

Summary

- SCiO Cup is a portable NIRS instrument for quick dry matter determination
- Variation in moisture, particle size, temperature, and instrument affect accuracy of NIRS predictions
- Dairyland Laboratories evaluated accuracy of SCiO Cup dry matter determination compared to laboratory analysis for alfalfa haylage, corn silage, and fresh hay across five instruments over a wide range of moisture and temperature
- Dairyland Laboratories recommends using SCiO Cup for dry matter determination within target harvest ranges for alfalfa haylage, corn silage, and fresh hay
- When measured at cold temperatures above freezing, the corn silage model illustrated a slope issue, though the standard error was still within expectations.

Background

Monitoring dry matter (DM) is crucial for crop producers to determine when to harvest and for formulators to adjust rations to maintain a consistent nutrient profile in the diet. Formulators may opt to send samples to commercial laboratories for analysis, which is considered the “gold standard”, but the cost is relatively high and results take several days. Other options include drying the sample in a microwave or using a Koster Tester. These methods are both prone to error and can result in fires if the sample is not monitored in the microwave or if the Koster tester is placed near flammable material. Food dehydrators offer simplicity and safety, but it may still take over a day for results (Dyk, 2010).

The SCiO Cup (Figure 1; Consumer Physics, St. Cloud, MN), a portable on-farm near infrared spectroscopy (NIRS) device, offers another option. Producers can scan samples and receive DM predictions instantly. On-farm NIRS offers convenience due to the speed of the analysis, reduced cost, and non-consumption of the sample. The SCiO Cup offers a unique design, including a wide scanning surface and lid to reduce light leak.

Variation in moisture and particle size can cause issues with NIRS prediction accuracy. This is true even with laboratory NIRS instruments when samples are dried to > 90% residual DM and finely ground (Baker et al., 1994). These issues are exacerbated for on-farm NIRS instruments because samples are not dried or ground prior to scanning. Goeser (2022) evaluated SCiO Cup for corn and legume silages across multiple locations and SCiO Cups (instruments) and reported acceptable agreement between the SCiO Cup and laboratory DM. While they used multiple cups in their study, they did not report instrument to instrument variation. The recommended method of scanning the same samples on all instruments to standardize them and decrease variation with calibration transfer (Bouveresse and Campbell, 2008) is not feasible for on-farm NIRS due to complications with preserving fresh and wet samples. Furthermore, temperature fluctuation in the field is another concern for on-farm NIRS. Even small changes in temperature in a laboratory setting affect NIRS predictions (Shenk and Westerhaus, 1991).

Therefore, the objectives of this study were to:

- evaluate SCiO Cup predictions of dry matter against laboratory analysis,
- compare variation among instruments,
- and evaluate accuracy of SCiO Cup predictions when temperature fluctuates.

Methods

Experiment 1

Ensiled alfalfa haylage (AH; $n = 35$), ensiled whole plant corn (WPC; $n = 30$), and fresh hay (FH; $n = 30$) were collected from incoming customer samples at Dairyland Laboratories (Arcadia, WI; Table 1). Samples were first scanned three times using a SCiO Cup (Consumer Physics), using the “Legume silage”, “Corn silage, and “Green chop haylages” applets (widgets) for AH, WPC, and FH, respectively. The contents of the cup were emptied, mixed by hand, then re-introduced to the cup between each scan. This procedure was repeated across each of five (1 – 5) SCiO Cup instruments. Contents were transferred to plastic cannisters, weighed, then dried in a forced air oven at $< 60^{\circ}\text{C}$ to approximately 95% residual DM. The samples were weighed then ground to pass a 1-mm screen in an abrasion mill (Udy Corp., Fort Collins, CO) and analyzed by near-infrared spectroscopy (NIRS) developed by Dairyland Laboratories to determine residual DM (NFTA Method 2.1.4; Shreve et al., 2006) and chemical composition (ash: method 942.05, CP: method 990.0, aNDFom: method 2002.04, EE: method 920.39, AOAC International, 2012, and starch using the acetate buffer method described by Hall (2009) with YSI Biochemistry Analyzer modification). Final DM was determined according to Equation [1]:

$$\frac{\text{Dry weight}}{\text{Wet weight}} \times \text{Residual dry matter} \quad [1]$$

The standard error of prediction (SEP) was calculated according to Equation [2]:

$$\sqrt{\frac{\sum(x_i - y_i)^2}{n}} \quad [2]$$

where $x_i - y_i$ = difference between results obtained by laboratory DM (x_i) and reference method (y_i) on sample i and n = total number of samples. The bias was calculated according to Equation [3]:

$$\frac{\sum(x_i - y_i)}{n} \quad [3]$$

Data were averaged across the effect of instrument and regression analysis was performed using the ‘lm’ function in R. Deviation of slopes from 1 was determined using the ‘lstrends’ function from the ‘emmeans’ package and deviation of intercepts from 0 was determined using the ‘emmeans’ function (Lenth, 2022). Significance was declared at $P < 0.05$ and tendencies are discussed when $0.05 < P < 0.10$.

