



Determination of fatty acid groups in intramuscular fat of pig by FT-NIRS within the project TREASURE

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Task 3.2: Evaluation of sensory, healthy, technological and typical attributes of regional pork products from untapped breeds and new products issued from local chains

developed by partners within national or European research projects. In particular the fresh meat and pork products submitted to chemical analyses will be utilized to perform NIRS analysis in order to create a regression equation and implement the NIRS methodology as new method to predict meat quality, with special emphasis on prediction of lipid content and fatty acid composition (UNIFI, KIS).

Quantitative chemical analyses used for the determination of FA are:

- a. costly
- b. time consuming

NIRS

- rapid and non-destructive: neither requiring reagents nor producing waste
- widely used **in research** for large-scale **meat quality evaluation**
- already applied to study the FA content in Iberian pig fat, intact pork loins and grounded beef.

		Fatty Acid			Fatty Acid
Animals	Lipid	Fat	IMF		lean
512	270	372	452		197

**Present research
considered 165
fresh samples of:**

Partner	Breed	Sampled
KIS	Krskopolje	5
CICYTEX	Iberico	15
UNIFI	Cinta Senese	20
LUHS	Lithuanian White	24
LUHS	Lithuanian Wattle	9
CSIC	Iberico	5
UNIZG	Turopolje	5
PFOS	Crna Slavonska	16
IRTA	Negre Mallorqui	5
BESH	Schwabish Hallisches	16
UEVORA	Alentejana	10
UEVORA	Bisara	9
IPVC	Bisara	16
INIA	Iberico	5
INRA/IFIP	Gascon	5
Total		165

12 breeds in 8 different countries:



The map shows the following breeds and their locations:

- Schwäbisch-Hällisches** (Germany): A black pig with a white saddle, surrounded by piglets.
- Senojo tipo Lietuvos baltosios** (Lithuania): A group of white piglets.
- Lietuvos vietines** (Lithuania): A group of black and white spotted pigs.
- Gascon** (France): A group of black pigs in a field.
- Krškopolje** (Croatia): A black pig in a stall.
- Bisaro** (Italy): A large white pig.
- Crna slavonska** (Croatia): A group of black pigs in a field.
- Alentejano** (Portugal): A group of black pigs in a field.
- Iberico** (Spain): A black pig in a field.
- Negre Mallorquí** (Spain): A group of black pigs in a field.
- Cinta Senese** (Italy): A black pig in a stall.
- Turopolje** (Croatia): A black pig in a field.



Samples collection: *Longissimus dorsi*

Slices



Grounded



**FT- NIRs
spectra
acquisition**



Analysis with reference methods

**From 3999 a 9999
wavenumber (cm⁻¹)**

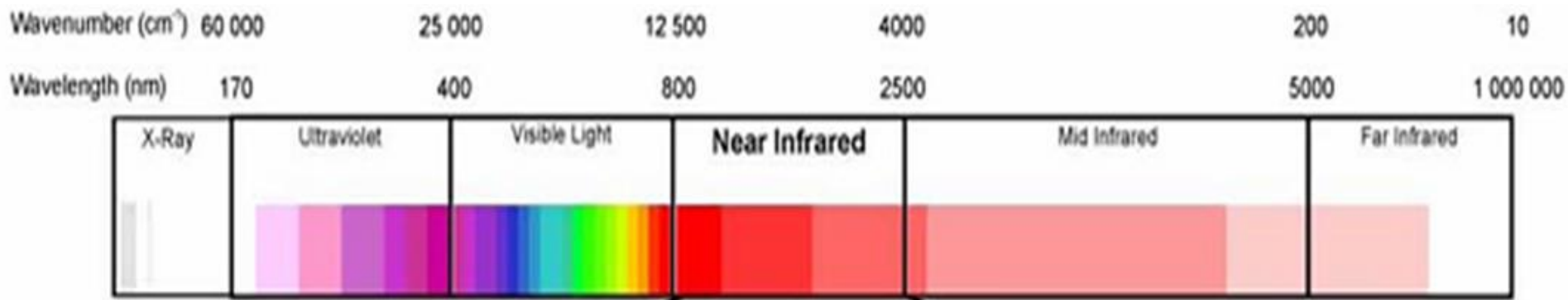
- Lipid were extracted from intramuscular samples
- Fatty Acid Profile through Gas Chromatography



Specific Aim

To evaluate the potential use of FT-NIRS for predicting:

