ICAR CERTIFICATION

Company name: FOSS Analytical A/S

Instrument name: **MilkoScan™ 7 RM**

Milk species: Cow Milk

Version March 2020

Author: Silvia Orlandini

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Preface

MilkoScan™ 7 RM was introduced onto the market at the end of 2016. It is an automatic, high resolution Fourier Transform mid InfraRed (FTIR) spectrophotometer, dedicated for rapid and high capacity determination of milk components. This instrument is provided with Diamond or CaF2 cuvette identified by a red or green interferometer box. The instrument speed is up to 600 samples/h. The instrument is applied in central milk testing laboratories and dairy laboratories for milk recording analysis and milk payment testing.

The instrument can be provided with calibrations for several milk parameters. In this certification, for ICAR DHI purposes, its performance has been evaluated in phase I (single lab test) and in phase II (interlaboratory study) for fat, protein, lactose and urea.

The performance will be evaluated according to limits reported in:

- ICAR protocol “Procedure 1 of Section 12 of ICAR Guidelines – Protocols for Evaluation of Milk Analysers for ICAR Approval” aligned with
- ISO 8196-3|IDF 128-3:2009 – Milk — Definition and evaluation of the overall accuracy of alternative methods of milk analysis — Part 3: Protocol for the evaluation and validation of alternative quantitative methods of milk analysis and
- ISO 9622|IDF 141:2013 Milk and liquid milk products — Guidelines for the application of mid-infrared spectrometry

ISO 8196-3|IDF 128-3:2009 is currently under revision and the next edition will report limits to validate the FTIR instruments and the validation criteria of a new instrument against the previous generation. These limits will be considered as informative.

Summary

Principle

MilkoScan™ 7 RM is an automatic, high resolution spectrophotometer, based on Fourier Transform Infrared (FTIR). The instrument is provided with a diamond or CaF2 cuvette and works at a speed of up to 600 samples/h.

Scope

The scope of the validation was raw cow milk.

Data evaluated

Raw data from Milchprüfring-Bayern (mpr) 2019-2020 Phase I and Phase II

Actalia report 2016 (Phase I)

Interlaboratory study (ILS) statistical elaboration from Qlip (NL)-2020

Table. Performance summary table.

Results of phase I					
Repeatability one instrument		fat range protein range r ≤ 0,019 g/100g	2-4 2-5,8	g/100g g/100g	Limit r≤0,04 (0,02) g/100g
Carry over		$CO_{H/L}$	≤ 0,4 %		Limit CO H/L ≤ 1%
Reproducibility Intralaboratory Phase I		$R_{intrafat}$ $R_{intraprotein}$	≤ 0,06 ≤ 0,06	g/100g g/100g	Limit R_{intra}≤0,08 (0,04) g/100g
Linearity		r linearity	≤ 1%		Limit r linearity ≤ 1%
Accuracy	Accuracy (comparison with FT 6000)	S_{yx} Fat-Protein-Lactose S_{yx} Urea	≤ 0,014 = 2,3	g/100g mg/dl	Limit Syx ≤ 0,04 g/100g
	Accuracy comparison Gerber-Amido black)	S_{yxfat} $S_{yxprotein}$	≤ 0,04 ≤ 0,06	g/100g	Limit Syx ≤ 0,1 (0,06) g/100g
Results of phase II					
Repeatability Interlaboratory Study (ILS)		r_{fat} $r_{protein}$ $r_{lactose}$ r_{urea}	=0,02 =0,02 =0,02 =2,5	g/100g mg/dl	Limit r ≤ 0,04 (0,02) g/100g Limit r ≤ 3,92 mg/dl
Reproducibility Intralaboratory		R_{fat} $R_{protein}$ $R_{lactose}$ R_{urea}	=0,04 =0,03 =0,03 =4	g/100g mg/dl	Limit R_{intra} ≤ 0,08 (0,04) g/100g Limit R_{intra} ≤ not specified
Reproducibility Interlaboratory Study (ILS)		R_{fat} $R_{protein}$ $R_{lactose}$ R_{urea}	=0,06 =0,06 =0,06 =4,7	g/100g mg/dl	Limit R ≤ 0,11 * g/100g Limit R ≤ not specified

*Limit reported in the ISO 9622/IDF 141
Note: the values in red mean that they are going to be approved in the next version of ICAR protocol and ISO 8196-3/IDF128-3

Robustness

Pilot samples analysed at Milchprüfring Bayern (DE) on 12 MilkoScan™ 7 RM each 45 DHI samples for a period of 4 working days gave only 1,55 % of sample out of the tolerance of +/- 0,05 (Limit 5%) for fat parameter.

MilkoScan™ 7 RM showed a very stable performance in between periodic maintenance each 3 months (appr. 450-500 *103 samples).

Final conclusion

The data provided by Milchprüfring Bayern (DE), Actalia (FR) report and results of an interlaboratory study (ILS) organized by Qlip (NL) gave robust evidence of adequate

instrument performance. The instrument performance obtained in the preliminary phase I were confirmed in phase II.

The instrument complies with all criteria limits defined in the ICAR “Procedure 1 of Section 12 of ICAR Guidelines – Protocols for Evaluation of Milk Analyses for ICAR Approval” aligned with the ISO 8196-3|IDF 128-3:2009 and ISO 9622|IDF 141.

Based on the results in this report the FOSS MilkoScan™ 7 RM can be granted the ICAR certificate for milk analysers.

1 Introduction

ICAR certification for milk analysers is intended to serve as a standardised process for certification of milk analysers and to describe instrument performance according to ICAR international guidelines for the DHI analyses for cow milk. ICAR subcontracted expert and accredited laboratories, namelyMilchprüfring Bayern (DE) and Qlip (NL). Milchprüfring Bayern collected the data for phase I and provided information and data on the instrument robustness, Qlip prepared the samples for the Interlaboratory study (ILS), calibration samples for FTIR analyses, chemical analyses of ILS samples and performed the statistical analyses. Also, data of the validation study conducted at Actalia in 2016 were evaluated. The tests performed in phase I, phase II and in the ILS study are robust and independent tools to calculate the performance of this instrument that is operated in milk-testing laboratories for DHI purposes.

2 Company name and instrument under evaluation

Manufacturer FOSS Analytical A/S
Nils FOSS Allé 1, 3400 Hillerød, Denmark

Instrument: MilkoScan™ 7 RM

3 Data evaluated by ICAR

Raw data from Milchprüfring Bayern (mpr) in 2019-2020 (Phase I and Phase II)

Actalia report 2016 (Phase I)

Interlaboratory study (ILS) and statistical elaboration by from Qlip in 2020

4 Instrument principle

The MilkoScan™ 7 RM is an automatic, high resolution spectrophotometer, based on Fourier transform infrared (FTIR), used for the rapid determination of components in milk.

The sample is taken automatically, homogenized, heated loaded in the accumulator and successively injected into the cuvette.

After sample pretreatment, uptake through a pipette, sample homogenization and temperature conditioning, the sample is measured with an infrared spectrometer that records the quantity of radiation absorbed in transmittance in the mid-infrared region (925 - 5000 cm-1). The spectral data are transformed into estimates of constituent concentrations through calibration models developed on representative samples from the population to be tested.

The interferogram is then transformed to spectrum data, and sent through a series of calculations, ending with the final calibrated measured value. After each sample intake the filter pipette is flushed. The first part of the sample volume intake is used to flush the entire flow system and thus to minimize the carry over effect.

Figure 1. Scheme for measurement principle MilkoScan 7 RM (source MilkoScan™ 7 RM User Manual 6007 4040 / Rev. 2).

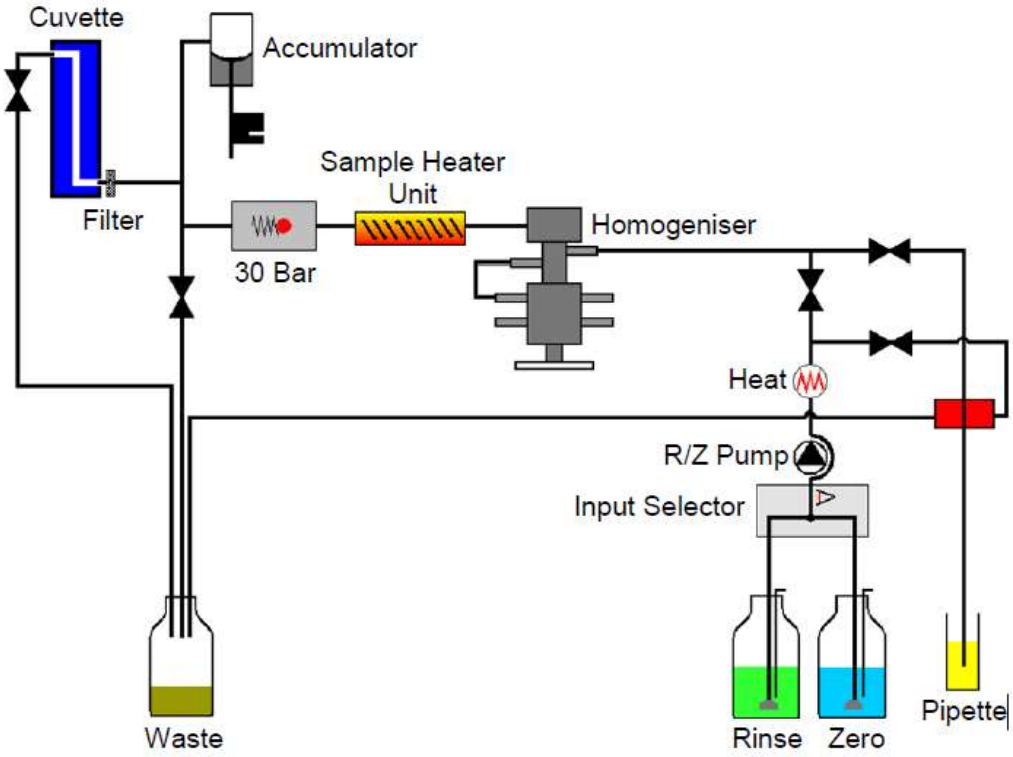


Figure 2. MilkoScan™ 7 RM.

5 ICAR evaluation criteria

The FOSS MilkoScan™ 7 RM was evaluated against the limits in ICAR protocol “Procedure 1 of Section 12 of ICAR Guidelines – Protocols for Evaluation of Milk Analyses for ICAR Approval”. Furthermore, FOSS performance was compared against the proposed limits (values in red) for FTIR instruments see Table 1 (only informative). Its accuracy was also

compared to the accuracy of the FOSS MilkoScan FT6000. MilkoScan FT6000 was already granted the ICAR grandfather exception in January 2020.

Table 1. ICAR evaluation criteria limits.

	Criteria limits					Criteria limits			
Measurand (Units)	**Fat g/100 g**	**Protein g/100 g**	**Lactose g/100 g**	**Urea mg/100 g**	**Measurand (units)**	**Fat g/100 g**	**Protein g/100 g**	**Lactose g/100 g**	**Urea mg/100 g**
Range, D*L*range — Whole	2,0 to 6,0	2,5 to 4,5	4,0 to 5,5	10,0 to 70,0	**Accuracy(1)**				
Carry-over ratio limit, *LC*	1	1	1	2	**Comparison of alternative against reference method**				
Sequence number, *NC*	20	20	20	20	Individual animal milk samples				
Minimum range, D*L*test	4	3	1,5	45	Standard deviation, *syx*	0,1	0,1	0,1	6
Linearity: ratio limit, D*e*/D*L*	0,01	0,01	0,01	0,02	Standard deviation, *syx*	0,06	0,06	0,06	6
Replicate number for linearity, *NL*	6	6	6	6	Number of individual animal milk samples, *Na*	100	100	100	100
Maximun range, D*L*test	4	4	4	100	Herd bulk milk samples'				
Intra laboratory reproducibility					Standard deviation, *syx*	0,07	0,07	0,07	4
Average standard deviation, s_{Rintra} *(Filter Instr.)*	0,02	0,02	0,02		Standard deviation, *syx*	0,05	0,05	0,05	4
Average standard deviation, s_{Rintra} *(FT Instr.)*	0,014	0,014	0,014		Number of herds, *Nh*1	5	5	5	5
Average reproducibility intra, R (Filter Instr.)	0,06	0,06	0,06		Number of herd bulk milk samples'	60	60	60	60
Average reproducibility intra R (FT)	0,04	0,04	0,04		**Accuracy (2)**				
Repeatability					**Comparison between two different instruments model**				
					Individual animal milk samples				
Average standard deviation of repeatability, s_r (Filter Instr.)	0,014	0,014	0,014		Standard deviation, *syx*	0,04	0,04	0,04	6

	Criteria limits					Criteria limits			
Measurand	**Fat**	**Protein**	**Lactose**	**Urea**	**Measurand**	**Fat**	**Protein**	**Lactose**	**Urea**
(Units)	**g/100 g**	**g/100 g**	**g/100 g**	**mg/100 g**	**(units)**	**g/100 g**	**g/100 g**	**g/100 g**	**mg/100 g**
Average standard deviation of repeatability, s_r (FT)	0,008	0,008	0,008	1,4	Number of individual animal milk samples, *Na*	100	100	100	100
Average repeatability, r (Filter Instr.)	0,04	0,04	0,04		Herd bulk milk samples'				
Average repeatability, r (FT)	0,02	0,02	0,02	3,92	Standard deviation, *syx*	0,03	0,03	0,03	4
Calibration									
Mean bias	± 0,05	± 0,05	± 0,05	± 1,2	Number of herds, *Nh1*	5	5	5	5
Slope,b	1 ± 0,05	1 ± 0,05	1 ± 0,10	1 ± 0,10	Number of herd bulk milk samples'	60	60	60	60

6 Phase I - Repeatability

The repeatability test is one of the basic and most important tests to perform during the validation study. The repeatability describes the minimum instrument variance results and it is part of intra- and interlaboratory reproducibility.

The data obtained during the stability tests performed in Phase I on a pilot sample and one instrument have been used to calculate the instrument repeatability. Each pilot sample has been analysed in 2 or 3 replicates. The tests conducted in phase one on MilkoScan™ 7 RM at Milchprüfring Bayern and Actalia provided results that fits the ICAR repeatability limit of r ≤0,04 g/100 g. The repeatability was calculated also during the accuracy test on 134 single cow milk samples, in a fat range of 2-4 g/100g and in a protein range of 2-5,8 g/100g, the repeatability values are r ≤0,02 (Table 2).

Table 2. Repeatability (r) and daily stability (R_{intra})results on pilot sample and single cow milk.

Type of Sample	N	Laboratory	Parameter	mean	r	R (intra)	Ref.	Limit
Pilot	20	mpr	Fat	2,01	0,02	0,05	Tab.A1	sr≤ 0,014 (0,008) r≤0,04 (0,02) $sR_{(intra)}$≤0,02(0,014) $R_{(intra)}$≤0,06 (0,04)
Pilot	20	mpr	Fat	3,55	0,01	0,05	Tab.A1	
Pilot	20	mpr	Fat	4,98	0,01	0,06	Tab.A1	
Pilot	20	mpr	Protein	2,61	0,01	0,04	Tab.A2	
Pilot	20	mpr	Protein	3,34	0,01	0,04	Tab.A2	
Pilot	20	mpr	Protein	4,05	0,02	0,04	Tab.A2	
Pilot	20	Actalia	Fat	1,93	0,01	0,02	Actalia report 2016	
Pilot	20	Actalia	Fat	3,90	0,02	0,02	Actalia report 2016	
Pilot	20	Actalia	Fat	5,83	0,03	0,03	Actalia report 2016	
Pilot	20	Actalia	Fat	7,50	0,04	0,04	Actalia report 2016	
Pilot	20	Actalia	Protein	2,05	0,02	0,02	Actalia report 2016	
Pilot	20	Actalia	Protein	2,91	0,01	0,02	Actalia report 2016	
Pilot	20	Actalia	Protein	3,84	0,02	0,03	Actalia report 2016	
Pilot	20	Actalia	Protein	5,76	0,02	0,02	Actalia report 2016	
Single cow	134	Actalia	Fat	3,96	0,02		Actalia report 2016	
Single cow	134	Actalia	Protein	3,47	0,02		Actalia report 2016	

Conclusion

The repeatability values obtained comply with the ICAR limit of ≤0,04. The repeatability will be further evaluated in phase II considering the robustness tests and the interlaboratory study (ILS)

7 Phase I - Intralaboratory reproducibility- Stability one instrument

Intralaboratory reproducibility ($R_{intra\ Phase\ I}$) of the pilot sample on one instrument (the same pilot sample is analysed by the same instrument at different time during the working day) and described the instrument stability well. Milchprüfring Bayern and Actalia prepared different concentration of pilot samples for fat and protein to evaluate the instrument daily reproducibility (R_{intra}). A fat range of 2-8 g/100g and a protein range of 2-5,8 g/100g were tested (Table 3 –Table A1 -Table A2).