Experiment 2

Samples of WPC and AH were randomly selected for a temperature challenge. The samples were divided into three representative subsamples using the coning and quartering technique. One subsample was cooled to 4°C in a refrigerator before scanning, a second was scanned at room temperature (20°C), and the final subsample was warmed to 39°C in an incubator prior to scanning. All samples were scanned as described previously for Experiment 1 and laboratory DM was determined according to Equation [1]. One SCiO Cup was used for the temperature challenge in order to maintain temperature of the samples during scanning. The SEP, and bias were determined according to Equations [2] and [3], respectively. Regression analysis was performed as described previously for Experiment 1.

Results and Discussion

Experiment 1

Samples were selected to cover the SCiO Cup calibration ranges (Table 1) over a range of sampling methods (conventionally harvested, processed with a chipper/shredder, or scissor clippings). Dry matter and chemical composition of the samples is presented in Table 2. One AH was outside the upper limit for the calibration and was removed from the study. Alfalfa for hay or haylage should be 20-25% DM at cutting, which is covered by the “Green chop haylage” widget, and dried to 30-35% DM for haylage (Digman et al., 2011), which is covered by the “Legume silage” widget.

The comparison of SCiO Cup DM averaged across the five instruments to laboratory analysis for AH is presented in Figure 2. The slope and intercept of the regression line were compared to a 1 to 1 bisector ($Y = x$), where SCiO Cup DM has the same value as laboratory analysis. The slope of the regression line for SCiO Cup vs. laboratory DM was not significantly different from 1, the intercept was not significantly different from 0, and R^2 was 0.9789, indicating that none of the samples tested deviated from the bisector line. These data, with a standard error of 0.9896, indicate that SCiO Cup DM is an acceptable substitute for laboratory DM for AH when using the “Legume silage” widget at room temperature (20°C).

Figure 3 indicates that SCiO Cup predicts DM for WPC with a standard error of $SEP = 1.5215$ and an $R^2 = 0.9453$. The intercept was not different from 0, but the slope tended to be greater than 1 ($P = 0.08$), indicating a tendency to overpredict DM at the upper end of the calibration range (25-55% DM, Table 1). When the samples exceeding 45% DM ($n = 6$) were removed, the slope and intercept did not deviate from the bisector line ($P = 0.1996$ and $P = 0.1623$, respectively). Corn silage should be harvested between 30-40% DM, depending on conservation method (Bagg, 2012). While our data indicate that SCiO Cup is an acceptable substitute for laboratory DM for WPC within this range, caution should be taken what true values are near the ends of the calibration range.

The FH comparisons are presented in Figure 4. Data were averaged across the five instruments and slope and intercept of the regression line were compared to a 1 to 1 bisector. With an R^2 of 0.9734, SCiO Cup explained more than 97% of the variation in dry matter. The SEP of 2.1382 did not exceed expected on-farm variation reported by Weiss et al. (2012), indicating that the error of the instrument is lower than expected variation from multiple samples of the same feed source. The slope and intercept significantly deviated from the $Y = x$ bisector ($P < 0.05$), indicating that instrument underpredicted values at the high end of the range and overpredicted at the low end of the range. SCiO Cup is an acceptable alternative to laboratory analysis for DM in FH when using the “Green chop haylages” widget.

The SEP , bias, and R^2 across instruments are presented in Table 3. The R^2 was always lowest for WPC, which might be attributed to larger particle size variation for WPC compared to AH or FH, although particle size was not measured in this study. It may also reflect the heterogeneity of WPC compared to hay - measuring DM in kernels and forage particles simultaneously may be more difficult when samples are not dried or ground. Weiss et al. (2012) reported the standard deviation for DM measured for WPC and haylage measured over 14 consecutive days on eight farms using laboratory-based oven DM was 2.07 and 3.70, respectively, which is slightly greater than the SEP and bias we observed. The SEP and bias were greatest for AH and FH scanned on Instrument 1 but do not exceed the variation reported by Weiss et al. (2012).