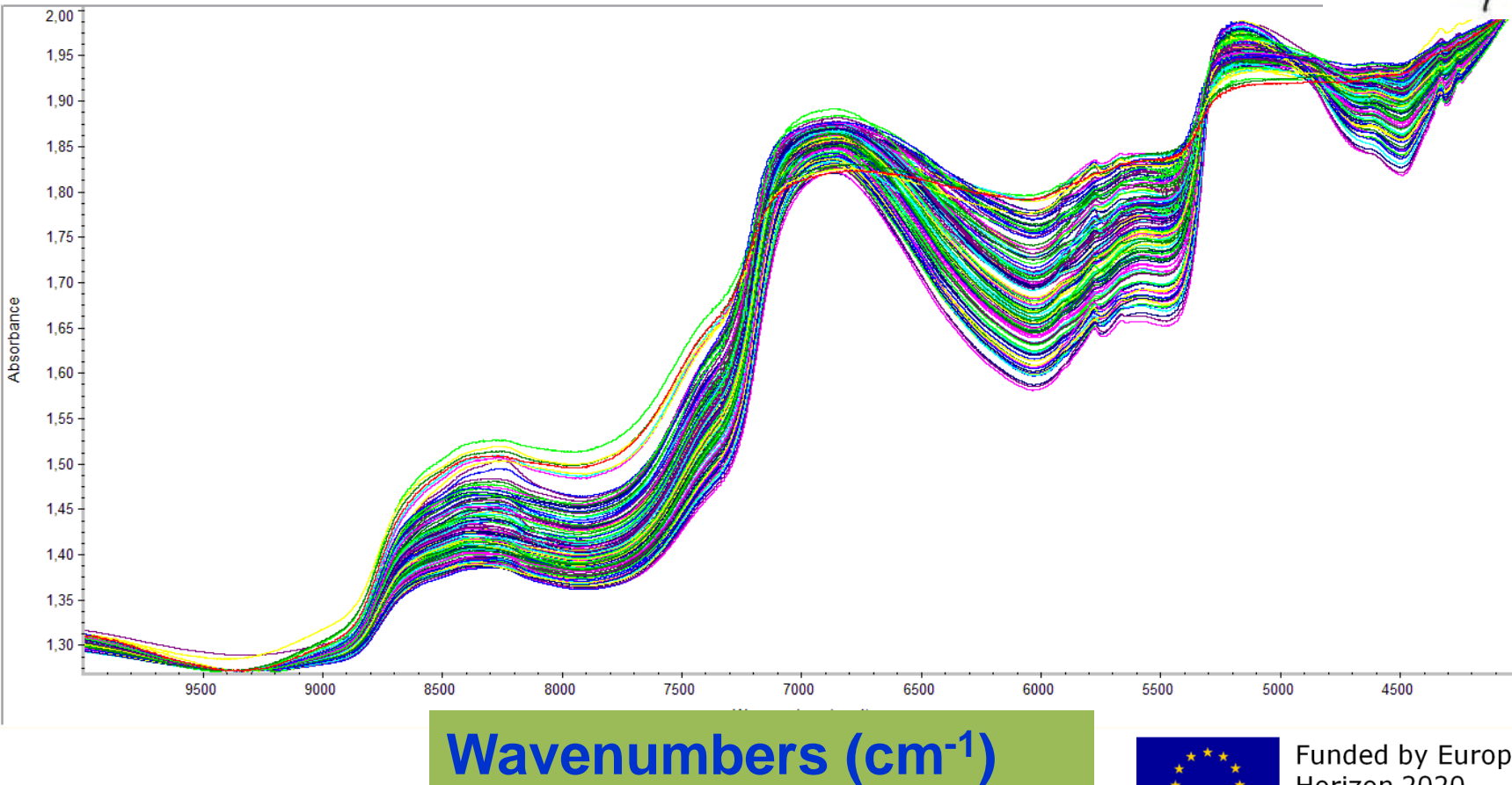
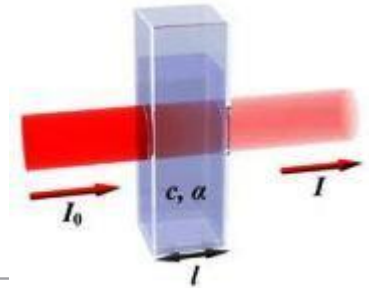
- ▶ Intramuscular Fat **IMF**
- ▶ fatty acids **C18:1n9ct** **C18:3n3** **C20:3 n6**
- ▶ FA groups **SFA** **MUFA** **PUFA**
 - n-6 PUFA** **n-3 PUFA** **n-4 PUFA** **n-1 PUFA**
- ▶ Ratios **PUFA/SFA** **n-6/n-3**