The stability values obtained are for fat $R_{intrafat}$=0,056 g/100 g and for protein $R_{intraprotein}$=0,040 g/100 g. The ICAR limit is R_{intra}≤0,08 (sR_{intra}≤0,028).

Table 3 Instrument stability (Milchprüfring Bayern)

	Fat				Protein			
	sr	sR_{intra}	r	R_{intra}	sr	sR_{intra}	r	R_{intra}
Low	0,008	0,019	0,023	0,054	0,005	0,015	0,014	0,042
Medium	0,005	0,017	0,014	0,048	0,004	0,013	0,011	0,037
High	0,005	0,023	0,014	0,065	0,006	0,014	0,017	0,040
OverAll	0,006	0,020	0,017	0,056	0,005	0,014	0,014	0,040
Limit	0,014	0,028	0,04	0,08	0,014	0,028	0,04	0,08

Conclusion for stability

The instrment daily reproducibility (R_{intra}) complies with the ICAR limit.

8 Phase I - Carry over

ICAR protocol, ISO 8196-3/IDF 128-3 and ISO 9622/IDF 141 define the carry-over as the residual volume of the previous sample as a percentage of the total volume of the instrument cell after a single pumping sequence of a sample through the instrument cell.

Internal factors/issues affecting carry over include pump settings, flow system deficiencies and compensation factors. External factors affecting carry-over include transfer from the stirrer and pipette.

After each measurement, the analytical circuit is cleaned to the possibility of transfer of a portion of a milk sample to the successive sample to a minimum (ISO 9622/IDF 141).

The carry-over tests have been executed at Milchprüfring Bayern and at Actalia for the parameters fat and protein. One individual cow milk and distilled water have been analyzed according the following sequence: MILK - MILK -WATER -WATER * repeated twenty times for the classic components: Fat and Protein

The efficiency of this is measured by the Ratio:

$$CO = [(\Sigma \text{ (Water 1)} - \Sigma \text{ (Water 2)}) / (\Sigma \text{ (Milk 2)} - \Sigma \text{ (Water 2)})] \times 100$$

The obtained value has to be lower than 1%.

Milchprüfring Bayern calculated the carry-over for fat parameter and obtained a carryover ratio for fat high to low CO H/L= 0,14 % and for Actalia reported the carry-over ratio for fat H/L = 0,31% and carry-over for protein H/L = 0,37%.

Table 4 Carry-over results

	Fat Range g/100g	CO Fat %
Carry Over ratio H/L(mpr)	0-9,60	0,14
Carry Over ratio H/L(Actalia)	1,9-7,48	0,31
	Protein Range g/100g	CO Protein %
Carry Over ratio H/L(Actalia)	2,05-5,76	0,37
Limit CO H/L		1%

Conclusion

The CO tests performed at Milchprüfring Bayern and Actalia revealed a carry over below the limit of 1% each for fat and protein (Table 4 and Table A3)

9 Linearity

Linearity expresses the constancy of the ratio between the increase in the concentration of an analyte and the corresponding result of an alternative method. A linear alternative method can be helpful to check the calibration adjustment, too.

The ratio De=range of residuals and Dc= range of concentration was calculated according the formula:

$$r = \frac{(e_{max} - e_{min})}{(M_{max} - M_{min})} \times 100$$

Where:

e_{max} is the numerical value of the maximum residual from the regression;

e_{min} is the numerical value of the minimum residual from the regression;

M_{max} is the numerical value of the upper measured value for the concerned set of samples;

M_{min} is the numerical value of the lower measured value for the concerned set of samples

The linearity test has been executed by Milchprüfring Bayern and Actalia for fat and protein.

Milchprüfring Bayern prepared 9 samples regularly distributed over a fat range of 2,25-4,92 g/100g and of protein 2,84-4,12 g//100g. The samples have been produced by mixing cream and skimmed milk to create the fat range and mixing filtrate and retentate obtained by tangential ultrafiltration. The two sets of samples have been analyzed in two replicates in increasing and in decreasing order of fat and protein content.

The result for the linearity ratio was $r_{fat(mpr)}$= 0,96% and $r_{protein(mpr)}$= 0,76 % (Table 5 and Table A4).

Actalia used the same scheme with 11 samples in a fat range of 1,96-7,84 g/100g and 15 samples in a protein range of 0-7,8 g//100g.

The linearity resulted ratio was $r_{fat(Actalia)}$= 0,76% $r_{protein(Actalia)}$= 0,26%.

Table 5 Linearity tests results.

		mpr	Actalia	Limit
Fat	range	2,25-4,92	1,96-7,84	1%
	n	9	11	
	r=De/Dc	**0,96%**	**0,76%**	
Protein	range	2,84-4,12	0-7,8	
	n	9	15	
	r=De/Dc	**0,76%**	**0,26%**	

Conclusion

Linearity test for fat and protein, executed in two different validation centers, complies with the ICAR protocol, ISO 8196-3 and in the ISO 9622/IDF 141 where the ratio, *r*, is indicated to be equal or less than 1 %.

10 Accuracy evaluation

The accuracy test was executed with different schemes:

a. accuracy test (mpr) MilkoScan™ 7 RM vs FT 6000
b. accuracy test (Actalia) MilkoScan™ 7 RM vs chemical methods
c. calibration evaluation (11.7.1)
d. comparison between instrument measurement and calibration samples values (11.7.2)
e. accuracy results for interlaboratory study cow samples vs reference methods (11.7.3)

10.1 Accuracy test (Milchprüfring-Bayern) MilkoScan™ 7 RM vs FT 6000

Milchprüfring Bayern calculated the accuracy comparing MilkoScan™ 7 RM vs FT 6000 for the parameters fat, protein, lactose and urea. The calibration of both instruments, MilkoScan™ 7 RM and FT 6000 have been adjusted using QSE calibration materials for fat, protein, lactose and urea characterized with reference methods, Röse Gottlieb, Kjeldhal, HPLC and differential pH method, respectively. The QSE calibration materials have a range of concentration applicable for the DHI cow milk sample analyses. 657 individual cow milk sample have been analyzed in single replicate by both instruments. This experimental plan is slightly different from the ICAR protocol, which prescribes to analyze the sample in 2 or 3 replicates and to consider the mean instrument value for the comparison with the anchor (reference) value. However, in this extensive accuracy test the experimental condition are more restrictive due to the single analysis executed on each instrument (Table 6).

Table 6. Accuracy test (Milchprüfring-Bayern) MilkoScan™ 7 RM vs FT 6000.

	FT 6000	MSC 7 RM			
	Fat (Yr)	**Fat (Xc)**	**Estimates Yc(x)**	**Differences d=X-Yc**	**Residuals (e)**
N	657	657	657	657	657
Mean	4,076	4,070	4,076	-0,007	0,000
SD	0,747	0,754	0,747	0,007	0,011
Minimum	1,590	1,570	1,599	-0,029	-0,040
Maximum	7,510	7,550	7,525	0,025	0,059
D = Max-Min	5,920	5,980	5,926	0,054	0,099
Slope	0,9910				
Intercept	0,0432				

			Limit	**Result**
Mean d =	**-0,007**	à	**0,050**	positive
Sd =	**0,007**	à	**0,070**	positive
Syx	**0,011**	à	**0,040**	positive
Slope =	**0,99**	à	**0,95<slope<1,05**	positive
intercept	**0,04**			

	FT 6000	MSC 7 RM			
	Lactose (Xc)	**Lactose (Xc)**	**Estimates Yc(x)**	**Differences d=X-Yc**	**residuals (e)**
N	657	657	657	657	657
Mean	4,818	4,825	4,825	0,000	-0,006
SD	0,199	0,197	0,195	0,002	0,014
Minimum	3,890	3,920	3,928	-0,008	-0,073
Maximum	5,260	5,250	5,246	0,004	0,043
D = Max-Min	1,370	1,330	1,318	0,012	0,115
Slope	1,0093				
Intercept	-0,0515				

			Limit	**Result**
Mean d =	**0,000**	à	**0,050**	positive
Sd =	**0,002**	à	**0,070**	positive
Syx	**0,014**	à	**0,040**	positive
Slope =	**1,01**	à	**0,90<slope<1,10**	positive
intercept	**-0,05**			

	FT 6000	MSC 7 RM			
	Protein (Yr)	Protein (Xc)	Estimates Yc(x)	Differences d=X-Yc	Residuals (e)
N	657	657	657	657	657
Mean	3,442	3,436	3,448	-0,012	-0,006
SD	0,364	0,372	0,368	0,003	0,013
Minimum	2,350	2,330	2,352	-0,022	-0,141
Maximum	4,660	4,690	4,691	-0,001	0,034
D = Max-Min	2,310	2,360	2,339	0,021	0,175
Slope	0,9794				
Intercept	0,0777				

			Limit	Result
Mean d =	**-0,012**	à	**0,050**	positive
Sd =	**0,003**	à	**0,070**	positive
Syx	**0,013**	à	**0,040**	positive
Slope =	**0,98**	à	**0,95<slope<1,05**	positive
intercept	**0,08**			

	FT 6000	MSC 7 RM			
	Urea (Yr)	Urea (Xc)	Estimates Yc(x)	Differences d=X-Yc	Residuals (e)
N	657	657	657	657	657
Mean	21,416	21,037	20,891	0,146	0,525
SD	7,711	7,783	7,713	0,070	2,293
Minimum	2,100	0,000	0,043	-0,043	-8,287
Maximum	62,800	64,900	64,361	0,539	6,719
D = Max-Min	60,700	64,900	64,318	0,582	15,006
Slope	0,9469				
Intercept	1,4960				

			Limit	Result
Mean d =	**0,146**	à	**1,200**	positive
Sd =	**0,070**	à	**0,070**	positive
Syx	**2,293**	à	**6,000**	positive
Slope =	**0,95**	à	**0,90<slope<1,10**	positive
intercept	**1,50**			

The results obtained were:

For fat s $_{yxfat}$ = 0,011, for protein s $_{yxprotein}$ = 0,013, for lactose s $_{yxlactose}$= 0,014 for s $_{yxurea}$= 2,29.

10.2 Accuracy test (Actalia) MilkoScan™ 7 RM vs chemical methods

At Actalia the accuracy test was executed after the basic manufacture calibration was adjusted with Actalia SRM (Table 7) in the range of the DHI samples. The accuracy test was performed on 131 and 134 single cow milk samples for fat and protein, respectively. The samples have been tested in double with the instrument and with chemical methods. For the statistical treatment, the mean value of two replicates executed on each sample with chemical methods (Gerber acido-butyrometric -fat, Amido black-protein) and the mean value of two instrument replicates have been considered. The data provided by Actalia have been converted from g/l to g/100g.

Table 7. Accuracy test (Actalia) MilkoScan™ 7 RM vs chemical methods.

	Fat	Protein	Limit	Limit FT
n	131	134		
min	1,96	2,77		
max	5,86	4,26		
Y	3,84	3,44		
Sy	0,59	0,37		
d	0,07	0,004		
Sd	0,04	0,06		
S_{yx}	0,04	0,06	0,1	0,06
S_{yx} %	1,09	0,4		
b	0,98	0,96		
a	0,07	1,1		

The results obtained were:

For fat s_{yxfat} =0,014, for protein $s_{yxprotein}$ =0,06

Conclusion for accuracy

The accuracy test executed comparing MilkoScan™ 7 RM with previous generation of instrument FT 6000 resulted in a standard deviation of accuracy s_{yx} below the proposed ICAR limits for all the 4 parameters.

The accuracy test for fat and protein evaluated vs the chemical methods Gerber and Amido Black resulted in the ICAR limit for FT instruments.

For this first part of MilkoScan™ 7 RM accuracy evaluation the results are positive. The overall accuracy evaluation is reported in section 11.

11 Interlaboratory study

An interlaboratory study (ILS) was organised in order to validate some of the criteria obtained in phase I in one single laboratory.

Qlip organised the ILS according to ISO 5725-1, ISO 5725-2 and IDF Bulletin 453/2012. It shall be noted that Qlip is accredited with ISO 17043 and for this the ILS statistical treatment is in the scope of this accreditation.

11.1 Participant laboratories

Ten laboratories from eight countries participated in the validation study with a total number of 12 MilkoScan™ 7 RM. The name of the laboratories is presented in alphabetic order in Table 8.

Table 8. Participant laboratories in the interlaboratory study.

Laboratory Name	
Ass. Regionale Allevatori Puglia	Italy
Ass. Regionale Allevatori lombardia	Italy

Coprinsem	Chile
Eastern Laboratory Services	USA
Federeazione Latterie Alto Adige Soc. Agr. Coop.	Italy
LIGAL	Spein
Milchprüfring Bayern	Germany
National Milk Laboratories	United Kingdom
Qlip	Netherlands

11.2 Interlaboratory Study (ILS) design

The participant laboratories have been provided with the following material (per instrument):

- 1 vial of 60 ml containing water for temperature control upon arrival (code: water);
- 1 vial of 60 ml containing blank solution (FOSS Zero Liquid), (code: blank);
- 1 vial of 37 ml for spectrum standardization (FOSS FTIR Equalizer), (code: Equalizer)
- Eight single cow milk and 2 bulk milk samples were split in double blind each and this way a set of in total twenty vials was created. 20 vials of 60 ml containing preserved raw milk for measurements with the MSC 7 (code: MSC 1 to MSC 2, see Table 9).
- Note: samples were preserved with 0,02% Na- azide + 0,005% bronopol + 0,0004% patent blue (end concentration in the milk).
- 2 Qlip sets consisting of 7 samples each for FAT calibration, (code: F 0 to F 6)
- 2 Qlip sets consisting of 5 samples each for PROTEIN calibration, (code: P 1 to P 5)
- 2 Qlip sets consisting of 5 samples each for LACTOSE calibration, (code: L 1 to L 5)
- 2 Qlip sets consisting of 6 samples each for UREA calibration, (code: U 1 to U 6)

- Note: Qlip calibration samples are preserved with 0,03% Na- azide + 0,0004% eosin (end concentration in the milk).

The sample concentration (11.7.1) covered the analytical application for ICAR interest

Table 9. Sample identification and type of milk.

Milk		Sample ID	
Individual cow milk	1	MSC 2	MSC 12
	2	MSC 3	MSC 15
	3	MSC 6	MSC 13
	4	MSC 9	MSC 16
	5	MSC 4	MSC 20
	6	MSC 7	MSC 18
	7	MSC 10	MSC 17
	8	MSC 8	MSC 19
Herd	9	MSC 1	MSC 11
	10	MSC 5	MSC 14

11.3 Transport condition and delivery time

All samples were delivered within a maximum of 72 hours with two exceptions. The samples temperature at arrival was between 1°C and 6,9°C. The sample quality reported by all the

laboratories was good and all the samples delivered have been analysed. The two laboratories that received the parcel on 30 November and on 2 December did not show any outlier result during the statistical evaluation.

11.4 ILS samples Homogenity

Each of the 10 MSC samples was tested in two replicates

The sample homogeneity was checked considering the fat concentration.

According to ISO 13528 the variance between vials (se) should not be bigger than 0,3*sR of the method used to check the homogeneity.