The average instrument bias (average difference between laboratory and SCiO measurement) was 0.65 across all instruments and feed types, with a maximum bias of 1.78 for instrument 1 on alfalfa haylage and a minimum bias of 0.08 on instrument 3 for fresh hay. R^2 ranged from 0.9168 to 0.9790 and SEP ranged from 1.02 to 2.62. These data, plus the fact that biases for each instrument were different across feed types, indicate that instrument to instrument variation is low and is limited primarily by the modeling process, not the hardware itself.

Experiment 2

Chemical composition of the samples is presented in Table 4. Two AH samples were flagged as outliers by SCiO Cup. The samples were within the calibration ranges reported by SCiO Cup and were both warmed to 39°C. It is unclear why these samples flagged while the other samples warmed to 39°C did not. SCiO Cup does not provide results when a sample is deemed an outlier, so all of the treatments (cooled to 4°C, room temperature, and warmed to 39°C) for those samples were removed from the study.

The comparison of SCiO Cup DM predictions to laboratory analysis for AH scanned at different temperatures is presented in Figure 5. The slope for the AH samples scanned at 4°C did not deviate from 1 but intercept tended to deviate from 0 ($P = 0.06$). SCiO Cup overpredicted DM for most ($n = 12$) of the AH samples scanned at 4°C, resulting in a bias of -1.39. When the samples were scanned at room temperature (20°C), slope and intercept did not deviate from 1 or 0, respectively, similar to the observations in Experiment 1. Although the SEP and bias were less for AH warmed to 39°C than for AH scanned at room temperature, the slope and intercept for AH that were warmed tended to deviate from the $Y = x$ bisector ($P = 0.09$ and $P = 0.08$, respectively), whereas AH scanned at room temperature did not.

The comparison of SCiO Cup DM predictions to laboratory analysis for WPC scanned at different temperatures is presented in Figure 6. When measured at cold temperatures above freezing (4°C), the slope for the corn silage was greater than 1 ($P < 0.05$) and intercept was less than 0 ($P < 0.05$). Most ($n = 9$) of the samples scanned at 4°C were overpredicted, resulting in a bias of -1.94. The slope and intercept for the samples scanned at room temperature (20°C) did not deviate from 0, similar to the observations in Experiment 1 when DM was within the target range for harvest. The slope and intercept for the samples warmed prior to scanning (39°C) also did not deviate from 1 or 0, respectively, but SEP was higher compared to samples scanned at room temperature. Consistent with the observations for AH, SEP and bias were not outside the range of acceptable variation at any temperature, but SCiO Cup DM predictions were optimal at room temperature (Table 5). The fact that we did not observe a similar issue with alfalfa haylage may indicate it can be overcome in model development.

Conclusions

- SCiO Cup predicted dry matter for alfalfa haylage ranging from 30 – 55% with reasonable accuracy and no slope or bias concerns.
- For fresh hay, SCiO Cup dry matter predictions vs. laboratory analysis tended to deviate from $Y = x$, but SEP and bias remained below reported on-farm dry matter variation.
- Accuracy decreased above 45% dry matter for corn silage, but that is above the target range to harvest.
- Variation among the five instruments tested was minimal.
- SCiO predictions of dry matter for alfalfa haylage were acceptable at warm or cold temperatures, but optimal at room temperature.
- Predictions of dry matter for corn silage were more sensitive to variations in temperature than alfalfa haylage.
- Dairyland Labs recommends using SCiO for ensiled alfalfa haylage, corn silage, and fresh hay within target dry matter ranges, but accuracy is reduced as temperature deviates from room temperature.

References

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Table 1. SCiO Cup calibration ranges

Widget	Dry matter range, %	Temperature range, °C
Legume silage	25 - 65	5 - 35
Corn silage	25 - 55	3 - 35
Green chop haylages	11 - 70	7 - 35

Table 2. Summary of chemical composition determined by NIRS for samples in Experiment 1

	Dry matter, %	Crude protein, %DM	aNDFom, %DM	Fat, %DM	Ash, %DM	Starch, %DM
Alfalfa haylage (n = 34)	29.4 - 54.0	17.4 - 26.5	31.2 - 45.1	2.76 - 4.45	9.17 - 14.9	0.20 - 4.35
Corn silage (n = 30)	29.5 - 47.7	7.08 - 14.2	25.2 - 42.6	2.97 - 4.54	3.46 - 7.65	28.4 - 43.3
Fresh hay (n = 30)	11.1 - 62.5	15.1 - 30.2	27.5 - 56.1	2.19 - 4.54	9.64 - 15.5	0.25 - 4.28