Scan of samples

FT- NIRS (Antaris II):

- 2 scan for each sample (32 scans 16^{-1} cm)
- Absorbance mode



Wavenumbers (cm^{-1})



Mathematical pre-treatments

- Multiplicative Scatter Correction
 - 1st and 2nd derivative
- Smoothing: Savitzky-Golay Filter
 - outliers' spectra

Calibration

Partial Least Square Regression
80% of total samples

Validation

20% of total samples
(randomly selected)

Results

R^2
RMSEC

Coefficient of determination
Root mean square error

R_p^2
RMSECP



Intramuscular Fat and Fatty Acid composition (all units in %)

Parameter n= 165	Mean	SD	Minimum	Maximum
IMF	4.30	2.33	0.56	14.15
SFA	38.71	2.44	31.87	45.61
MUFA	52.48	3.22	43.23	59.45
PUFA	8.26	2.72	3.31	19.50
n-6 PUFA	8.01	2.61	3.24	18.40
n-3 PUFA	0.26	0.14	0.07	1.10
n-4 PUFA	0.01	0.01	0.00	0.08
n-1 PUFA	0.20	0.14	0.03	1.04
C18:1n9ct	42.30	2.45	34.47	48.19
C18:3n3	0.31	0.13	0.11	0.78
C20:3n6	0.16	0.08	0.04	0.58
<i>Ratios</i>				
PUFA/SFA	0.22	0.08	0.08	0.55
n-6/n-3	35.23	11.84	16.69	61.35