ISO 9622/IDF 141:2013 report sR=0,04. It means that the variance between the vials should not be bigger than se ≤ 0,013. All the samples tested showed a variance between vials (se) lower than the calculated limit Table 10.

Table 10 Sample homogeneity between vials

Limits ISO 13528
se≤ 0,3*sR

	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
	MSC 2 MSC 12	MSC 3 MSC 15	MSC 6 MCS 13	MSC 9 MSC 16	MSC 4 MSC 20	MSC 7 MSC 18	MSC 10 MSC 17	MSC 8 MCS 19	MSC 1 MSC 11	MSC 5 MSC 14
level	**2,27**	**4,05**	**4,85**	**3,99**	**2,48**	**3,40**	**3,07**	**5,39**	**4,95**	**4,27**
sr	0,005	0,004	0,004	0,004	0,005	0,003	0,005	0,005	0,003	0,003
r	0,014	0,013	0,013	0,011	0,014	0,009	0,014	0,016	0,009	0,009
sr%	0,22%	0,11%	0,09%	0,10%	0,20%	0,09%	0,16%	0,10%	0,06%	0,07%
sd	0,004	0,006	0,003	0,004	0,005	0,002	0,004	0,003	0,003	0,005
se	0,002	0,005	0,001	0,003	0,003	0,000	0,001	0,000	0,003	0,004
sR ISO 9622	0,04	0,04	0,04	0,04	0,04	0,04	0,04	0,04	0,04	0,04
Limit se	0,013	0,013	0,013	0,013	0,013	0,013	0,013	0,013	0,013	0,013

11.5 ILS execution

All the participant laboratories received detailed instructions on how to treat the samples and how to prepare the instrument according specific manufactures instructions in order to obtain the best standardised conditions. All the tests have been executed on 29 November 2019 with four exceptions, two on 28 November and two on 2 December.

A dedicated job was created to analyse the ICAR ILS samples. It was requested to set slope=1 and Bias=0 for each parameter.

After the instrument was standardisation with FOSS FT Equalizer the blank samples and the ILS samples have been analysed in duplicates.

After the results registration it was requested to measure the Qlip calibration samples in calibration mode. The slope and bias proposed by the instrument have been reported in the result sheet and used to recalculate the results. The second set of Qlip calibration samples have been analysed to verify the calibration.

11.6 Laboratory results and statistical treatment

For each parameter the data have been statistically treated as follows :

f. raw data with instrument set with slope=1 and bias =0.
For the statistical treatment the first replicate of each vial and the first replicate of its double blind were considered.

g. outliers: Cochran and Grubbs tests and deletion of the identified outlier. Straggler data have not been removed.

h. data recalculated with instrument slope and bias obtained during the calibration adjustment Table B1

i. outlier Cochran and Grubbs tests and deletion of the identified outlier. Straggler data have not been removed.

j. calibration evaluation (11.7.1)

k. comparison of the instrument measurement results with calibration sample values (11.7.2)

l. accuracy results for interlaboratory study cow samples vs reference methods (11.7.3)

m. precision calculation (11.7.4).

11.7 Calibration evaluation

During the interlaboratory study the ILS samples were analysed using 12 instruments with the basic manufacture calibration slope=1 and bias=0. In a second step the laboratories adjusted the basic calibration using Qlip SRM for the parameters fat (g/100g), protein (g/100g), lactose (g/100g) and urea (mg/dl). The slope obtained has been evaluated according to the ICAR limits being 1+/- 0,05 for fat and protein , 1+/- 0,10 for lactose and urea. Furthermore, the interval of confidence was calculated for each instrument.

The mean bias, defined as mean of the difference between the instrument result and calibration samples value, were evaluated to be in the ICAR limit of
Mean $_{\text{biasFat-Protein-Lactose}}$ =+/- 0,05 for fat protein lactose and Mean $\text{bias}_{\text{Urea}}$ =+/-1,2

Standard deviation of difference is reported as informative.

The criteria illustrated above are summarized in Table 11.

Table 11. Calibration data.

	Slope			Intercept				
Fat	-	Lowest 95%	Highest 95%		Lowest 95%	Highest 95%	Mean bias	sd
ICAR limit	**1+ 0,05**						**+/-0,05**	
L1F	1,03	1,029	1,041	- 0,000	- 0,025	0,019	0,004	0,016
L2F	0,98	0,977	0,989	0,110	0,092	0,132	0,002	0,012
L3F	1,01	1,003	1,016	0,050	0,031	0,075	0,001	0,011
L5F	0,98	0,974	0,995	0,163	0,122	0,197	0,001	0,019
L7F	1,03	1,020	1,037	0,006	- 0,025	0,037	0,003	0,021
L8F	1,03	1,019	1,034	0,019	- 0,004	0,047	0,006	0,014
L10F	1,03	1,018	1,031	0,017	- 0,005	0,043	0,001	0,014
L13F	1,00	0,992	1,003	0,070	0,043	0,083	0,005	0,013
L14F	1,01	1,003	1,019	0,004	- 0,024	0,033	0,002	0,015
L15F	1,02	1,009	1,024	0,166	0,140	0,191	0,002	0,011
L16F	0,99	0,983	0,999	0,169	0,138	0,196	0,001	0,015
L17F	1,05	1,039	1,053	0,014	- 0,011	0,036	0,039	0,031

SRM	F1	F2	F3	F4	F5	F6	F7
Fat RM	0,06	1,05	2,05	3,03	4,03	5	5,98

	Slope			Intercept				
Lactose	-	Lowest 95%	Highest 95%		Lowest 95%	Highest 95%	**Mean bias**	**sd**
ICAR limit	**1+ 0,10**						**+/-0,05**	
L1L	1,007	0,998	1,017	- 0,173	- 0,220	- 0,134	0,001	0,007
L2L	1,006	1,000	1,015	- 0,350	- 0,392	- 0,321	0,000	0,005
L3L	1,027	1,022	1,031	- 0,330	- 0,345	- 0,305	0,004	0,007
L5L	0,988	0,981	0,996	0,020	- 0,015	0,051	0,002	0,004
L7L	1,008	1,002	1,011	0,017	0,006	0,043	0,002	0,003
L8L	0,996	0,993	0,998	0,031	0,021	0,045	0,001	0,004
L10L	1,006	0,996	1,012	0,002	- 0,026	0,043	0,003	0,007
L13L	0,991	0,988	0,993	0,020	0,007	0,029	0,005	0,005
L14L	1,003	0,993	1,012	- 0,157	- 0,200	- 0,113	0,000	0,022
L15L	1,007	0,995	1,021	0,001	- 0,062	0,054	0,012	0,008
L16L	1,000	0,989	1,011	0,001	- 0,046	0,052	0,001	0,007
L17L	1,010	0,996	1,022	- 0,205	- 0,260	- 0,143	0,033	0,004

SRM Lactose	L 1	L 2	L 3	L 4	L 5
RM	2,92	3,64	4,34	5,01	5,75

	Slope			Intercept				
Protein	-	Lowest 95%	Highest 95%		Lowest 95%	Highest 95%	**Mean bias**	**sd**
ICAR limit	**1+ 0,05**						**+/-0,05**	
L1P	1,01	0,996	1,018	- 0,016	- 0,058	0,026	0,003	0,008
L2P	1,00	0,993	1,008	- 0,010	- 0,039	0,019	0,001	0,009
L3P	1,03	1,016	1,035	- 0,050	- 0,089	- 0,018	0,002	0,008
L5P	1,00	0,993	1,005	- 0,051	- 0,070	- 0,021	0,006	0,009
L7P	1,01	1,003	1,018	- 0,010	- 0,043	0,014	0,014	0,007
L8P	1,01	1,001	1,018	- 0,005	- 0,041	0,024	0,004	0,007
L10P	1,00	0,992	1,016	0,014	- 0,031	0,058	0,002	0,009
L13P	1,01	1,002	1,024	- 0,040	- 0,082	0,004	0,004	0,009
L14P	1,00	0,984	1,006	0,015	- 0,037	0,046	0,012	0,006
L15P	1,03	1,017	1,034	- 0,060	- 0,096	- 0,030	0,010	0,007
L16P	1,02	1,005	1,030	- 0,062	- 0,112	- 0,015	0,002	0,006
L17P	1,01	1,002	1,026	- 0,020	- 0,072	0,020	0,005	0,007
SRM Protein	P 1	P 2	P 3	P 4	P 5			
RM	1,89	2,78	3,65	4,54	5,4			

	Slope			Intercept				
Urea	-	Lowest 95%	Highest 95%		Lowest 95%	Highest 95%	**Mean bias**	**sd**
ICAR limit	**1+ 0,10**						**`+/-1,2**	
L1U	1,07	1,034	1,113	-24,970	-27,668	-22,324	0,1	0,808
L2U	1,04	1,016	1,059	-8,500	-9,760	-7,437	1,0	0,602
L3U	1,11	1,078	1,152	-15,800	-17,930	-13,731	0,0	0,592
L5U	1,09	1,086	1,125	-24,990	-27,095	-24,463	2,1	1,131
L7U	1,08	1,052	1,116	-14,622	-16,482	-12,809	1,0	0,425
L8U	1,06	1,032	1,103	-21,996	-10,846	-7,053	0,4	0,618
L10U	1,10	1,075	1,128	-25,220	-27,066	-23,590	0,9	1,016
L13U	1,09	1,052	1,132	-18,000	-20,848	-16,045	0,7	0,516
L14U	1,14	1,125	1,148	-8,760	-9,369	-8,197	0,7	0,840
L15U	1,08	1,061	1,107	-12,350	-13,614	-11,045	0,8	0,351
L16U	1,07	1,032	1,103	-8,980	-10,846	-7,053	0,2	0,430
L17U	1,06	1,031	1,086	-21,390	-23,192	-19,658	0,9	0,930
SRM Urea	U1	U2	U3	U4	U5	U6		
RM	18	24	33	49	63	78		

Conclusion

The calibration evaluation of all the 12 instruments did not show values out of the established limits with the exception of parameter urea instrument L5U mean $bias_{Urea}$=2,1.mg/dl. The laboratory did not reported any information to explain this results. Considering the successive tests it is clear that the instrument was not correctly adjusted for the high concentration U6=78 mg/dl (Tab 12). Despite that the overall instrument accuracy resulted optimal in urea the validation range 13-46 mg/dl

11.7.1 Comparison of the instrument measurement results with calibration sample values

In order to validate the measurement procedure the difference between the certified and measured value with its uncertainty were calculated and evaluated.

The comparison of $\Delta_m \leq U_\Delta$ where:

Δ_m = the absolute difference between the mean measured value and calibration sample value

U_Δ = is the uncertainty of Δ_m

$$u_\Delta = \sqrt{u^2_m + u^2_{RM}}$$

Defining:

U_Δ is the expanded uncertainty of u_Δ

$U_\Delta = u_\Delta * 2$

$u^2_\Delta = u^2_m + u^2_{SRM}$

u^2_m= uncertainty of measurement result

u^2_m was considered the standard deviation of interlaboratory study (sRILS) obtained for each parameter $um = sR_{ILS}$

Table 12. Comparison between instruments results and calibration sample value ($\Delta_m \leq U_\Delta$).

Fat	**F1**	**F2**	**F3**	**F4**	**F5**	**F6**	**F7**
RM	0,06	1,05	2,05	3,03	4,03	5,00	5,98
UΔ	**0,048**	**0,048**	**0,048**	**0,048**	**0,048**	**0,048**	**0,048**
L1F	0,030	0,010	0,020	0,005	0,010	0,010	0,000
L2F	0,020	0,010	0,020	0,000	0,005	0,000	0,000
L3F	0,015	0,015	0,015	0,000	0,000	0,000	0,005
L5F	0,000	0,020	0,000	0,020	0,020	0,000	0,030
L7F	0,030	0,020	0,030	0,015	0,000	0,000	0,015
L8F	0,025	0,010	0,020	0,005	0,010	0,010	0,010
L10F	0,020	0,010	0,020	0,000	0,000	0,000	0,015
L13F	0,030	0,000	0,010	0,010	0,010	0,000	0,005
L14F	0,020	0,015	0,020	0,000	0,005	0,010	0,015
L15F	0,010	0,010	0,010	0,020	0,010	0,000	0,005
L16F	0,015	0,010	0,010	0,020	0,010	0,005	0,020
L17F	0,020	0,000	0,010	0,040	0,055	0,065	0,085

U ref material	0,018
u ref material for two replicates	0,0064
sR ILS_{Fat}	0,023
UΔ	**0,048**

Lactose	**L1**	**L2**	**L3**	**L4**	**L5**
RM	2,92	3,64	4,34	5,01	5,75
UΔ	**0,047**	**0,047**	**0,047**	**0,047**	**0,047**
L1L	0,010	0,005	0,005	0,005	0,000
L2L	0,005	0,005	0,000	0,005	0,005
L3L	0,015	0,000	0,000	0,000	0,005
L5L	0,000	0,010	0,000	0,000	0,000
L7L	0,005	0,000	0,005	0,000	0,000
L8L	0,005	0,005	0,000	0,005	0,000
L10L	0,000	0,010	0,010	0,000	0,005
L13L	0,005	0,000	0,000	0,010	0,010
L14L	0,007	0,006	0,004	0,002	0,005
L15L	0,000	0,020	0,020	0,010	0,010
L16L	0,010	0,005	0,005	0,005	0,000
L17L	0,030	0,040	0,030	0,030	0,035

U ref material	0,03
U ref material for two replicates	0,0106
sR $ILS_{Lactose}$	0,021
UΔ	**0,047**

Protein	P1	P2	P3	P4	P5
RM	1,89	2,78	3,65	4,54	5,40
UΔ	**0,047**	**0,047**	**0,047**	**0,047**	**0,047**
L1P	0,005	0,005	0,005	0,015	0,005
L2P	0,010	0,005	0,000	0,010	0,010
L3P	0,010	0,000	0,000	0,010	0,010
L5P	0,010	0,010	0,010	0,010	0,010
L7P	0,020	0,005	0,010	0,020	0,015
L8P	0,000	0,015	0,005	0,000	0,000
L10P	0,000	0,010	0,000	0,015	0,005
L13P	0,000	0,010	0,010	0,010	0,010
L14P	0,010	0,020	0,010	0,005	0,015
L15P	0,010	0,000	0,010	0,020	0,010
L16P	0,005	0,000	0,005	0,010	0,000
L17P	0,010	0,000	0,000	0,015	0,000

U ref material	0,025
u ref material for two replicates	0,0088
sR $ILS_{Protein}$	0,022
UΔ	**0,047**

Urea	U1	U2	U3	U4	U5	U6
RM	18	24	33	49	63	78
UΔ	**3,7**	**3,7**	**3,7**	**3,7**	**3,7**	**3,7**
L1U	0,4	1,4	0,1	0,9	0,4	0,8
L2U	1,9	1,1	1,1	1,1	0,3	0,3
L3U	0,4	0,1	0,4	0,4	0,2	1,2
L5U	2,1	1,5	1,4	0,7	2,7	3,9
L7U	1,1	0,8	0,6	1,8	0,7	0,9
L8U	0,5	0,9	0,6	0,8	0,2	0,9
L10U	1,6	0,3	1,7	2,2	0,3	0,1
L13U	0,5	1,5	1,0	0,5	0,0	0,5
L14U	2,3	0,9	0,2	0,1	0,1	0,6
L15U	1,5	0,8	0,9	0,9	0,5	0,5
L16U	0,1	0,8	0,0	0,1	0,4	0,6
L17U	0,4	0,5	1,5	0,9	1,4	2,1

U ref material	2
U ref material for two replicates	0,7
sR ILS_{Urea}	1,7
UΔ	**3,7**

Conclusion

The comparison of the instruments measurement results with calibration samples values demonstrated that the instruments were correctly calibrated with the exception of instrument L17F for fat calibration samples F5,F6 and F7 and instrument L5U for urea calibration sample U6 that showed a value being slightly bigger than the calculated limit.

