Performance of Sysmex® XT, Sysmex® XN-Vet and manual methods on Chicken blood: a comparison study

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INTRODUCTION

CBC is an essential part of health evaluation when there is increased mortality in avian farms. Though, data regarding broiler chickens' haematology is scarce in scientific literature. Because they have nucleated red blood cells and thrombocytes, birds' haematology parameters are challenging to evaluate and manual methods are preferred. However, it is time consuming, especially when dealing with large numbers series. We hypothesized that new generation haematology analysers might be of help.

Objective of the study: To compare performance between data obtained by Sysmex XT, Sysmex XN-Vet and manual methods to determine if automated analyser can be used in poultry haematology.
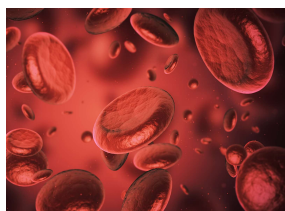
EXPERIMENTAL DESIGN

The prospective study was performed between November 2021 and November 2022. Experiments were carried out on fresh EDTA anti-coagulated blood (4ml tubes) from 309 ROSS308 male chicken, aged from 10 to 35 days, with unknown health status. Tubes filled with less than 1ml blood or containing clots were discarded. All samples were processed within 36 hours after collection. All samples were submitted to automatic analysis of RBCs and WBCs parameters on Sysmex®XT-2000 and 52 samples were also analysed with XN-Vet.
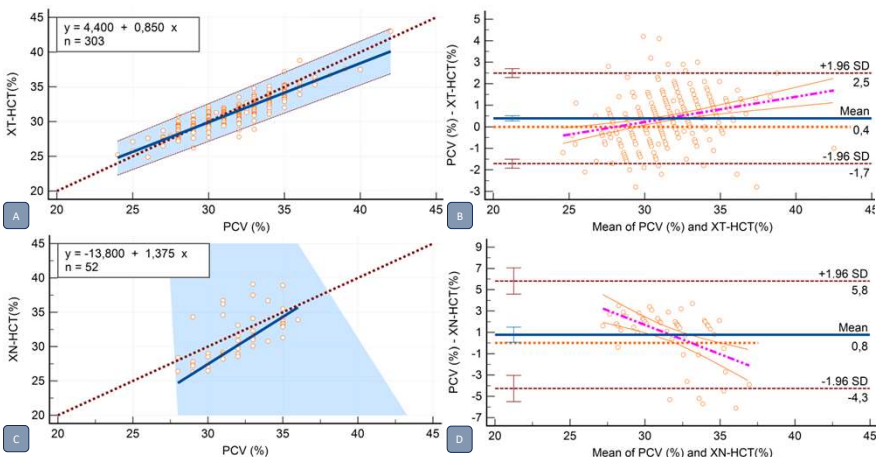
Manual methods (n=304) included packed cell volume (PCV), performed in duplicate (13.000g centrifugation, 10 min) and white blood cell (WBC) counts determined in a Neubauer chamber using Natt–Herrick's method. Differential counts were made on May Grunwald Giemsa-stained blood smears.

Statistical comparison of the results obtained with the automated and the manual methods was performed with a Spearman rank correlation and Passing–Bablok regression analysis. Bland–Altman diagrams showing the mean bias ±1.96 standard deviation were drawn. Correlations were ranked as "excellent" for Spearman r = 0.93–0.99, "good" for r = 0.80–0.92, "fair" for r = 0.59–0.79, and "poor" for r< 0.59.

RESULTS & DISCUSSION

- There was good correlation between the gold standard PCV and HCT measured with Sysmex® XT analyser XT-HCT (r=0.89 [0.86;0.91]) and fair correlation between XN-HCT and PCV (r=0.69 [0.51;0.81]).

- In all cases, a systematic error (SE) was present. When looking at the regression curve on Passing-Bablok graph (Figure 1), SE could be unbundled in a constant error, corresponding to the intercept and a proportional error, corresponding to the slope.

Figure 1 A. C. Passing-Bablok regression analysis of XT-analyser HCT (%) vs. manual method PCV (%), XN-HCT vs. PCV respectively with regression lines (blue), confidence bands (dashed), identity lines (red). B. D. Bland-Altman analysis, plot of differences between PCV and XT-HCT, PCV and XN-HCT vs. the mean of the two measurements respectively, equality lines (dotted), means (blue), regression lines (pink) with confidence interval limits.

- Constant and proportional errors, as well as the bias, were summarized in Table 1.

	Constant error	Proportional error	Bias (%)
XT-HCT vs. PCV	4.40 (95%CI 2.80;5.58)	0.85 (95%CI 0.81;0.90)	1.15 (95%CI 0.77;1.54)
XN-HCT vs. PCV	-13.80 (95%CI -30.20;-6.93)	1.38 (95%CI 1.16;1.90)	3.16 (95%CI 1.07;5.25)

Table 1 Constant, proportional errors and bias for both XT and XN-HCT vs. PCV

- These data suggested that Sysmex® XT analyser performed better than XN-Vet in automatic determination of HCT when compared to the gold standard PCV.

- As for both analysers a proportional error was identified, caution is advised for extreme values' interpretation. This is especially true for XN Vet due to the limited number of data available and the narrower range of values obtained.

- Moreover, as equality's line was not within the confidence interval of the mean difference, the methods couldn't be regarded as interchangeable, and, specific reference intervals for the different methods should be provided.

- Considering previously published CV for XN-Vet (Grebert, 2021) and XT analysers (Bourgès-Abella, 2011) below 1%, the bias were considered acceptable as TE$_{Obs}$ (2CV + bias): 5.16 and 3.15 respectively were lower than TE$_a$: 10% proposed in ASVCP guidelines.

- There was poor correlation and no agreement between WBCs manual counts and automated counts (Figure 2).
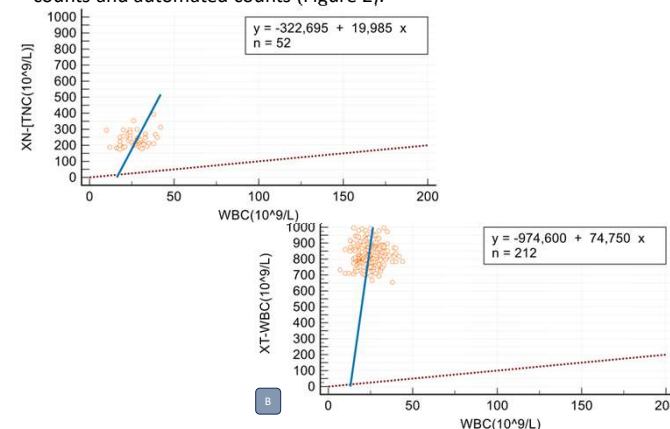

Figure 2 A. B. Passing-Bablok regression analysis of XT-WBC vs. manual method, XN-TNCC vs. manual method respectively, regression lines (blue), identity lines (red).

CONCLUSIONS

- XT and XN-Vet automated analysers could be used to evaluate haematocrit in poultry.
- XT performed better than XN-Vet for HTC determination.
- Specific reference intervals for the methods should be provided.
- The analysers were not usable to evaluate WBCs in chickens.

Suggested Literature : Al-Nedawi A. Reference hematology for commercial Ross 308 broilers. OJVR, volume 22 (7):566-570, 2018.
Goodwin M. PCV reference intervals to aid in the diagnosis of anemia and polycythemia in young broiler chickens. Avian Diseases 36:440-443, 1992.

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