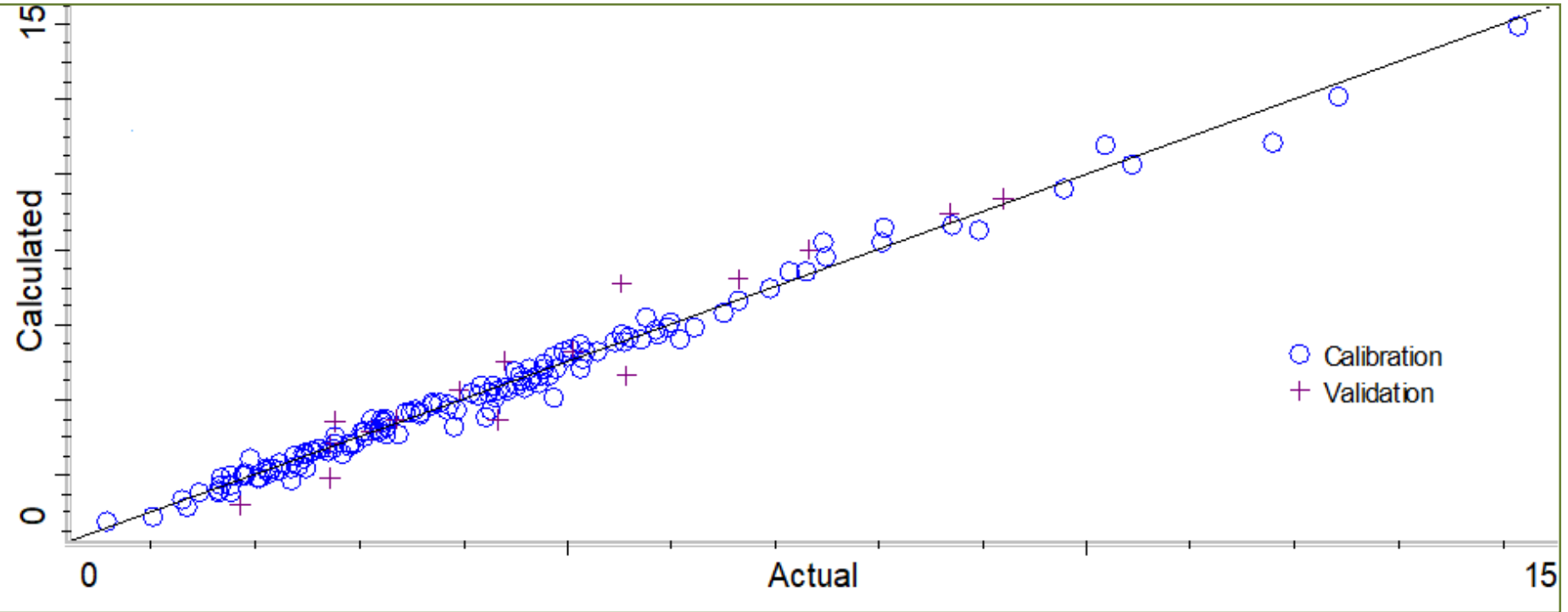


NIRS spectra of grounded meat



IMF

R^2_c 0.994 R^2_p **0.966**
RMSEC 0.245 RMSEP 0.664



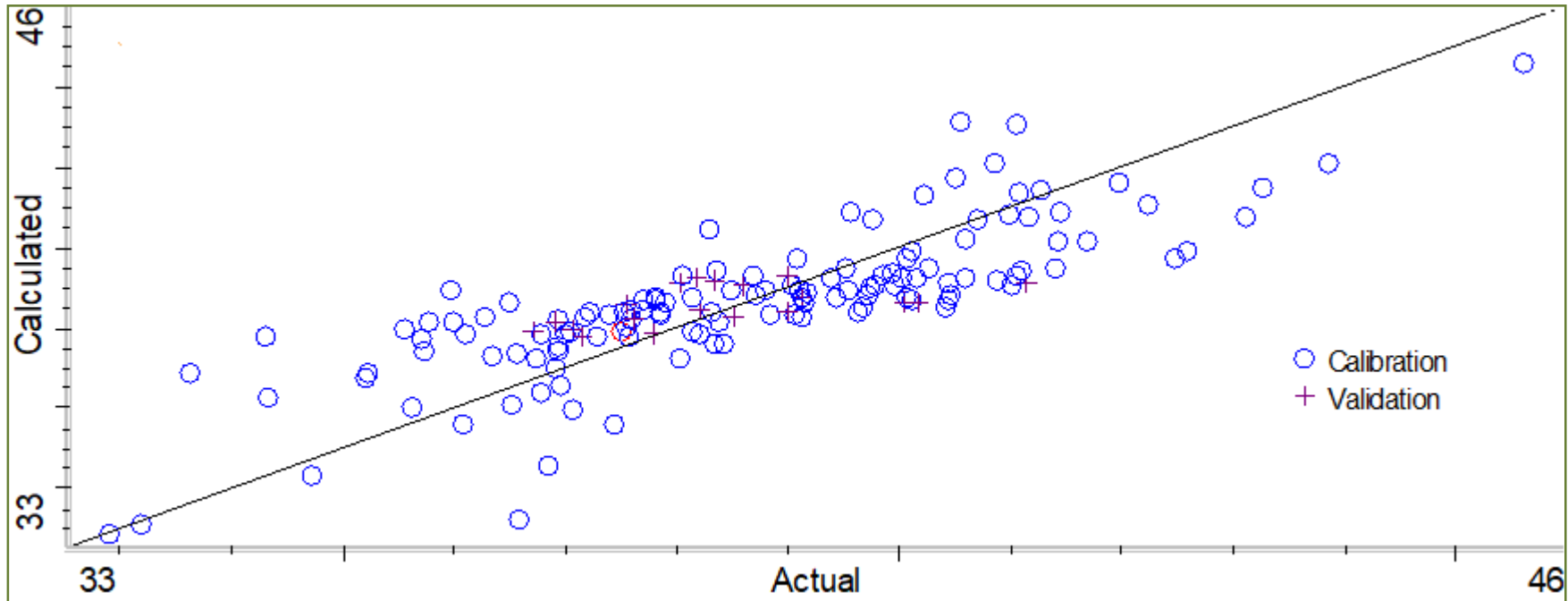
Prevolnik Povše et al. 2017
Pork fresh meat **LD muscle** (n = 75)

	R^2_c	SE_C	R^2_{cv}	SE_{CV}
IMF	0.99	0.14	0.95	0.25



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R^2_c **0.817** R^2_p **0.771**
 RMSEC 1.30 RMSEP 0.975