The instruments measuring procedure of calibration samples is positive.

11.7.2 Accuracy results for interlaboratory study cow samples vs reference methods

The ILS results obtained with the manufacture basic calibration have been recalculated using the slope and bias proposed by the instruments during the calibration adjustment. The recalculated instrumental data of ILS samples (1-10) have been compared with the results of reference methods Röse Gottlieb, Kjeldahl, HPLC and differential pH for fat, protein, lactose and urea, respectively.

The table 13 reports the results for the mean bias$_{(before)}$ obtained with slope=1 and bias=0 (instrument results- reference method results) and mean bias$_{(after)}$ (instrument results recalculated - reference method results), standard deviation of accuracy (syx $_{after}$) obtained considering the recalculated results and the reference method results. The mean bias is calculated per lab on the ILS samples, before and after recalculation (Table 13). Standard deviation of accuracy (s_{xy}) is evaluated on the recalculated results.

Table 13. Accuracy evaluation on ILS cow samples.

Accuracy evaluation with vallidation samples (ILS)

Mean bias calculated per lab for the ILS samples, before and after recalculation

Fat	**mean bias**		**syx**
	before	after	after
ICAR limit			**0,1**
ICAR limit FT			**0,06**
L1F	-0,12	0,00	0,04
L2F	-0,03	0,01	0,03
L3F	-0,08	0,01	0,03
L5F	-0,09	0,01	0,04
L7F	-0,11	0,00	0,04
L8F	-0,11	0,01	0,04
L10F	-0,11	0,00	0,04
L13F	-0,04	0,02	0,03
L14F	-0,04	0,01	0,04
L15F	-0,21	0,02	0,02
L16F	-0,13	0,00	0,02
L17F	-0,13	0,06	0,05

Protein	**mean bias**		**syx**
	before	after	after
ICAR limit			**0,1**
ICAR limit FT			**0,06**
L1P	-0,03	-0,02	0,03
L2P	0,02	0,01	0,03
L3P	-0,04	0,01	0,04
L5P	0,06	0,01	0,05
L7P	-0,04	-0,02	0,02
L8P	-0,04	-0,02	0,02
L10P	-0,04	-0,01	0,02
L13P	-0,01	0,00	0,03
L14P	0,01	0,00	0,03
L15P	-0,04	0,00	0,06
L16P	0,00	0,00	0,05
L17P	-0,04	-0,01	0,03

ILS samples	**1**	**2**	**3**	**4**	**5**
Röse Gottlieb	2,31	4	4,83	3,99	2,53
ILS samples	**6**	**7**	**8**	**9**	**10**
Röse Gottlieb	3,38	3,08	5,39	4,94	4,27

ILS samples	**1**	**2**	**3**	**4**	**5**
Kjeldhal	3,25	4,55	3,95	4,42	3,15
ILS samples	**6**	**7**	**8**	**9**	**10**
Kjeldhal	3,10	3,46	4,77	3,76	3,57

Lactose	**mean bias**		**syx**
	before	after	after
ICAR limit			**0,1**
ICAR limit FT			**0,06**
L1L	0,12	-0,02	0,04
L2L	0,27	-0,05	0,06
L3L	0,16	-0,05	0,05
L5L	0,03	-0,01	0,06
L7L	-0,07	-0,02	0,05
L8L	-0,03	-0,02	0,05
L10L	-0,04	-0,01	0,05
L13L	0,01	-0,01	0,05
L14L	0,13	0,24	0,04
L15L	-0,06	-0,03	0,06
L16L	-0,02	-0,02	0,06

Urea	**mean bias**		**syx**
	before	after	after
ICAR limit			**6**
L1U	20,14	-1,62	4,89
L2U	7,35	0,01	4,08
L3U	9,99	-1,95	3,83
L5U	21,41	0,58	4,35
L7U	10,36	-1,39	2,94
L8U	18,49	-0,83	4,87
L10U	19,50	-1,39	4,52
L13U	14,25	-0,24	3,94
L14U	3,31	-1,74	2,98
L15U	9,52	0,00	3,19
L16U	6,85	-0,05	2,62

Lactose	mean bias		syx
L17L	0,10	-0,05	0,04

Urea	mean bias		syx
L17U	16,90	-2,12	4,40

ILS samples	1	2	3	4	5
HPLC	4,93	4,63	4,45	4,54	4,69
ILS samples	6	7	8	9	10
HPLC	4,61	4,54	3,91	4,54	4,57

ILS samples	1	2	3	4	5
Diff.pH	27	31	46	29	25
ILS samples	6	7	8	9	10
Diff.pH	15	13	19	17	17

Conclusion

All twelve instruments that participated in the interlaboratory study for the parameters fat, protein, lactose and urea, after the calibration adjustment, generated data that compared with reference chemical methods produced mean bias and standard deviation of accuracy values in the range of the established ICAR limits. The overall evaluation of MilkoScan™ 7 RM accuracy, including section 10, is positive.

11.7.3 Precision calculation

The precision calculation, specifically repeatability and interlaboratory reproducibility, was done according to ISO 5725-2.

In tables B1 fat- B2 protein-B3 lactose,B4 urea are reported, for each instrument, the data obtained with slope=1 and bias=0 before the calibration adjustment and the re-calculated values with the slope and bias proposed. The identified outliers for Cochran and Grubbs are identified in yellow (Cochran) , red (Grubbs), and orange (straggler). The repeatability and reproducibility criteria have been calculated, after outlier deletion, before and after the calibration.

The precision of each sample and the overall precision for the four parameters is reported in Table 14.

For the parameter fat laboratory L5F was an outlier for Cochran test on samples 7 and 9 before and after the data re-calculation. Repeatability $r_{\text{fat before}}$ = 0,023 g/100g and reproducibility $_{\text{before}}$ =0,157 g/100g before the calibration adjustment and $r_{\text{fat after}}$ = 0,023 g/100g and reproducibility $R_{\text{fat after}}$ = 0,064 g/100g after the calibration.

For the parameter protein no outlier for Cochran and Grubs tests have been identified

Repeatability $r_{\text{protein before}}$ = 0,023 g/100g and reproducibility $R_{\text{protein before}}$ = 0,157 g/100g before the calibration adjustment and $r_{\text{protein after}}$ = 0,023 g/100g and reproducibility $R_{\text{protein after}}$=0,064 g/100g after the calibration

For the parameter lactose instrument L14L was outlier for Grubbs test for all the samples after the calibration adjustment

Repeatability $r_{\text{lactose before}}$ = 0,018 g/100g and reproducibility $R_{\text{lactose before}}$ = 0,305 g/100g before the calibration adjustment and $r_{\text{lactose after}}$ = 0,018 g/100g and reproducibility $R_{\text{lactose after}}$ = 0,060 g/100g after the calibration.

For the parameter urea, no outlier for Cochran and Grubs tests have been identified.

Repeatability r $_{urea\ before}$ = 2,3 mg/dl and reproducibility R $_{urea\ before}$ = 17,5 mg/dl before the calibration adjustment and r $_{urea\ after}$ = 2,5 mg/dl and reproducibility R $_{urea\ after}$ = 4,7 mg/dl after the calibration adjustment.

Tab 14. Repeatability (r) and Reproducibility (R).

Cochran outlier removed data obtained with slope=1 and Bias=0 before calibration)
Cochran outlier removed data obtained with slope and Bias after calibration)
Repeatability blind duplicates
Reproducibility, R

Fat	Before calibration		After calibration	
	r	R	r	R
ICAR Limit	**0,04**		**0,04**	
ICAR Limit	**0,02**		**0,02**	
Limit ISO 9622	**0,04**	**0,11**	**0,04**	**0,11**
SAMPLE 1	0,03	0,14	0,03	0,04
SAMPLE 2	0,03	0,16	0,03	0,09
SAMPLE 3	0,03	0,16	0,03	0,07
SAMPLE 4	0,02	0,17	0,02	0,06
SAMPLE 5	0,03	0,13	0,03	0,05
SAMPLE 6	0,02	0,12	0,02	0,06
SAMPLE 7	0,01	0,12	0,01	0,05
SAMPLE 8	0,02	0,19	0,03	0,09
SAMPLE 9	0,02	0,19	0,02	0,08
SAMPLE 10	0,02	0,17	0,02	0,05
Overall	0,02	0,16	0,02	0,06

Protein	Before calibration		After calibration	
	r	R	r	R
ICAR Limit	**0,04**		**0,04**	
ICAR Limit	**0,02**		**0,02**	
Limit ISO 9622	**0,04**	**0,11**	**0,04**	**0,11**
SAMPLE 1	0,02	0,08	0,02	0,07
SAMPLE 2	0,01	0,10	0,01	0,04
SAMPLE 3	0,02	0,10	0,02	0,06
SAMPLE 4	0,01	0,12	0,01	0,04
SAMPLE 5	0,03	0,10	0,03	0,06
SAMPLE 6	0,02	0,09	0,02	0,05
SAMPLE 7	0,02	0,11	0,02	0,06
SAMPLE 8	0,03	0,16	0,03	0,11
SAMPLE 9	0,04	0,11	0,04	0,05
SAMPLE 10	0,01	0,09	0,01	0,03
Overall	0,02	0,11	0,02	0,06

	Before calibration		After calibration	
Lactose	r	R	r	R
ICAR Limit	**0,04**		**0,04**	
ICAR Limit	**0,02**		**0,02**	
Limit ISO 9622	**0,04**	**0,11**	**0,04**	**0,11**
SAMPLE 1	0,02	0,31	0,02	0,05
SAMPLE 2	0,01	0,34	0,01	0,05
SAMPLE 3	0,02	0,30	0,02	0,05
SAMPLE 4	0,02	0,32	0,02	0,04
SAMPLE 5	0,01	0,31	0,01	0,06
SAMPLE 6	0,01	0,28	0,01	0,07
SAMPLE 7	0,01	0,31	0,01	0,06
SAMPLE 8	0,01	0,26	0,01	0,11
SAMPLE 9	0,03	0,30	0,03	0,05
SAMPLE 10	0,01	0,31	0,01	0,04
Overall	0,02	0,31	0,02	0,06

Urea	Before calibration		After calibration	
	r	R	r	R
ICAR Limit				
ICAR Limit	**4**		**4**	
Limit ISO 8196-3	**4**		**4**	
SAMPLE 1	2,3	14,3	2,5	5,5
SAMPLE 2	2,1	17,5	2,4	3,9
SAMPLE 3	2,3	19,6	2,5	4,1
SAMPLE 4	3,1	17,7	3,4	3,4
SAMPLE 5	1,9	13,9	2,1	6,0
SAMPLE 6	1,4	15,3	1,5	3,4
SAMPLE 7	2,2	15,5	2,5	3,6
SAMPLE 8	2,2	22,0	2,4	6,4
SAMPLE 9	2,8	18,5	3,0	5,5
SAMPLE 10	2,2	17,2	2,4	3,7
Overall	2,3	17,5	2,5	4,7

Conclusion

The results show that the instrument repeatability values are lower than the established ICAR limits r=0,04 and is also equal to the proposed limit for FT instrument r=0,02 g/100g for fat protein and lactose.

The reproducibility obtained with the slope=1 and bias=0 shows that the instruments need to be calibrated as it is recommended by the quality assurance procedures. The reproducibility values obtained adjusting the instruments with the same calibration material is R=0,06 g/100g and thus lower than ISO 9622/IDF 141 limit of R $_{Limit}$ = 0,11 g/100g.

12 Robustness

Milchprüfring Bayern followed the robustness test for MilkoScan™ 7 RM from March 2018 to March 2019.

Twelve MilkoScan™ 7 RM are installed at Milchprüfring Bayern and they analyse on average 5000 samples/day on each instrument. This situation, with several instruments working in parallel and a big number of samples analysed provided a great number of data and information. The instrument maintenance is planned each three months (each 450-500 10^3 samples) and the manufacture reccomends to proceed with the spectrum standardization each month.The instruments showed to be very stable and basically it is not necessary, to standardise the spectrum between two maintenance visits.

At Milchprüfring Bayern each day a new pilot sample is prepared. The assigned value is determined after all the instruments are checked with calibration samples.

When the Dairy Herd Improvement (DHI) single cow milk samples are analyzed, two consecutive pilot samples are analyzed each 45 samples.

We have extracted the results from 4 working days. The 12 instruments analyzed a mean of 48696 DHI samples/day and 2371 pilot samples/day.

Considering a tolerance for fat parameter of +/- 0,05 g/100g with a 5% of probability that the samples are out of tolerance, only 1,55 % of pilot samples actually exceeded (analysed in single replicate) exceeded the fixed belt and were out of tolerance without any routine interruption (Table 15).

Table 15. Routine stability test.

	17/12/2019	**19/12/2019**	**14/01/2020**	**16/01/2020**	**Overall**
n DHI samples	53833	48618	47051	46374	48969
n pilot	2588	2329	2298	2269	2371
n samples out of tolerance	47	59	21	21	
n samples out of tolerance %	1,82%	2,53%	0,91%	0,93%	1,55%
n instruments	12	12	12	10	
tolerance fat	+/- 0,05				

The standard deviation of intralaboratory reproducibility (sRintra) was calculated considering the pilot repeatability (sr) and the pilot standard deviation between instruments (sL) of four working routine day.

Table 16. Calculation of standard deviation of intralaboratory reproducibility.

		17/12/ 2019	19/12/ 2019	14/01/ 2020	16/01/ 2020	SL	SRintra	Overall	SRintra Limit	SRintra Limit
Fat [%]	Pilot Mean	4,282	4,250	4,280	4,201	0,000	0,014	4,253	0,028	0.014
	sr	0,0121	0,0121	0,0131	0,0197			0,014		
	sr%	0,28%	0,28%	0,31%	0,47%			0,003		
	sd	0,0077	0,0047	0,0086	0,0046			0,007		
Protein [%]	Pilot Mean	3,574	3,570	3,585	3,587	0,004	0,009	3,579	0,028	0.014
	sr	0,0072	0,0075	0,0095	0,0074			0,008		
	sr%	0,20%	0,21%	0,26%	0,21%			0,002		
	sd	0,0056	0,0037	0,0080	0,0057			0,006		
Lactose [%]	Pilot Mean	4,758	4,766	4,780	4,767	0,004	0,009	4,768	0,028	0.014
	sr	0,0083	0,0085	0,0091	0,0084			0,009		
	sr%	0,17%	0,18%	0,19%	0,18%			0,002		
	sd	0,0056	0,0079	0,0040	0,0042			0,006		
Urea [mg/dl]	Pilot Mean	23,781	23,845	23,712	23,581	0,000	1,4	23,730		
	sr	1,2721	1,3551	1,6993	1,3271			1,4		
	sr%	5,35%	5,68%	7,17%	5,63%			0,060		
	sd	0,0536	0,0547	0,0543	0,0567			0,055		

Note: The values in red are going to be approved in the future.

The results obtained are:

SR_{intra} Fat =0,014 g/100g	R_{intra} Fat =0,04 g/100g	Limit= 0,08 (0,04) g/100g
sR_{intra} Protein =0,009 g/100g	R_{intra} Prot =0,02 g/100g	Limit= 0,08 (0,04) g/100g
sR_{intra} Lactose =0,009 g/100g	R_{intra} Lact =0,02 g/100g	Limit= 0,08 (0,04) g/100g
sR_{intra} Urea =1,4 mg/dl	R_{intra} Urea =3,92 mg/dl	Limit= not specified

Conclusion robustness

The robustness test conducted at Milchprüfring Bayern on the MilkoScan™ 7 RM instruments showed an excellent stability and precision.

The extensive tests showed that the standard deviation of intralaboratory reproducibility was always within the limits specified by ICAR and ISO 8196-3/IDF 128-3. To be noted that for all the parameters tested the values of standard deviation of intralaboratory reproducibility (sR_{intra}) are very close to the standard deviation of repeatability limit (sr =0,014 g/100g). For urea sR_{intra} = 1,4 that is the standard deviation of repeatability (sr) limit. These results assure a very small laboratory uncertainty.