Table 3. Standard error of prediction (SEP), bias, and R² across instrument for alfalfa haylage, corn silage, and fresh hay for Experiment 1

Feed Type	Statistic	Instrument				
		1	2	3	4	5
Alfalfa haylage (n = 34)	SEP	2.02	1.02	1.12	1.11	1.29
	Bias	1.78	0.14	-0.49	0.33	-0.46
	R ²	0.9790	0.9763	0.9790	0.9743	0.9666
Corn silage (n = 30)	SEP	1.41	2.01	1.50	2.16	1.34
	Bias	0.51	-0.89	-0.55	-1.37	-0.53
	R ²	0.9471	0.9168	0.9403	0.9323	0.9611
Fresh hay (n = 30)	SEP	2.62	2.12	2.33	2.11	2.21
	Bias	1.53	-0.15	0.08	0.37	0.67
	R ²	0.9757	0.9717	0.9658	0.9736	0.9731

Table 4. Summary of chemical composition determined by NIRS for samples in Experiment 2

	Dry matter, %	Crude protein, %DM	aNDFom, %DM	Fat, %DM	Ash, %DM	Starch, %DM
Alfalfa haylage (n = 13)	29.7 - 52.7	19.1 - 25.0	32.3 - 45.1	2.76 - 4.16	9.17 - 14.5	0.82 - 3.88
Corn silage (n = 10)	29.2 - 36.8	7.15 - 8.13	32.7 - 42.2	2.97 - 4.12	3.70 - 4.88	29.9 - 39.1

Table 5. Standard error of prediction (SEP), bias, and R² for alfalfa haylage or corn silage at different temperatures for Experiment 2

Feed Type	Statistic	Temperature		
		4°C	20°C	39°C
Alfalfa haylage (n = 13)	SEP	1.93	1.44	1.13
	Bias	-1.39	-0.68	-0.14
	R ²	0.9677	0.9703	0.9795
Corn silage (n = 10)	SEP	2.31	1.45	1.81
	Bias	-1.94	0.11	0.59
	R ²	0.8852	0.7297	0.5204



Figure 1. SCiO Cup: in-field NIRS instrument

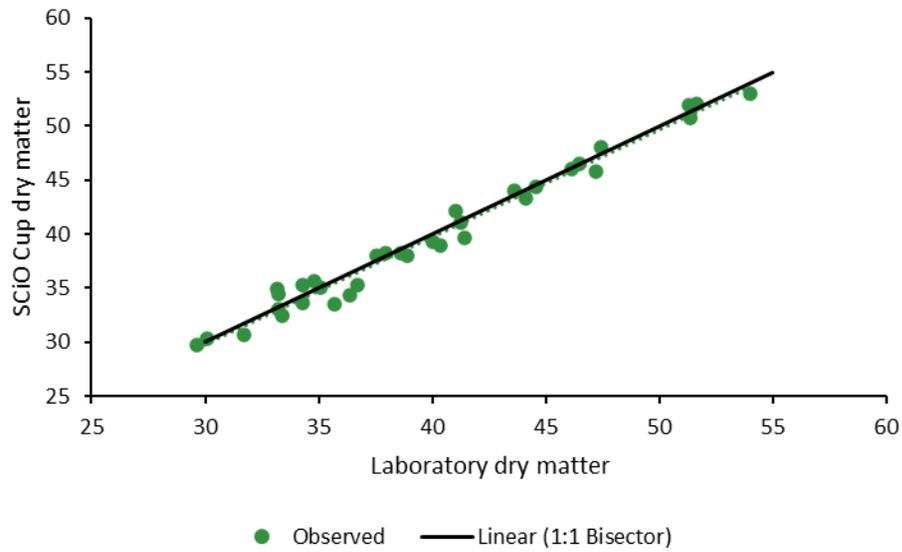


Figure 2. Comparison of dry matters determined by chemistry vs. SCiO for legume haylage (n = 34) averaged across five instruments. $Y = 0.9909x + 0.1037$, $SEP = 0.9896$, $R^2 = 0.9789$; slope was not different from 1 ($P = 0.7145$) and intercept was not different from 0 ($P = 0.921$)

