these additional samples that the equations were generated. Twenty-nine samples were anemic (PCV<0.38 L/L [<38%]), 38 were within the reference interval (PCV of 0.38–55 L/L [38–55%]), and 2 samples were polycythemic (PCV of 0.55 and 0.6 L/L [55–60%]). There was a total of 2 hemolyzed samples and 1 lipemic sample. There were no grossly icteric samples available. The R^2valuewhen only anemic samples, with a PCV<0.38 L/L [<38%], were included was 0.85 (n=29). The R^2 value for the more severely anemic samples, with a PCV <0.2 L/L (<20%), was 0.78 (n = 17).

The correlation between the measured PCV and extrapolated using the dropper in a controlled environment strengthened further, this yielded the equation of the line of y=−0.5407*x+74.49. This is demonstrated in Figure 9. The predicted PCV was calculated by inputting the MCI value into the equation of the line of best fit. With the additional nine clinical samples that had the PCV predicted based on the dropper method in a controlled environment, using the equation of the line (y = = −0.542x + 74.945) the data in Table 1 was achieved.

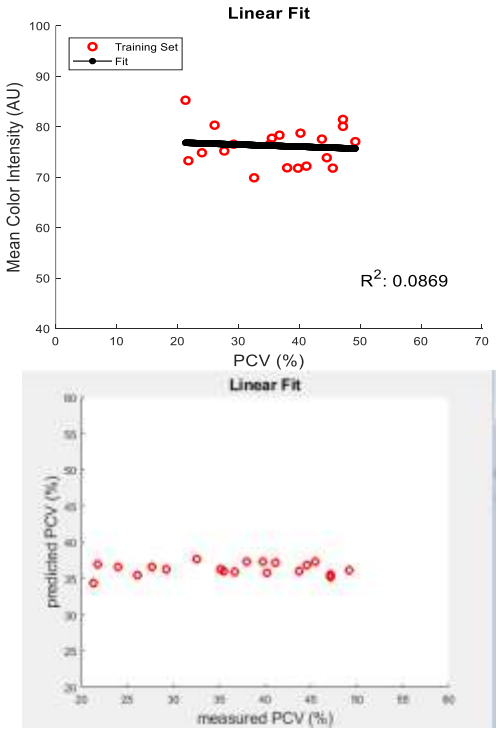


Figure 9: The linear correlation achieved between the image analysis when obtained with a controlled environment and the use of a dropper (mean color intensity) and the measured PCV (n = 20). Line of best fit has an R^2 of 0.0869 (P < .0001)

To account for there being a nonlinear relationship between PCV and relative color intensity, a 4-parameter logistic (4PL) curvewas generated. The larger data set, evaluating the samples photographed in the box with the use of the dropper, was assessed in this way. The R^2 for this 4PL generated curvewas 0.926 (P<.0001). Using the linearmodel, 95% of the degree of error (in percent) for the mean of the predicted PCV ranged from−7.8% to 7.6%. The standard deviation of the percent errorwas 31.9% for all the samples together. This standard deviation of the percent error was reassessed for the subgroups as follows: anemic 48.4%,within normal limits 9.1%, and erthryocytosis 1.4%.

Table 1: The predicted PCV utilizing the equation of the straight line generated by plotting the mean color intensity (MCI) against the measured PCV, using the dropper method within a controlled environment

	et	measured_PCV	predicted_PCV	PCV_difference
1	1	27.6840	36.5425	-8.8586
2	2	44.5173	36.8263	7.6910
3	3	26.0925	35.4554	-9.3629
4	4	32.5841	37.6703	-5.0862
5	5	49.2036	36.1479	13.0557
6	6	35.1331	36.2641	-1.1310
7	7	47.1771	35.2123	11.9648
8	8	47.1771	35.5057	11.6714
9	9	43.7145	36.0411	7.6734
10	10	39.7880	37.2654	2.5226

Description of code:

- First a folder having data i.e., blood images is selected.
- Selected blood images are used to calculate MCI (Mean Color Intensity).
- Then a folder having reports are selected.
- With the help of given information of RBC value and PCV value, measured PCV is calculated.
- Then for predicted PCV, Line of best fit is calculated by Least Square Method.
- PCV difference is then calculated by subtracting predicted PCV from measured PCV.
- Then Coefficient of Determination i.e., R^2 is calculated. It is used to tell that how much the algorithm is capable to model the data.
- Then graph is plotted between measured PCV vs MCI.

VI. Conclusion

Current PCV measurement method requires several pieces of equipment, which reduces its availability for both financial and logistic reasons, such as the need for electricity. The necessary equipment also reduces its portability, limiting its use in field situations. A smartphone application would overcome these limitations. The main limitation is that a controlled environment is required to improve the correlation and predictive ability of the technique. In this study, the controlled environment was deliberately created from widely available materials in order to maximize its applicability. One method by which environmental effects could be ameliorated would be the use of a standardized, widely available, reference point of color with which to normalize the results when processing the image.

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