13 Conclusion

The data provided by Milchprüfring Bayern, the Actalia report and results of an interlaboratory study (ILS) organized by Qlip provided robust evidence of the instrument's

performance. The instrument performance obtained in the preliminary phase I have been confirmed and actually improved in phase II.

The instrument complies with all criteria limits defined in the ICAR “Procedure 1 of Section 12 of ICAR Guidelines – Protocols for Evaluation of Milk Analyses for ICAR Approval” aligned with the ISO 8196-3:2009 – Milk — Definition and evaluation of the overall accuracy of alternative methods of milk analysis — Part 3: Protocol for the evaluation and validation of alternative quantitative methods of milk analysis for all the criteria tested and ISO 9622/IDF 141.

ICAR certifies the performance of MilkoScan™ 7 RM based on above described studies and results thereof. All studies were executed by independent and accredited laboratories and besides test of individual laboratories an extensive international accredited validation study was performed.

14 Acknowledgment

ICAR thanks the laboratories Milchprüfring Bayern (DE) and QLIP (NL) for the qualified work done, Actalia (FR) for providing national validation reports, and all the laboratories that participated in the interlaboratory study.

15 Reference documents

- Protocol for the Evaluation of Milk Analysers for ICAR Approval: 2019 https://www.icar.org/index.php/icar-recording-guidelines/
- ISO 9622 IDF 141: 2013 – Milk and liquid milk products — Guidelines for the application of mid-infrared spectrometry
- ISO 8196 IDF 128-3 :2009 – Milk — Definition and evaluation of the overall accuracy of alternative methods of milk analysis — Part 3: Protocol for the evaluation and validation of alternative quantitative methods of milk analysis
- Milchprüfring data MilkoScan™ 7 RM December 2019

16 Annex A - Table of results Phase I

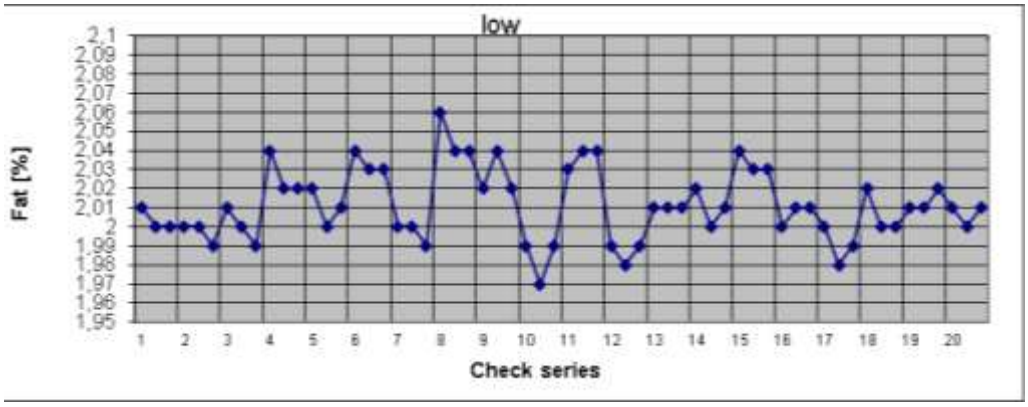
Table A1. Repeatability and stability on pilot samples for fat.

1) low fat concentration:

Check series q	Replicates n	Mean m	SD Sr(i)	Variance Var
20	2			
Sum	120,670	40,223		0,00140
Mean	2,011	2,011	0,008	0,000070
SD		0,018		
	1,97			

Cochran-Test: Cochran-Limit (P=0,95; n=3; q=20): 0,270				
0,095	<	0,270	→	Variance homogeneity
0,012	≤	0,019	→	Variance homogeneity

		Limit	
SD of repeatability Sr =	0,008	0,014	positive
SD between check series Sc =	0,017		
SD of daily reproducibility SR =	0,019	0,028	positive



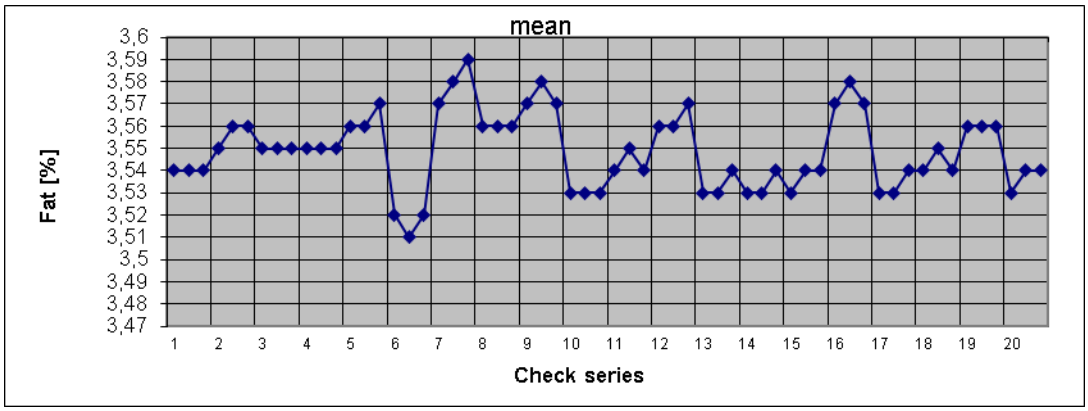
2) mean fat concentration:

Check series q	Replicates n	Mean m	SD Sr(i)	Variance Var
20	2			
Sum	170,340	70,977		0,00053
Mean	3,549	3,549	0,004	0,000027
SD		0,017		

3,51

Cochran-Test: Cochran-Limit (P=0,95; n=3; q=20): 0,270				
0,187	<	0,270	→	Variance homogeneity
0,010	≤	0,012	→	Variance homogeneity

		Limit	
SD of repeatability Sr =	0,005	0,014	positive
SD between check series Sc =	0,017		
SD of daily reproducibility SR =	0,017	0,028	positive



3) high fat concentration:

Check series q	Replicates n	Mean m	SD Sr(i)	Variance Var
20	2			
Sum	299,810	99,937		0,00043
Mean	4,997	4,997	0,004	0,000022
SD		0,023		

Cochran-Test: Cochran-Limit (P=0,95; n=3; q=20): 0,270				
0,077	<	0,270	→	Variance homogeneity
0,006	≤	0,011	→	Variance homogeneity

		Limit	
SD of repeatability Sr =	0,005	0,014	Positive
SD between check series Sc =	0,023		
SD of daily reproducibility SR =	0,023	0,028	positive

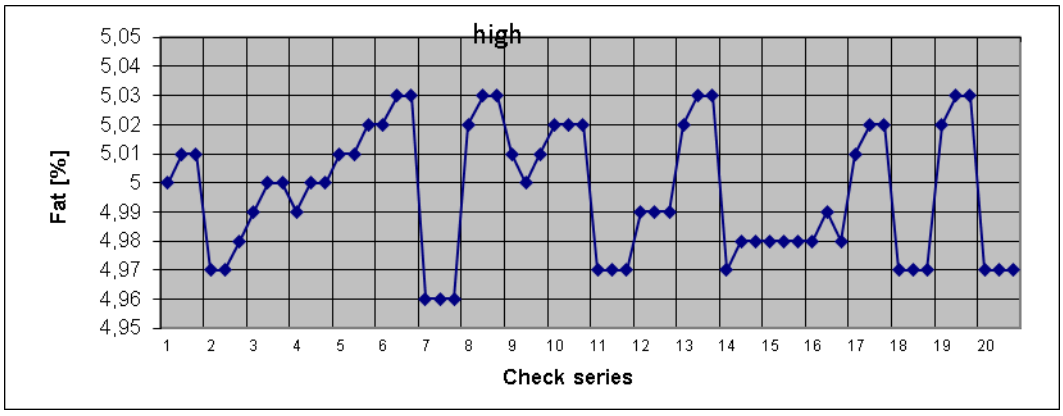


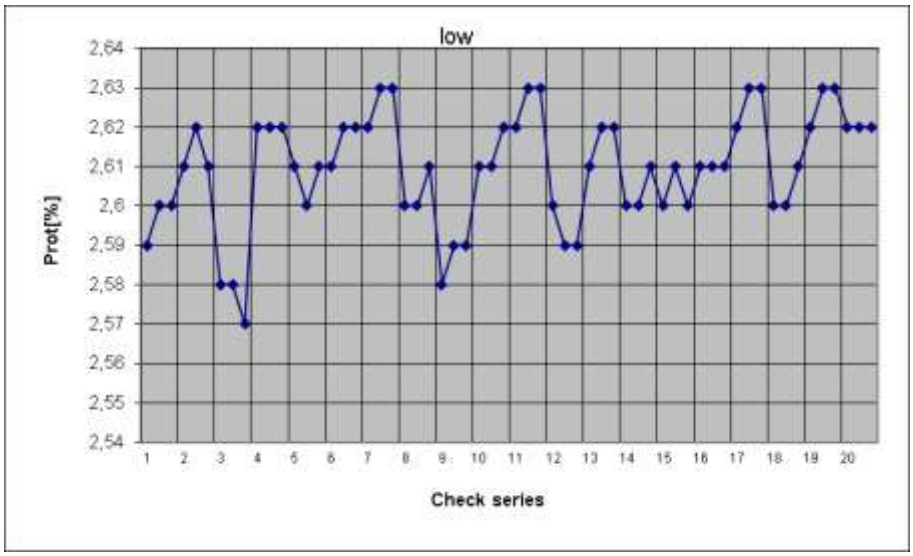
Table A2. Repeatability and stability on pilot samples for protein.

1) Low protein concentration:

Check series q	Replicates n	Mean m	SD Sr(i)	Variance Var
20	**2**			
Sum	156,570	52,190		0,00057
Mean	2,610	2,610	0,005	0,000028
SD		0,014		
	2,58			

Cochran-Test: Cochran-Limit (P=0,95; n=3; q=20): 0,270				
0,059	<	0,270	→	Variance homogeneity
0,006	≤	0,012	→	Variance homogeneity

		Limit	
SD of repeatability Sr =	**0,005**	**0,014**	**positive**
SD between check series Sc =	0,014		
SD of daily reproducibility SR =	**0,015**	**0,028**	**positive**



2) Mean protein concentration:

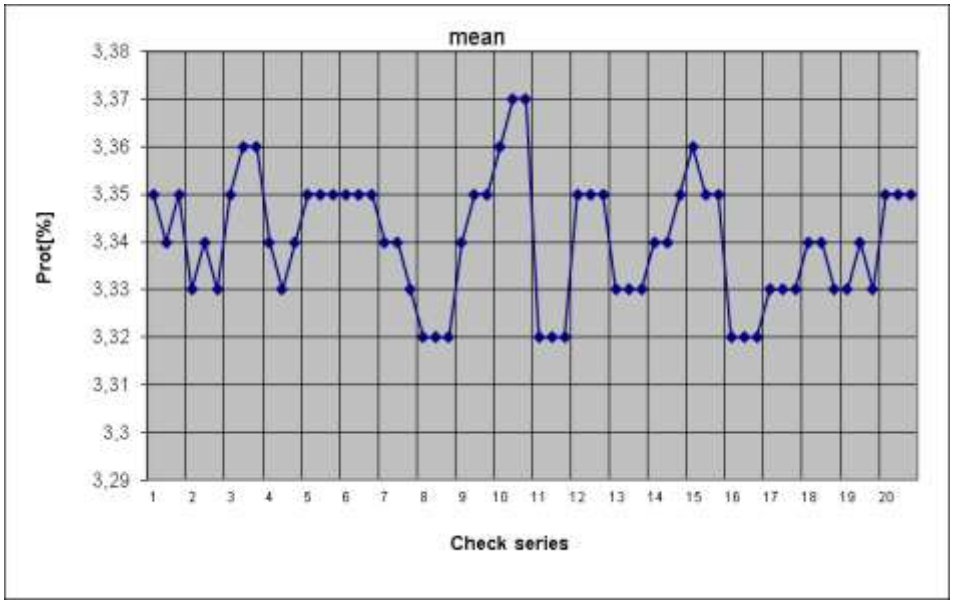
Check series q	Replicates n	Mean m	SD Sr(i)	Variance Var
20	**2**			
Sum	160,310	66,810		0,00037
Mean	3,340	3,341	0,003	0,000018
SD		0,013		

3,32

Cochran-Test:
Cochran-Limit (P=0,95; n=3; q=20): 0,270

0,091	<	0,270	→	Variance homogeneity
0,006	≤	0,010	→	Variance homogeneity

		Limit	
SD of repeatability Sr =	**0,004**	**0,014**	**positive**
SD between check series Sc =	0,013		
SD of daily reproducibility SR =	**0,013**	**0,028**	**positive**



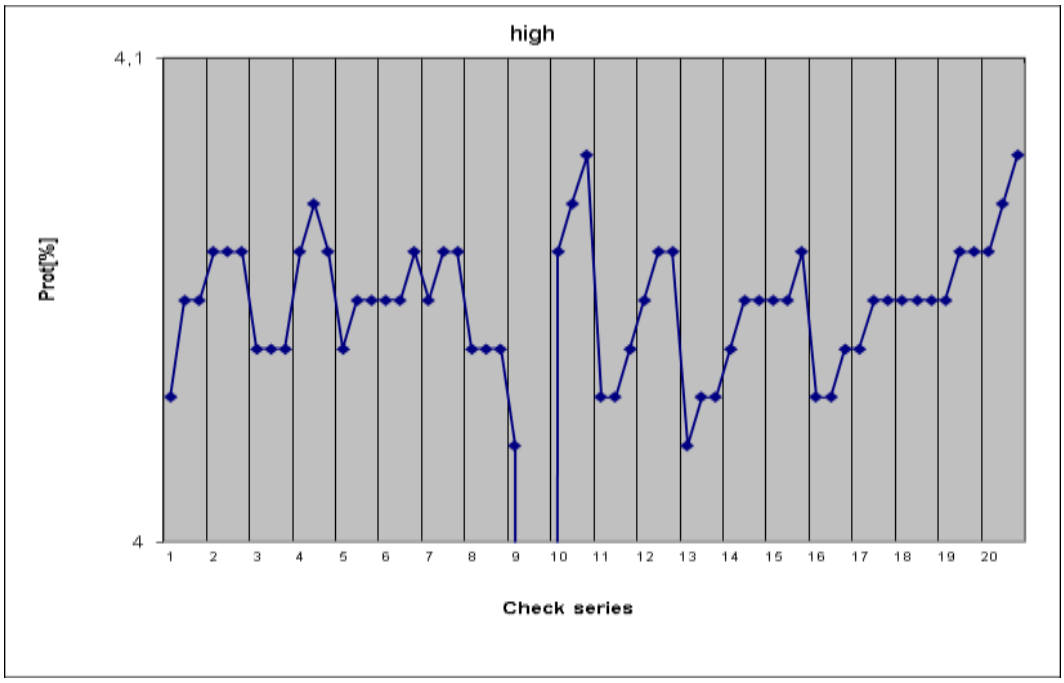
3) High protein concentration:

Check series q	Replicates n	Mean m	SD Sr(i)	Variance Var
20	2			
Sum	234,860	80,967		0,00073
Mean	4,049	4,048	0,005	0,000037
SD		0,013		

4,02

Cochran-Test: Cochran-Limit (P=0,95; n=3; q=20): 0,270				
0,182	<	0,270	→	Variance homogeneity
0,012	≤	0,014	→	Variance homogeneity

		Limit	
SD of repeatability Sr =	**0,006**	**0,014**	**positive**
SD between check series Sc =	0,013		
SD of daily reproducibility SR =	**0,014**	**0,028**	**positive**



Tab A3. Carry Over Milchprüfring-Bayern.