Gonzalez Martins et al. (2003) in **subcutaneous fat** of Iberian pig

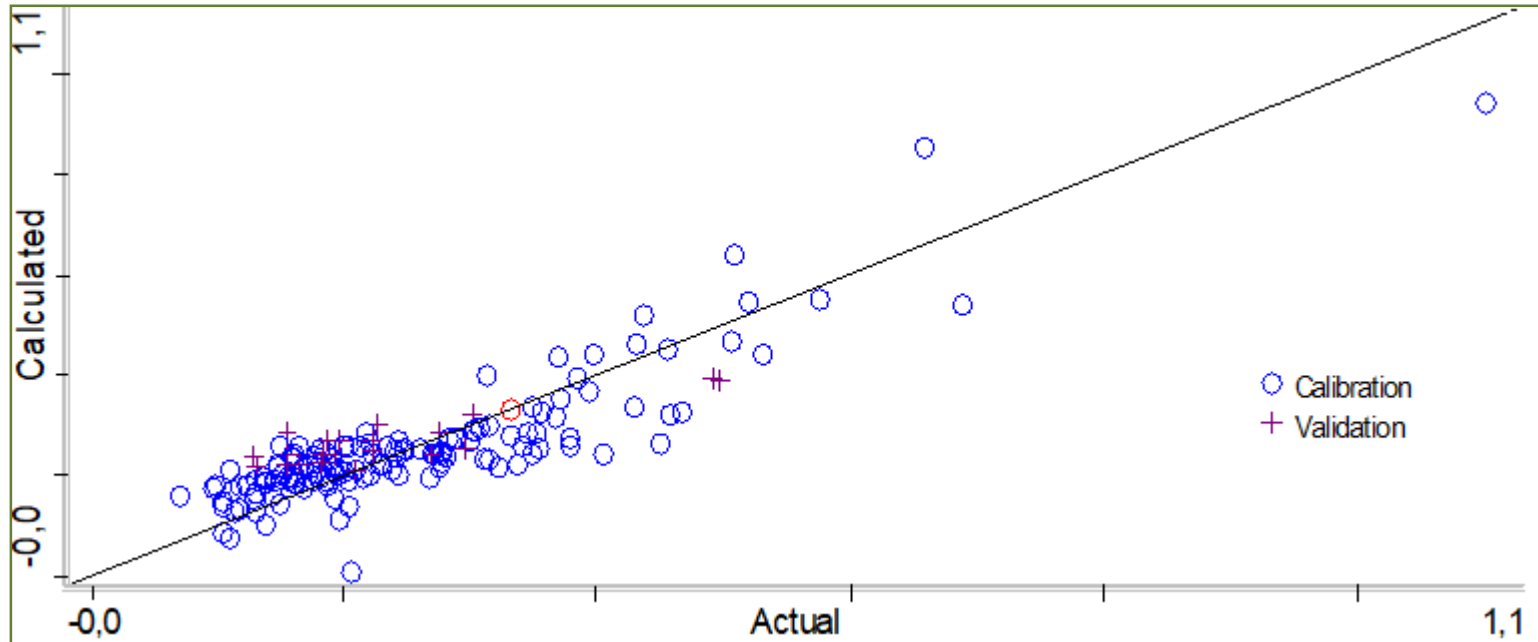
	RSQ	Se _c or SE _p	SE _{cv} SEP _c	Range	n	N. PC
Saturated						
CALIBRATION	0.96	0.86	1.10	22.09-47.31	157	12
EXT. VALIDATION	0.90	1.18	1.18	29.26-40.75	23	



PUFA



R^2_c 0.881 R^2_p **0.872**
 RMSEC 0.066 RMSEP 0.070



Gonzalez Martins et al. (2003) in **subcutaneous fat** of Iberian pig

	RSQ	Se _c or SE _p	SE _{CV} SEP _C	Range	n	N. PC
Polyunsaturated						
CALIBRATION	0.95	0.47	0.60	2.31-14.82	157	12
EXT. VALIDATION	0.88	0.76	0.78	5.44-13.31	23	