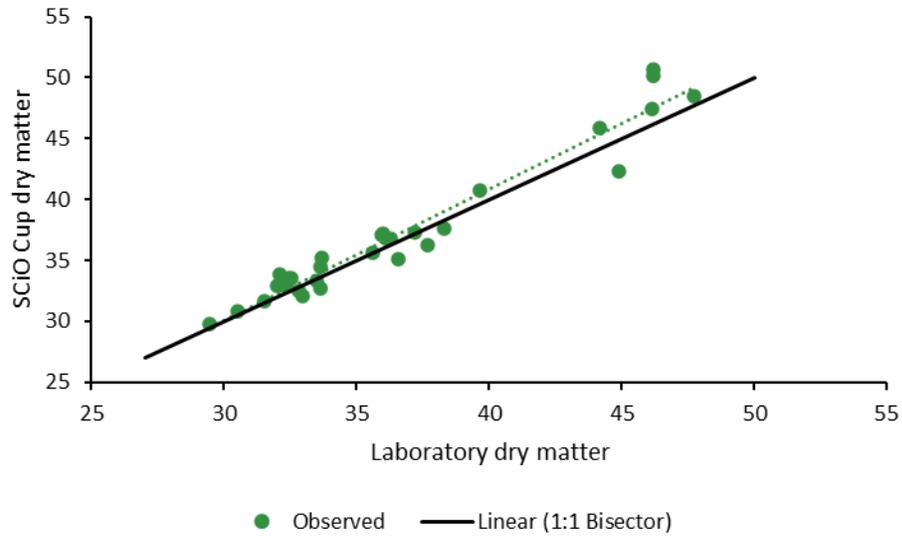


Figure 3. Comparison of dry matters determined by chemistry vs. SCiO for corn silage (n = 30) averaged across five instruments. $Y = 1.0794x - 2.3414$, $SEP = 1.5215$, $R^2 = 0.9453$; slope tended to be greater than 1 ($P = 0.08$) but intercept was not different from 0 ($P = 0.178$)

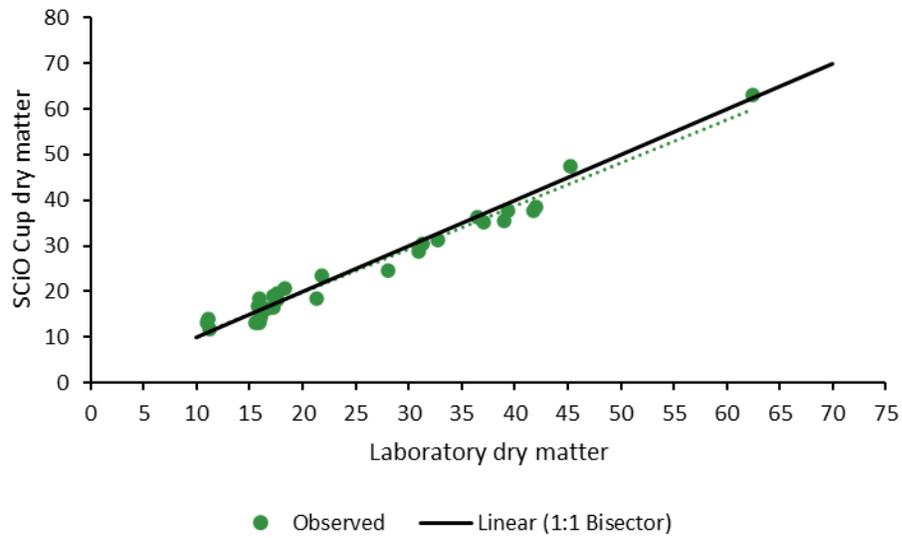


Figure 4. Comparison of dry matters determined by chemistry vs. SCiO for fresh hay (n = 30) averaged across five instruments. $Y = 0.9438x + 0.9251$, $SEP = 2.1382$, $R^2 = 0.9734$; slope was less than 1 ($P < 0.05$) and intercept greater than 0 ($P < 0.05$)