Carry-over

Fat

Sequence	Concentration				Difference			
	LL1	LL2	HL1	HL2	dL	dH	LL2 - REF LL	HL2 - REF HL
1	0,09	0,09	9,56	9,62	0,000	-0,060	0,00	0,02
2	0,09	0,08	9,59	9,62	0,010	-0,030	-0,01	0,02
3	0,1	0,09	9,6	9,62	0,010	-0,020	0,00	0,02
4	0,09	0,08	9,59	9,62	0,010	-0,030	-0,01	0,02
5	0,1	0,08	9,61	9,6	0,020	0,010	-0,01	0,00
6	0,1	0,08	9,59	9,61	0,020	-0,020	-0,01	0,01
7	0,1	0,08	9,6	9,63	0,020	-0,030	-0,01	0,03
8	0,09	0,08	9,58	9,61	0,010	-0,030	-0,01	0,01
9	0,1	0,08	9,59	9,6	0,020	-0,010	-0,01	0,00
10	0,09	0,08	9,6	9,6	0,010	0,000	-0,01	0,00
Mean	0,095	0,082	9,591	9,613	0,013	-0,022	-0,007	0,011
SD	0,005	0,004	0,014	0,011	0,007	0,019	0,004	0,011
N	10	10	10	10	10	10	10	10
REF LL:	0,09							
REF HL:	9,60							

Mean difference of concentration: 9,531

Carry-over ratios:		%	Limit	
	C. O. R. (H/L) =	0,14	1%	positive
	C. O. R. (L/H) =	-0,23	1%	positive

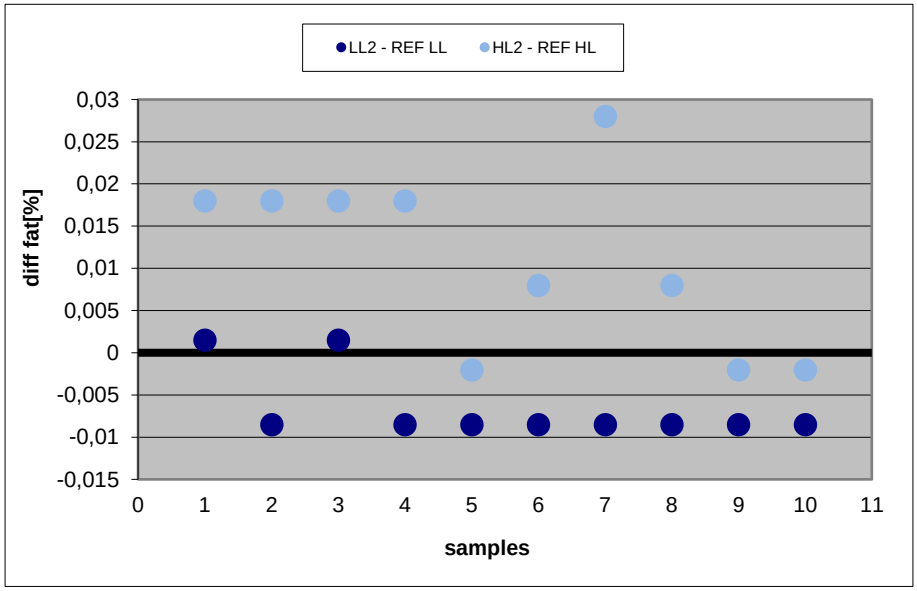


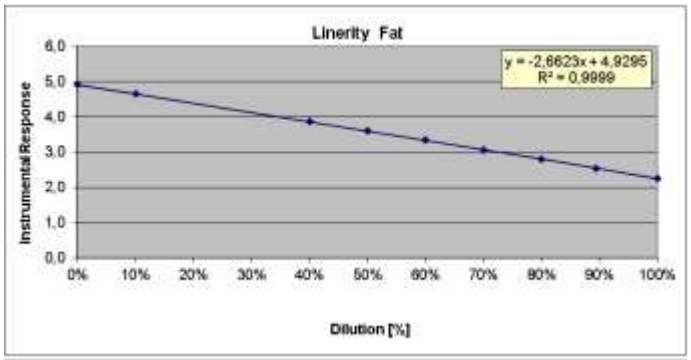
Table A4. Linearity test Milchprüfring-Bayern.

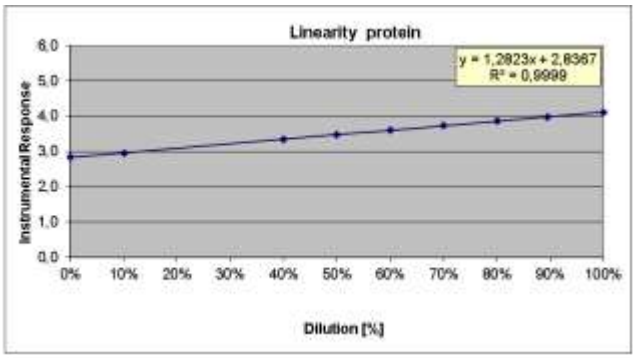
Linearity Fat g/100g

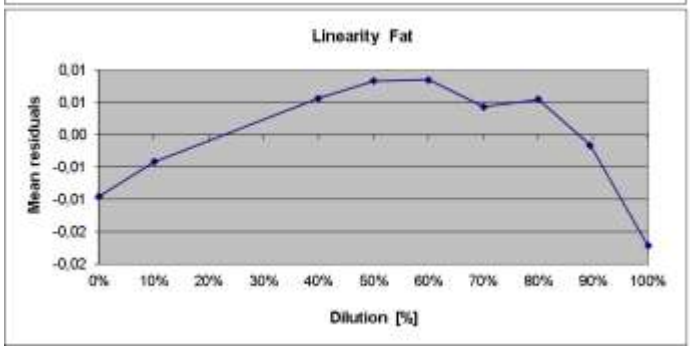
Sample	Dilution [m/m %] X	Replicates			Mean concentration Y	Mean residual e	SD Sr
		1	2	3			
1	100%	2,25	2,25	2,25	2,250	-0,017	0,000
2	89%	2,55	2,54	2,55	2,547	-0,002	0,006
3	80%	2,81	2,80	2,80	2,803	0,005	0,006
4	70%	3,07	3,07	3,07	3,070	0,004	0,000
5	60%	3,34	3,34	3,34	3,340	0,009	0,000
6	50%	3,60	3,61	3,61	3,607	0,008	0,006
7	40%	3,87	3,87	3,87	3,870	0,006	0,000
8	10%	4,66	4,66	4,65	4,657	-0,004	0,006
9	0%	4,92	4,92	4,92	4,920	-0,010	0,000
N	9	9	9	9	9	9	
Mean	0,555	3,452	3,451	3,451	3,451	0,000	0,004
SD	0,342	0,911	0,913	0,910	0,912	0,009	
Minimum	0,000	2,250	2,250	2,250	2,250	-0,017	
Maximum	1,000	4,920	4,920	4,920	4,920	0,009	
D = Max-Min	1,000	2,670	2,670	2,670	2,670	0,026	
Linear regression:							
Slope	-2,66234						
Intercept	4,92950						

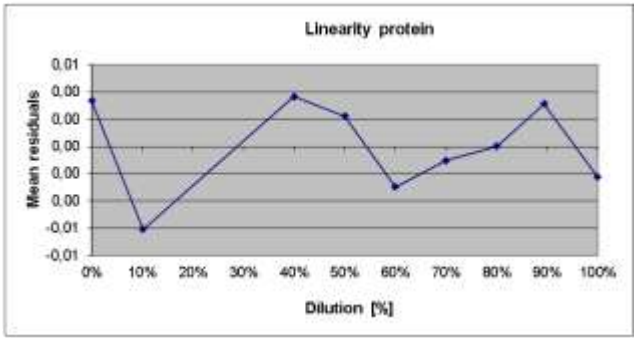
Linearity Protein g/100g

Sample	Dilution [m/m %] X	Replicates			Mean concentration Y	Mean residual e	SD Sr
		1	2	3			
1	100%	4,12	4,11	4,12	4,117	-0,002	0,006
2	89%	3,98	3,99	3,99	3,987	0,003	0,006
3	80%	3,86	3,86	3,87	3,863	0,000	0,006
4	70%	3,73	3,73	3,74	3,733	-0,001	0,006
5	60%	3,60	3,60	3,61	3,603	-0,003	0,006
6	50%	3,48	3,48	3,48	3,480	0,002	0,000
7	40%	3,36	3,35	3,35	3,353	0,004	0,006
8	10%	2,96	2,96	2,96	2,960	-0,006	0,000
9	0%	2,84	2,84	2,84	2,840	0,003	0,000
N	9	9	9	9	9	9	
Mean	0,555	3,548	3,547	3,551	3,549	0,000	0,005
SD	0,342	0,438	0,438	0,441	0,439	0,003	
Minimum	0,000	2,840	2,840	2,840	2,840	-0,006	
Maximum	1,000	4,120	4,110	4,120	4,117	0,004	
D = Max-Min	1,000	1,280	1,270	1,280	1,277	0,010	
Linear regression:							
Slope	1,28226						
Intercept	2,83666						


Linerity Fat
y = -2,6623x + 4,9295
R² = 0,9999
Instrumental Response
Dilution [%]


Linearity protein
y = 1,2823x + 2,8367
R² = 0,9999
Instrumental Response
Dilution [%]


Linearity Fat
Mean residuals
Dilution [%]


Linearity protein
Mean residuals
Dilution [%]

17 Annex B. Tables Interlaboratory study

Table B1 Fat results before and after calibration adjustment

Fat, g/100g

	Raw data slope=1 Intercept=0		**Data obtained with the calibration adjustment QLIP RM**		**Data Recalculated after calibration**			**Raw data slope=1 Intercept=0**		**Data obtained with the calibration adjustment QLIP RM**		**Data recalculated after calibration**	
	SAMPLE 1				**SAMPLE 1**			**SAMPLE 2**				**SAMPLE 2**	
Lab	MSC 2	MSC 12	slope	intercept	MSC 2	MSC 12	Lab	MSC 3	MSC 15	slope	intercept	MSC 3	MSC 15
L1F	2,23	2,23	1,0340	0,0000	2,31	2,31	L1F	3,94	3,93	1,0340	0,0000	4,07	4,06
L2F	2,23	2,24	0,9830	0,1100	2,30	2,31	L2F	4,00	4,00	0,9830	0,1100	4,04	4,04
L3F	2,23	2,24	1,0100	0,0500	2,30	2,31	L3F	3,96	3,96	1,0100	0,0500	4,05	4,05
L5F	2,15	2,18	0,9834	0,1632	2,28	2,31	L5F	3,94	3,92	0,9834	0,1632	4,04	4,02
L7F	2,22	2,24	1,0281	0,0058	2,29	2,31	L7F	3,92	3,95	1,0281	0,0058	4,04	4,07
L8F	2,23	2,24	1,0271	0,0195	2,31	2,32	L8F	3,93	3,95	1,0271	0,0195	4,06	4,08
L10F	2,24	2,24	1,0250	0,0171	2,31	2,31	L10F	3,95	3,95	1,0250	0,0171	4,07	4,07
L13F	2,24	2,24	0,9970	0,0700	2,30	2,30	L13F	3,99	3,99	0,9970	0,0700	4,05	4,05
L14F	2,29	2,29	1,0117	0,0042	2,32	2,32	L14F	4,01	4,03	1,0117	0,0042	4,06	4,08
L15F	2,10	2,11	1,0165	0,1657	2,30	2,31	L15F	3,80	3,82	1,0165	0,1657	4,03	4,05
L16F	2,15	2,16	0,9904	0,1686	2,30	2,31	L16F	3,88	3,90	0,9904	0,1686	4,01	4,03
L17F	2,22	2,23	1,0458	0,0136	2,34	2,35	L17F	3,93	3,94	1,0458	0,0136	4,12	4,13

	Raw data slope=1 Intercept=0		**Data obtained with the calibration adjustment QLIP RM**		**Data recalculated after calibration**			**Raw data slope=1 Intercept=0**		**Data obtained with the calibration adjustment QLIP RM**		**Data recalculated after calibration**	
	SAMPLE 3				**SAMPLE 3**			**SAMPLE 4**				**SAMPLE 4**	
Lab	MSC 6	MSC 13	slope	intercept	MSC 6	MSC 13	Lab	MSC 9	MSC 16	slope	intercept	MSC 9	MSC 16
L1F	4,67	4,66	1,0340	0,0000	4,83	4,82	L1F	3,86	3,85	1,0340	0,0000	3,99	3,98
L2F	4,80	4,78	0,9830	0,1100	4,83	4,81	L2F	3,95	3,94	0,9830	0,1100	3,99	3,98
L3F	4,73	4,72	1,0100	0,0500	4,83	4,82	L3F	3,91	3,90	1,0100	0,0500	4,00	3,99
L5F	4,75	4,73	0,9834	0,1632	4,83	4,81	L5F	3,92	3,93	0,9834	0,1632	4,02	4,03
L7F	4,68	4,68	1,0281	0,0058	4,82	4,82	L7F	3,87	3,86	1,0281	0,0058	3,98	3,97
L8F	4,68	4,68	1,0271	0,0195	4,83	4,83	L8F	3,87	3,87	1,0271	0,0195	3,99	3,99
L10F	4,70	4,69	1,0250	0,0171	4,83	4,82	L10F	3,87	3,87	1,0250	0,0171	3,98	3,98
L13F	4,79	4,77	0,9970	0,0700	4,85	4,83	L13F	3,95	3,96	0,9970	0,0700	4,01	4,02
L14F	4,78	4,76	1,0117	0,0042	4,84	4,82	L14F	3,95	3,95	1,0117	0,0042	4,00	4,00
L15F	4,59	4,59	1,0165	0,1657	4,83	4,83	L15F	3,76	3,74	1,0165	0,1657	3,99	3,97
L16F	4,70	4,69	0,9904	0,1686	4,82	4,81	L16F	3,84	3,83	0,9904	0,1686	3,97	3,96
L17F	4,68	4,67	1,0458	0,0136	4,91	4,90	L17F	3,85	3,86	1,0458	0,0136	4,04	4,05

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 5				SAMPLE 5			SAMPLE 6				SAMPLE 6	
Lab	MSC 4	MSC 20	slope	intercept	MSC 4	MSC 20	Lab	MSC 7	MSC 18	slope	intercept	MSC 7	MSC 18
L1F	2,41	2,41	1,0340	0,0000	2,49	2,49	L1F	3,28	3,27	1,0340	0,0000	3,39	3,38
L2F	2,45	2,46	0,9830	0,1100	2,52	2,53	L2F	3,36	3,36	0,9830	0,1100	3,41	3,41
L3F	2,44	2,45	1,0100	0,0500	2,51	2,52	L3F	3,34	3,33	1,0100	0,0500	3,42	3,41
L5F	2,42	2,40	0,9834	0,1632	2,54	2,52	L5F	3,33	3,32	0,9834	0,1632	3,44	3,43
L7F	2,41	2,43	1,0281	0,0058	2,48	2,50	L7F	3,30	3,29	1,0281	0,0058	3,40	3,39
L8F	2,41	2,41	1,0271	0,0195	2,49	2,49	L8F	3,27	3,27	1,0271	0,0195	3,38	3,38
L10F	2,40	2,42	1,0250	0,0171	2,48	2,50	L10F	3,29	3,29	1,0250	0,0171	3,39	3,39
L13F	2,46	2,47	0,9970	0,0700	2,52	2,53	L13F	3,36	3,35	0,9970	0,0700	3,42	3,41
L14F	2,47	2,47	1,0117	0,0042	2,50	2,50	L14F	3,35	3,35	1,0117	0,0042	3,39	3,39
L15F	2,30	2,32	1,0165	0,1657	2,50	2,52	L15F	3,22	3,21	1,0165	0,1657	3,44	3,43
L16F	2,36	2,36	0,9904	0,1686	2,51	2,51	L16F	3,29	3,29	0,9904	0,1686	3,43	3,43
L17F	2,41	2,40	1,0458	0,0136	2,53	2,52	L17F	3,26	3,28	1,0458	0,0136	3,42	3,44