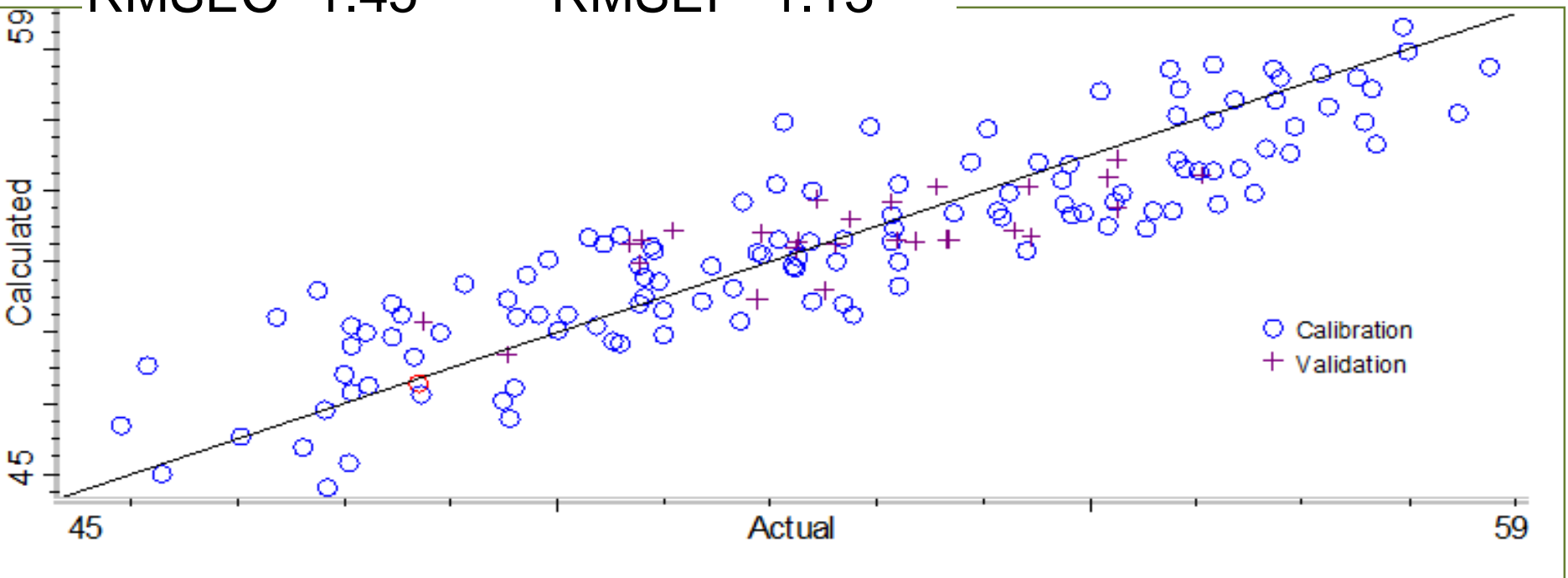


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MUFA



R^2_c **0.894** R^2_p **0.770**
 RMSEC **1.45** RMSEP **1.13**



Gonzalez Martins et al. (2003) in **subcutaneous fat** of Iberian pig

	RSQ	Se _C or SE _P	SE _{CV} SEP _C	Range	n	N. PC
Monounsaturated						
CALIBRATION	0.89	0.98	1.50	47.37-65.62	157	12
EXT. VALIDATION	0.74	1.21	1.23	51.43-60.70	23	



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PLS calibration and validation models

Trait	Calibration		Validation		Math treat
	R^2_c	RMSEC	R^2_p	RMSEP	
IMF	0.994	0.24	0.966	0.66	1;2
SFA	0.817	1.30	0.771	0.97	1;3;4
MUFA	0.894	1.45	0.770	1.13	1;3;4
PUFA	0.881	0.66	0.872	0.70	1;3

Math treatment:

1. Multiplicative Scatter Correction
2. First Derivative
3. Second Derivative
4. Savitzky-Golay filter



PLS calibration and validation models

Trait	Calibration		Validation		Math treat
	R^2_c	RMSEC	R^2_p	RMSEP	
n-6 PUFA	0.754	1.43	0.694	1.52	1;3
C18:1 n9ct	0.768	1.46	0.687	1.19	1;3;4
<i>Ratios</i>					
n-6/n-3	0.824	6.77	0.801	5.94	1;3

Math treatment:

1. Multiplicative Scatter Correction
2. First Derivative
3. Second Derivative
4. Savitzky-Golay filter



PLS calibration and validation models

Trait	Calibration		Validation		Math treat
	R^2_c	RMSEC	R^2_p	RMSEP	
n-3 PUFA	0.756	0.07	0.701	0.06	1
n-4 PUFA	0.721	0.00	0.671	0.00	1;3;4
n-1 PUFA	0.834	0.06	0.648	0.05	1;2
C18:3 n3	0.812	0.07	0.768	0.07	1;2;4
C20:3 n6	0.740	0.05	0.721	0.04	1;2;4
<i>Ratios</i>					
PUFA/SFA	0.704	0.44	0.686	0.37	1;3

Math treatment:

1. Multiplicative Scatter Correction
2. First Derivative
3. Second Derivative
4. Savitzky-Golay filter



Resuming

- ▶ Developed model applicable for IMF quality control
- ▶ Promising results for FA groups
- ▶ Good results even when low proportion of FA
- ▶ Single fatty acid even if highly represented gave poor result
- ▶ Mathematical pre-treatments needed
- ▶ Possible application for Nutritional Label
- ▶ Additional work expected



Thanks for your attention