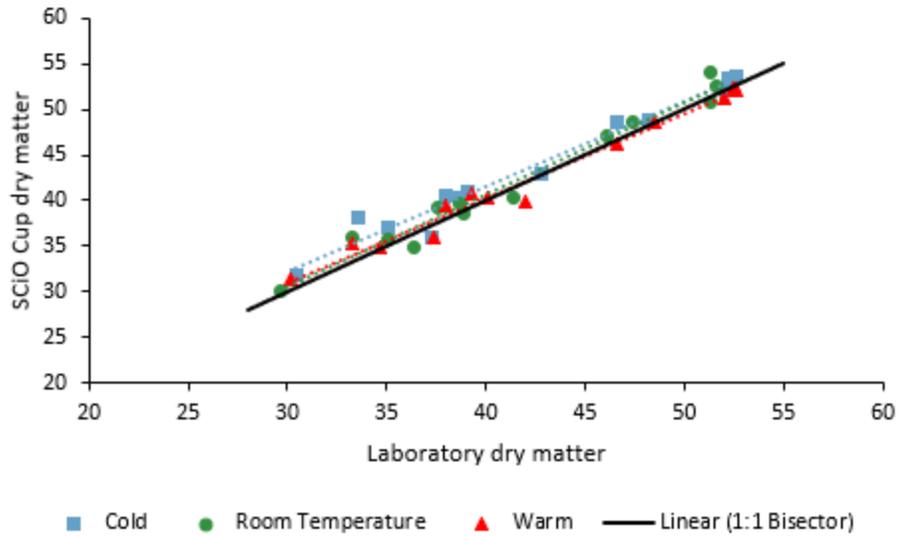


Figure 5. Effect of temperature on accuracy of SCiO Cup for alfalfa haylage (n = 13) when temperature is: a) Cold (4°C): $Y = 0.9311x + 4.2822$, $SEP = 1.93$, $R^2 = 0.9677$, slope was not different from 1 ($P = 0.185$) and intercept tended to be different from 0 ($P = 0.06$); b) Room temperature (20°C): $Y = 1.0119x + 0.1895$, $SEP = 1.44$, $R^2 = 0.9703$, slope was not different from 1 ($P = 0.8201$) and intercept was not different from 0 ($P = 0.9317$); or c) Warm (39°C): $Y = 0.9288x + 3.1293$, $SEP = 1.13$, $R^2 = 0.9795$, slope tended to be less than 1 ($P = 0.09$) and intercept tended to be different from 0 ($P = 0.08$)

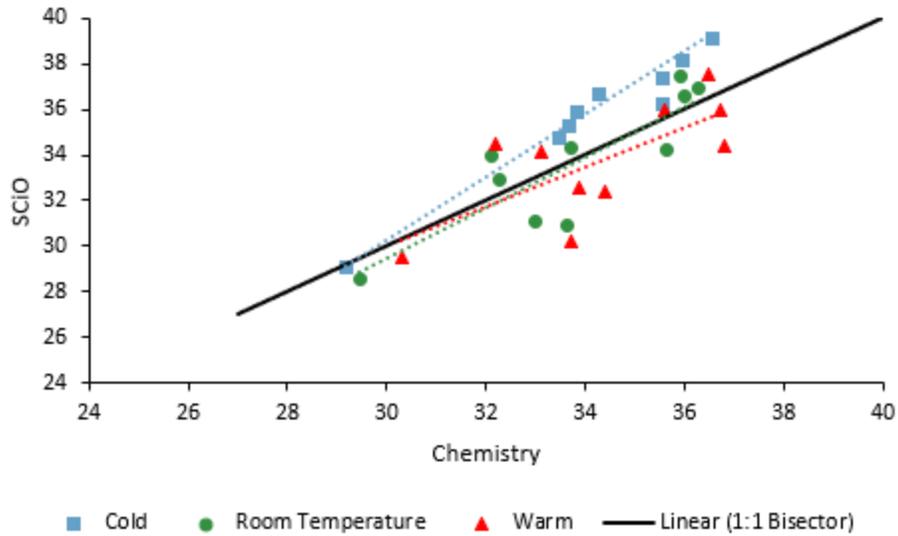


Figure 6. Effect of temperature on accuracy of SCiO Cup for corn silage (n = 10) when temperature is: a) Cold (4°C): $Y = 1.3788x - 11.0721$, $SEP = 2.31$, $R^2 = 0.8852$, slope greater than 1 ($P < 0.05$) and intercept less than 0 ($P < 0.05$), b) Room temperature (20°C): $Y = 1.1167x - 4.0539$, $SEP = 1.45$, $R^2 = 0.7297$, slope not different from 1 ($P = 0.5592$) and intercept not different from 0 ($P = 0.5504$), c) Warm (39°C): $Y = 0.8673x + 3.9599$, $SEP = 1.81$, $R^2 = 0.5204$, slope not different from 1 ($P = 0.5855$) and intercept not different from 0 ($P = 0.6350$)