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibratio			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 7				SAMPLE 7			SAMPLE 8				SAMPLE 8	
Lab	FM3	FM5	slope	intercept	FM3	FM5	Lab	FM15	FM17	slope	intercept	FM15	FM17
L1F	2,99	2,99	1,0340	0,0000	3,09	3,09	L1F	5,14	5,14	1,0340	0,0000	5,31	5,31
L2F	3,03	3,02	0,9830	0,1100	3,09	3,08	L2F	5,32	5,32	0,9830	0,1100	5,34	5,34
L3F	3,01	3,01	1,0100	0,0500	3,09	3,09	L3F	5,25	5,24	1,0100	0,0500	5,35	5,34
L5F	3,00	2,97	0,9834	0,1632	3,11	3,08	L5F	5,27	5,26	0,9834	0,1632	5,35	5,34
L7F	3,00	2,99	1,0281	0,0058	3,09	3,08	L7F	5,18	5,18	1,0281	0,0058	5,33	5,33
L8F	2,99	3,00	1,0271	0,0195	3,09	3,10	L8F	5,16	5,17	1,0271	0,0195	5,32	5,33
L10F	3,00	3,00	1,0250	0,0171	3,09	3,09	L10F	5,16	5,17	1,0250	0,0171	5,31	5,32
L13F	3,03	3,03	0,9970	0,0700	3,09	3,09	L13F	5,30	5,30	0,9970	0,0700	5,35	5,35
L14F	3,06	3,06	1,0117	0,0042	3,10	3,10	L14F	5,25	5,27	1,0117	0,0042	5,32	5,34
L15F	2,90	2,90	1,0165	0,1657	3,11	3,11	L15F	5,16	5,16	1,0165	0,1657	5,41	5,41
L16F	2,96	2,96	0,9904	0,1686	3,10	3,10	L16F	5,27	5,27	0,9904	0,1686	5,39	5,39
L17F	2,99	2,99	1,0458	0,0136	3,14	3,14	L17F	5,13	5,10	1,0458	0,0136	5,38	5,35

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibratio			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 9				SAMPLE 9			SAMPLE 10				SAMPLE 10	
Lab	FM11	FM13	slope	intercept	FM11	FM13	Lab	FM23	FM24	slope	intercept	FM23	FM24
L1F	4,82	4,81	1,0340	0,0000	4,98	4,97	L1F	4,16	4,16	1,0340	0,0000	4,30	4,30
L2F	4,96	4,96	0,9830	0,1100	4,99	4,99	L2F	4,28	4,29	0,9830	0,1100	4,32	4,33
L3F	4,89	4,88	1,0100	0,0500	4,99	4,98	L3F	4,22	4,23	1,0100	0,0500	4,31	4,32
L5F	4,83	4,90	0,9834	0,1632	4,91	4,98	L5F	4,22	4,23	0,9834	0,1632	4,31	4,32
L7F	4,82	4,83	1,0281	0,0058	4,96	4,97	L7F	4,18	4,19	1,0281	0,0058	4,30	4,31
L8F	4,83	4,84	1,0271	0,0195	4,98	4,99	L8F	4,18	4,18	1,0271	0,0195	4,31	4,31
L10F	4,85	4,84	1,0250	0,0171	4,99	4,98	L10F	4,19	4,19	1,0250	0,0171	4,31	4,31
L13F	4,91	4,93	0,9970	0,0700	4,97	4,99	L13F	4,26	4,26	0,9970	0,0700	4,32	4,32
L14F	4,92	4,92	1,0117	0,0042	4,98	4,98	L14F	4,25	4,27	1,0117	0,0042	4,30	4,32
L15F	4,71	4,71	1,0165	0,1657	4,95	4,95	L15F	4,06	4,08	1,0165	0,1657	4,29	4,31
L16F	4,82	4,82	0,9904	0,1686	4,94	4,94	L16F	4,16	4,17	0,9904	0,1686	4,29	4,30
L17F	4,82	4,81	1,0458	0,0136	5,05	5,04	L17F	4,17	4,15	1,0458	0,0136	4,37	4,35

Grubbs oulier
Grubbs straggler
Cochran outlier

Table B2 Protein results before and after calibration adjustment.

Protein, g/100g

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 1				SAMPLE 1			SAMPLE 2				SAMPLE 2	
Lab	MSC 2	MSC 12	slope	intercept	MSC 2	MSC 12	Lab	MSC 3	MSC 15	slope	intercept	MSC 3	MSC 15
L1P	3,23	3,22	1,0070	-0,0161	3,24	3,23	L1P	4,51	4,52	1,0070	-0,0161	4,53	4,54
L2P	3,27	3,26	1,0000	-0,0100	3,26	3,25	L2P	4,55	4,55	1,0000	-0,0100	4,54	4,54
L3P	3,22	3,21	1,0250	-0,0500	3,25	3,24	L3P	4,49	4,48	1,0250	-0,0500	4,55	4,54
L5P	3,24	3,25	1,0000	-0,0511	3,19	3,20	L5P	4,58	4,59	1,0000	-0,0511	4,53	4,54
L7P	3,20	3,22	1,0092	-0,0097	3,22	3,24	L7P	4,49	4,50	1,0092	-0,0097	4,52	4,53
L8P	3,21	3,22	1,0081	-0,0049	3,23	3,24	L8P	4,50	4,50	1,0081	-0,0049	4,53	4,53
L10P	3,22	3,22	1,0040	0,0140	3,25	3,25	L10P	4,50	4,50	1,0040	0,0140	4,53	4,53
L13P	3,23	3,24	1,0130	-0,0400	3,23	3,24	L13P	4,53	4,53	1,0130	-0,0400	4,55	4,55
L14P	3,26	3,26	0,9960	0,0148	3,26	3,26	L14P	4,55	4,55	0,9960	0,0148	4,55	4,55
L15P	3,17	3,17	1,0254	-0,0604	3,19	3,19	L15P	4,45	4,46	1,0254	-0,0604	4,50	4,51
L16P	3,20	3,19	1,0171	-0,0624	3,19	3,18	L16P	4,50	4,50	1,0171	-0,0624	4,51	4,51
L17P	3,21	3,21	1,0130	-0,0200	3,23	3,23	L17P	4,51	4,50	1,0130	-0,0200	4,55	4,54

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 3				SAMPLE 3			SAMPLE 4				SAMPLE 4	
lab	MSC 6	MSC 13	slope	intercept	MSC 6	MSC 13	lab	MSC 9	MSC 16	slope	intercept	MSC 9	MSC 16
L1P	3,89	3,89	1,0070	-0,0161	3,90	3,90	L1P	4,39	4,40	1,0070	-0,0161	4,40	4,41
L2P	3,94	3,94	1,0000	-0,0100	3,93	3,93	L2P	4,44	4,43	1,0000	-0,0100	4,43	4,42
L3P	3,88	3,89	1,0250	-0,0500	3,93	3,94	L3P	4,37	4,36	1,0250	-0,0500	4,43	4,42
L5P	3,95	3,93	1,0000	-0,0511	3,90	3,88	L5P	4,51	4,51	1,0000	-0,0511	4,46	4,46
L7P	3,87	3,88	1,0092	-0,0097	3,90	3,91	L7P	4,38	4,38	1,0092	-0,0097	4,41	4,41
L8P	3,88	3,87	1,0081	-0,0049	3,91	3,90	L8P	4,39	4,39	1,0081	-0,0049	4,42	4,42
L10P	3,87	3,87	1,0040	0,0140	3,90	3,90	L10P	4,38	4,38	1,0040	0,0140	4,41	4,41
L13P	3,93	3,93	1,0130	-0,0400	3,94	3,94	L13P	4,41	4,41	1,0130	-0,0400	4,43	4,43
L14P	3,92	3,92	0,9960	0,0148	3,92	3,92	L14P	4,45	4,44	0,9960	0,0148	4,45	4,44
L15P	3,83	3,83	1,0254	-0,0604	3,87	3,87	L15P	4,38	4,38	1,0254	-0,0604	4,43	4,43
L16P	3,87	3,87	1,0171	-0,0624	3,87	3,87	L16P	4,43	4,43	1,0171	-0,0624	4,44	4,44
L17P	3,87	3,88	1,0130	-0,0200	3,90	3,91	L17P	4,38	4,38	1,0130	-0,0200	4,42	4,42

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 5				SAMPLE 5			SAMPLE 6				SAMPLE 6	
lab	MSC 4	MSC 20	slope	intercept	MSC 4	MSC 20	lab	MSC 7	MSC 18	slope	intercept	MSC 7	MSC 18
L1P	3,11	3,13	1,0070	-0,0161	3,12	3,14	L1P	3,06	3,06	1,0070	-0,0161	3,07	3,07
L2P	3,14	3,15	1,0000	-0,0100	3,13	3,14	L2P	3,11	3,11	1,0000	-0,0100	3,10	3,10
L3P	3,10	3,10	1,0250	-0,0500	3,13	3,13	L3P	3,07	3,08	1,0250	-0,0500	3,10	3,11
L5P	3,22	3,23	1,0000	-0,0511	3,17	3,18	L5P	3,17	3,15	1,0000	-0,0511	3,12	3,10
L7P	3,11	3,13	1,0092	-0,0097	3,13	3,15	L7P	3,05	3,05	1,0092	-0,0097	3,07	3,07
L8P	3,12	3,12	1,0081	-0,0049	3,14	3,14	L8P	3,06	3,06	1,0081	-0,0049	3,08	3,08
L10P	3,12	3,13	1,0040	0,0140	3,15	3,16	L10P	3,05	3,05	1,0040	0,0140	3,08	3,08
L13P	3,11	3,13	1,0130	-0,0400	3,11	3,13	L13P	3,10	3,10	1,0130	-0,0400	3,10	3,10
L14P	3,17	3,15	0,9960	0,0148	3,17	3,15	L14P	3,09	3,09	0,9960	0,0148	3,09	3,09
L15P	3,15	3,16	1,0254	-0,0604	3,17	3,18	L15P	3,09	3,08	1,0254	-0,0604	3,11	3,10
L16P	3,17	3,17	1,0171	-0,0624	3,16	3,16	L16P	3,11	3,10	1,0171	-0,0624	3,10	3,09
L17P	3,11	3,11	1,0130	-0,0200	3,13	3,13	L17P	3,05	3,05	1,0130	-0,0200	3,07	3,07

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 7				SAMPLE 7			SAMPLE 8				SAMPLE 8	
lab	FM3	FM5	slope	intercept	FM3	FM5	lab	FM15	FM17	slope	intercept	FM15	FM17
L1P	3,47	3,45	1,0070	-0,0161	3,48	3,46	L1P	4,81	4,80	1,0070	-0,0161	4,83	4,82
L2P	3,53	3,52	1,0000	-0,0100	3,52	3,51	L2P	4,87	4,86	1,0000	-0,0100	4,86	4,85
L3P	3,47	3,47	1,0250	-0,0500	3,51	3,51	L3P	4,81	4,81	1,0250	-0,0500	4,88	4,88
L5P	3,57	3,57	1,0000	-0,0511	3,52	3,52	L5P	4,94	4,95	1,0000	-0,0511	4,89	4,90
L7P	3,45	3,45	1,0092	-0,0097	3,47	3,47	L7P	4,76	4,75	1,0092	-0,0097	4,79	4,78
L8P	3,45	3,45	1,0081	-0,0049	3,47	3,47	L8P	4,76	4,76	1,0081	-0,0049	4,79	4,79
L10P	3,45	3,44	1,0040	0,0140	3,48	3,47	L10P	4,76	4,76	1,0040	0,0140	4,79	4,79
L13P	3,49	3,49	1,0130	-0,0400	3,50	3,50	L13P	4,84	4,84	1,0130	-0,0400	4,86	4,86
L14P	3,50	3,50	0,9960	0,0148	3,50	3,50	L14P	4,82	4,82	0,9960	0,0148	4,82	4,82
L15P	3,48	3,48	1,0254	-0,0604	3,51	3,51	L15P	4,82	4,82	1,0254	-0,0604	4,88	4,88
L16P	3,50	3,51	1,0171	-0,0624	3,50	3,51	L16P	4,89	4,85	1,0171	-0,0624	4,91	4,87
L17P	3,44	3,45	1,0130	-0,0200	3,46	3,47	L17P	4,78	4,80	1,0130	-0,0200	4,82	4,84

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 9				SAMPLE 9			SAMPLE 10				SAMPLE 10	
lab	FM11	FM13	slope	intercept	FM11	FM13	lab	FM23	FM24	slope	intercept	FM23	FM24
L1P	3,71	3,71	1,0070	-0,0161	3,72	3,72	L1P	3,52	3,52	1,0070	-0,0161	3,53	3,53
L2P	3,74	3,75	1,0000	-0,0100	3,73	3,74	L2P	3,56	3,57	1,0000	-0,0100	3,55	3,56
L3P	3,69	3,69	1,0250	-0,0500	3,73	3,73	L3P	3,51	3,52	1,0250	-0,0500	3,55	3,56
L5P	3,81	3,82	1,0000	-0,0511	3,76	3,77	L5P	3,61	3,62	1,0000	-0,0511	3,56	3,57
L7P	3,67	3,71	1,0092	-0,0097	3,69	3,73	L7P	3,51	3,52	1,0092	-0,0097	3,53	3,54
L8P	3,67	3,71	1,0081	-0,0049	3,69	3,74	L8P	3,51	3,51	1,0081	-0,0049	3,53	3,53
L10P	3,70	3,70	1,0040	0,0140	3,73	3,73	L10P	3,51	3,51	1,0040	0,0140	3,54	3,54
L13P	3,73	3,73	1,0130	-0,0400	3,74	3,74	L13P	3,54	3,54	1,0130	-0,0400	3,55	3,55
L14P	3,74	3,74	0,9960	0,0148	3,74	3,74	L14P	3,54	3,55	0,9960	0,0148	3,54	3,55
L15P	3,69	3,71	1,0254	-0,0604	3,72	3,74	L15P	3,52	3,52	1,0254	-0,0604	3,55	3,55
L16P	3,73	3,75	1,0171	-0,0624	3,73	3,75	L16P	3,56	3,56	1,0171	-0,0624	3,56	3,56
L17P	3,70	3,70	1,0130	-0,0200	3,73	3,73	L17P	3,50	3,51	1,0130	-0,0200	3,53	3,54

Grubbs oulier
Grubbs straggler
Cochran outlier

Table B3. Lactose results before and after calibration adjustment.

Lactose, g/100g

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 1				SAMPLE 1			SAMPLE 2				SAMPLE 2	
Lab	MSC 2	MSC 12	slope	intercept	MSC 2	MSC 12	Lab	MSC 3	MSC 15	slope	intercept	MSC 3	MSC 15
L1P	5,08	5,08	1,01	-0,17	4,94	4,94	L1P	4,79	4,78	1,01	-0,17	4,65	4,64
L2P	5,22	5,23	1,01	-0,35	4,90	4,91	L2P	4,93	4,93	1,01	-0,35	4,61	4,61
L3P	5,12	5,11	1,03	-0,33	4,93	4,92	L3P	4,80	4,80	1,03	-0,33	4,60	4,60
L5P	4,98	4,99	0,99	0,02	4,94	4,95	L5P	4,66	4,65	0,99	0,02	4,62	4,61
L7P	4,86	4,88	1,01	0,02	4,92	4,94	L7P	4,55	4,56	1,01	0,02	4,60	4,61
L8P	4,92	4,92	1,00	0,03	4,93	4,93	L8P	4,60	4,60	1,00	0,03	4,61	4,61
L10P	4,91	4,91	1,01	0,00	4,94	4,94	L10P	4,58	4,59	1,01	0,00	4,61	4,62
L13P	4,97	4,97	0,99	0,02	4,95	4,95	L13P	4,64	4,64	0,99	0,02	4,62	4,62
L14P	5,10	5,10	1,02	0,01	5,21	5,21	L14P	4,80	4,79	1,02	0,01	4,91	4,90
L15P	4,90	4,89	1,01	0,00	4,93	4,92	L15P	4,57	4,57	1,01	0,00	4,60	4,60
L16P	4,93	4,93	1,00	0,00	4,93	4,93	L16P	4,61	4,61	1,00	0,00	4,61	4,61
L17P	5,06	5,06	1,01	-0,20	4,91	4,91	L17P	4,76	4,77	1,01	-0,20	4,60	4,61

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 3				SAMPLE 3			SAMPLE 4				SAMPLE 4	
Lab	MSC 6	MSC 13	slope	intercept	MSC 6	MSC 13	Lab	MSC 9	MSC 16	slope	intercept	MSC 9	MSC 16
L1P	4,55	4,54	1,01	-0,17	4,41	4,40	L1P	4,62	4,62	1,01	-0,17	4,48	4,48
L2P	4,68	4,68	1,01	-0,35	4,36	4,36	L2P	4,77	4,79	1,01	-0,35	4,45	4,47
L3P	4,56	4,56	1,03	-0,33	4,35	4,35	L3P	4,67	4,67	1,03	-0,33	4,47	4,47
L5P	4,43	4,43	0,99	0,02	4,40	4,40	L5P	4,52	4,53	0,99	0,02	4,49	4,50
L7P	4,33	4,35	1,01	0,02	4,38	4,40	L7P	4,43	4,43	1,01	0,02	4,48	4,48
L8P	4,38	4,39	1,00	0,03	4,39	4,40	L8P	4,46	4,47	1,00	0,03	4,47	4,48
L10P	4,37	4,37	1,01	0,00	4,40	4,40	L10P	4,45	4,45	1,01	0,00	4,48	4,48
L13P	4,41	4,43	0,99	0,02	4,39	4,41	L13P	4,50	4,50	0,99	0,02	4,48	4,48
L14P	4,55	4,55	1,02	0,01	4,65	4,65	L14P	4,63	4,63	1,02	0,01	4,73	4,73
L15P	4,35	4,35	1,01	0,00	4,38	4,38	L15P	4,43	4,44	1,01	0,00	4,46	4,47
L16P	4,39	4,39	1,00	0,00	4,39	4,39	L16P	4,47	4,47	1,00	0,00	4,47	4,47
L17P	4,52	4,53	1,01	-0,20	4,36	4,37	L17P	4,61	4,61	1,01	-0,20	4,45	4,45

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 5				SAMPLE 5			SAMPLE 6				SAMPLE 6	
Lab	MSC 4	MSC 20	slope	intercept	MSC 4	MSC 20	Lab	MSC 7	MSC 18	slope	intercept	MSC 7	MSC 18
L1P	4,78	4,77	1,01	-0,17	4,64	4,63	L1P	4,69	4,69	1,01	-0,17	4,55	4,55
L2P	4,90	4,90	1,01	-0,35	4,58	4,58	L2P	4,86	4,86	1,01	-0,35	4,54	4,54
L3P	4,80	4,79	1,03	-0,33	4,60	4,59	L3P	4,75	4,75	1,03	-0,33	4,55	4,55
L5P	4,66	4,67	0,99	0,02	4,62	4,63	L5P	4,63	4,64	0,99	0,02	4,59	4,60
L7P	4,56	4,56	1,01	0,02	4,61	4,61	L7P	4,53	4,53	1,01	0,02	4,58	4,58
L8P	4,61	4,60	1,00	0,03	4,62	4,61	L8P	4,57	4,57	1,00	0,03	4,58	4,58
L10P	4,59	4,59	1,01	0,00	4,62	4,62	L10P	4,56	4,56	1,01	0,00	4,59	4,59
L13P	4,65	4,65	0,99	0,02	4,63	4,63	L13P	4,61	4,62	0,99	0,02	4,59	4,60
L14P	4,80	4,80	1,02	0,01	4,91	4,91	L14P	4,71	4,71	1,02	0,01	4,82	4,82
L15P	4,58	4,57	1,01	0,00	4,61	4,60	L15P	4,54	4,54	1,01	0,00	4,57	4,57
L16P	4,61	4,61	1,00	0,00	4,61	4,61	L16P	4,58	4,58	1,00	0,00	4,58	4,58
L17P	4,76	4,76	1,01	-0,20	4,60	4,60	L17P	4,67	4,68	1,01	-0,20	4,51	4,52

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 7				SAMPLE 7			SAMPLE 8				SAMPLE 8	
lab	FM3	FM5	slope	intercept	FM3	FM5	lab	FM15	FM17	slope	intercept	FM15	FM17
L1P	4,63	4,63	1,01	-0,17	4,49	4,49	L1P	4,11	4,11	1,01	-0,17	3,97	3,97
L2P	4,75	4,76	1,01	-0,35	4,43	4,44	L2P	4,31	4,31	1,01	-0,35	3,99	3,99
L3P	4,65	4,64	1,03	-0,33	4,45	4,44	L3P	4,19	4,20	1,03	-0,33	3,97	3,98
L5P	4,52	4,51	0,99	0,02	4,49	4,48	L5P	4,08	4,08	0,99	0,02	4,05	4,05
L7P	4,42	4,42	1,01	0,02	4,47	4,47	L7P	4,00	3,99	1,01	0,02	4,05	4,04
L8P	4,46	4,46	1,00	0,03	4,47	4,47	L8P	4,03	4,02	1,00	0,03	4,05	4,04
L10P	4,45	4,45	1,01	0,00	4,48	4,48	L10P	4,01	4,02	1,01	0,00	4,04	4,05
L13P	4,49	4,50	0,99	0,02	4,47	4,48	L13P	4,05	4,05	0,99	0,02	4,03	4,03
L14P	4,65	4,64	1,02	0,01	4,75	4,74	L14P	4,12	4,11	1,02	0,01	4,21	4,20
L15P	4,43	4,43	1,01	0,00	4,46	4,46	L15P	4,00	4,00	1,01	0,00	4,03	4,03
L16P	4,47	4,46	1,00	0,00	4,47	4,46	L16P	4,04	4,03	1,00	0,00	4,04	4,03
L17P	4,61	4,61	1,01	-0,20	4,45	4,45	L17P	4,10	4,10	1,01	-0,20	3,94	3,94

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 9				SAMPLE 9			SAMPLE 10				SAMPLE 10	
Lab	FM11	FM13	slope	intercept	FM11	FM13	Lab	FM23	FM24	slope	intercept	FM23	FM24
L1P	4,67	4,66	1,01	-0,17	4,53	4,52	L1P	4,71	4,71	1,01	-0,17	4,57	4,57
L2P	4,81	4,82	1,01	-0,35	4,49	4,50	L2P	4,87	4,87	1,01	-0,35	4,55	4,55
L3P	4,69	4,70	1,03	-0,33	4,49	4,50	L3P	4,76	4,75	1,03	-0,33	4,56	4,55
L5P	4,56	4,57	0,99	0,02	4,53	4,54	L5P	4,62	4,61	0,99	0,02	4,58	4,57
L7P	4,44	4,48	1,01	0,02	4,49	4,53	L7P	4,51	4,52	1,01	0,02	4,56	4,57
L8P	4,50	4,53	1,00	0,03	4,51	4,54	L8P	4,55	4,56	1,00	0,03	4,56	4,57
L10P	4,50	4,50	1,01	0,00	4,53	4,53	L10P	4,54	4,54	1,01	0,00	4,57	4,57
L13P	4,55	4,56	0,99	0,02	4,53	4,54	L13P	4,60	4,59	0,99	0,02	4,58	4,57
L14P	4,68	4,67	1,02	0,01	4,78	4,77	L14P	4,72	4,72	1,02	0,01	4,83	4,83
L15P	4,48	4,49	1,01	0,00	4,51	4,52	L15P	4,53	4,53	1,01	0,00	4,56	4,56
L16P	4,51	4,53	1,00	0,00	4,51	4,53	L16P	4,56	4,56	1,00	0,00	4,56	4,56
L17P	4,65	4,65	1,01	-0,20	4,49	4,49	L17P	4,69	4,70	1,01	-0,20	4,53	4,54

Grubbs oulier

Grubbs straggler

Cochran outlier

Table B4. Urea results before and after calibration adjustment.

Urea, mg/dl

	Raw data slope=1 Intercept=0		**Data obtained with the calibration adjustment QLIP RM**		**Data recalculated after calibration**			**Raw data slope=1 Intercept=0**		**Data obtained with the calibration adjustment QLIP RM**		**Data recalculated after calibration**	
	SAMPLE 1				**SAMPLE 1**			**SAMPLE 2**				**SAMPLE 2**	
Lab	MSC 2	MSC 12	slope	intercept	MSC 2	MSC 12	Lab	**MSC 3**		slope		**MSC 3**	
L1U	42	43	1,073	-24,970	20	21	L1L	52,40	53,30	1,07	-24,97	31,26	32,22
L2U	32	31	1,037	-8,500	25	24	L2L	41	40	1,037	-8,500	34	33
L3U	36	33	1,114	-15,800	24	21	L3L	44	43	1,114	-15,800	33	32
L5U	46	44	1,092	-24,990	25	23	L5L	56	54	1,092	-24,990	36	33
L7U	37	37	1,084	-14,622	25	25	L7L	42	42	1,084	-14,622	31	31
L8U	41	41	1,063	-21,996	22	22	L8L	51	51	1,063	-21,996	32	32
L10U	45	44	1,100	-25,220	24	23	L10L	53	52	1,100	-25,220	33	32
L13U	38	39	1,092	-18,000	23	25	L13L	48	47	1,092	-18,000	34	33
L14U	29	29	1,136	-8,760	24	24	L14L	35	35	1,136	-8,760	31	31
L15U	35	35	1,085	-12,350	26	26	L15L	42	43	1,085	-12,350	33	35
L16U	35	33	1,068	-8,980	28	26	L16L	40	39	1,068	-8,980	33	33
L17U	41	41	1,058	-21,390	22	22	L17L	49	49	1,058	-21,390	30	31

	Raw data slope=1 Intercept=0		**Data obtained with the calibration adjustment QLIP RM**		**Data recalculated after calibration**			**Raw data slope=1 Intercept=0**		**Data obtained with the calibration adjustment QLIP RM**		**Data recalculated after calibration**	
	SAMPLE 3				**SAMPLE 3**			**SAMPLE 4**				**SAMPLE 4**	
Lab	**MSC 6**		slope		**MSC 6**		Lab	**MSC 9**		slope		**MSC 9**	
L1L	67,50	67,20	1,07	-24,97	47,46	47,14	L1L	47,30	49,20	1,07	-24,97	25,78	27,82
L2L	54	53	1,037	-8,500	47	46	L2L	34	34	1,037	-8,500	27	27
L3L	55	54	1,114	-15,800	46	44	L3L	38	37	1,114	-15,800	27	26
L5L	68	68	1,092	-24,990	50	49	L5L	48	50	1,092	-24,990	27	29
L7L	55	54	1,084	-14,622	45	44	L7L	37	38	1,084	-14,622	26	27
L8L	65	64	1,063	-21,996	47	46	L8L	47	46	1,063	-21,996	28	26
L10L	66	64	1,100	-25,220	47	46	L10L	45	48	1,100	-25,220	25	27
L13L	58	59	1,092	-18,000	45	46	L13L	41	43	1,092	-18,000	27	29
L14L	46	48	1,136	-8,760	44	46	L14L	30	31	1,136	-8,760	25	26
L15L	54	54	1,085	-12,350	46	46	L15L	37	37	1,085	-12,350	28	28
L16L	52	50	1,068	-8,980	46	45	L16L	34	33	1,068	-8,980	27	26
L17L	62	64	1,058	-21,390	45	46	L17L	43	45	1,058	-21,390	25	27

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 5				**SAMPLE 5**			**SAMPLE 6**				**SAMPLE 6**	
Lab	**MSC 4**		slope		**MSC 4**		Lab	**MSC 7**		slope		**MSC 7**	
L1L	35,30	35,00	1,07	-24,97	12,91	12,59	L1L	36,00	35,20	1,07	-24,97	13,66	12,80
L2L	25	25	1,037	-8,500	17	17	L2L	23	23	1,037	-8,500	15	15
L3L	28	27	1,114	-15,800	15	14	L3L	26	25	1,114	-15,800	13	12
L5L	39	38	1,092	-24,990	18	17	L5L	35	36	1,092	-24,990	13	14
L7L	28	30	1,084	-14,622	16	18	L7L	26	27	1,084	-14,622	14	15
L8L	35	34	1,063	-21,996	15	14	L8L	34	34	1,063	-21,996	14	14
L10L	36	35	1,100	-25,220	14	13	L10L	34	34	1,100	-25,220	12	13
L13L	32	32	1,092	-18,000	17	17	L13L	30	30	1,092	-18,000	15	15
L14L	23	22	1,136	-8,760	17	17	L14L	20	20	1,136	-8,760	14	14
L15L	28	28	1,085	-12,350	18	18	L15L	25	25	1,085	-12,350	15	14
L16L	26	27	1,068	-8,980	19	20	L16L	23	23	1,068	-8,980	16	16
L17L	34	33	1,058	-21,390	14	14	L17L	31	32	1,058	-21,390	12	13

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 7				**SAMPLE 7**			**SAMPLE 8**				**SAMPLE 8**	
Lab	**FM3**		slope		**FM3**		Lab	**FM15**		slope		**FM15**	
L1L	31,30	31,70	1,07	-24,97	8,61	9,04	L1L	47,40	47,50	1,07	-24,97	25,89	26,00
L2L	18	19	1,037	-8,500	10	11	L2L	32	31	1,037	-8,500	25	24
L3L	22	21	1,114	-15,800	9	8	L3L	34	34	1,114	-15,800	22	22
L5L	33	31	1,092	-24,990	11	9	L5L	50	47	1,092	-24,990	30	26
L7L	23	24	1,084	-14,622	10	11	L7L	34	33	1,084	-14,622	22	21
L8L	31	30	1,063	-21,996	11	9	L8L	46	45	1,063	-21,996	26	26
L10L	32	32	1,100	-25,220	10	10	L10L	46	46	1,100	-25,220	26	25
L13L	26	27	1,092	-18,000	10	11	L13L	40	40	1,092	-18,000	26	26
L14L	18	17	1,136	-8,760	12	10	L14L	27	27	1,136	-8,760	22	22
L15L	22	22	1,085	-12,350	12	11	L15L	33	32	1,085	-12,350	23	23
L16L	20	21	1,068	-8,980	12	13	L16L	29	29	1,068	-8,980	22	22
L17L	29	30	1,058	-21,390	9	11	L17L	43	45	1,058	-21,390	24	26

	Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration			Raw data slope=1 Intercept=0		Data obtained with the calibration adjustment QLIP RM		Data recalculated after calibration	
	SAMPLE 9				**SAMPLE 9**			**SAMPLE 10**				**SAMPLE 10**	
Lab	**FM11**		slope		**FM11**		Lab	**FM23**		slope		**FM23**	
L1L	41,50	40,70	1,07	-24,97	19,56	18,70	L1L	38,90	38,40	1,07	-24,97	16,77	16,23
L2L	31	29	1,037	-8,500	24	21	L2L	25	26	1,037	-8,500	17	18
L3L	32	30	1,114	-15,800	20	18	L3L	30	28	1,114	-15,800	18	16
L5L	42	42	1,092	-24,990	21	21	L5L	41	39	1,092	-24,990	19	18
L7L	30	30	1,084	-14,622	18	18	L7L	28	28	1,084	-14,622	16	16
L8L	41	42	1,063	-21,996	21	22	L8L	37	37	1,063	-21,996	17	17
L10L	41	39	1,100	-25,220	19	18	L10L	39	37	1,100	-25,220	18	16
L13L	33	33	1,092	-18,000	18	18	L13L	33	34	1,092	-18,000	18	19
L14L	23	22	1,136	-8,760	17	16	L14L	22	21	1,136	-8,760	16	15
L15L	32	29	1,085	-12,350	22	19	L15L	28	28	1,085	-12,350	18	18
L16L	26	26	1,068	-8,980	19	19	L16L	25	26	1,068	-8,980	17	19
L17L	38	37	1,058	-21,390	19	18	L17L	34	35	1,058	-21,390	15	15

Grubbs oulier
Grubbs straggler
Cochran outlier