Bulgarian Journal of Agricultural Science, 18 (No 4) 2012, 455-481
Agricultural Academy

REVIEW OF *IN VIVO*-METHODS FOR QUANTITATIVE MEASUREMENT OF PROTEIN DEPOSITION RATE IN ANIMALS WITH EMPHASIZE ON SWINE

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Abstract

SIMEONOVA, M. L., N. A. TODOROV and A. P. SCHINCKEL, 2012. Review of *in vivo*-methods for quantitative measurement of protein deposition rate in animals with emphasize on swine. *Bulg. J. Agric. Sci.,* 18: 455 - 481

Most societal important breeding traits have an economic and a no economic value, and are sufficiently heritable for effective genetic selection. Genetic selection goals include the improvement of production traits, such as growth rate (g/day), meat percent (%), feed efficiency, and piglet production, because these traits are economically important. Because of their high relationship with the growth rate and feed efficiency, and the fact that they are inherently related with the animal health and welfare, the traits muscle lean gain (g/day) and protein deposition rate (g/day) are useful measurements. Intensive pork production within large animal populations requires a special care to reduce the negative consequences of animal production on the environment. Growth models can be used to evaluate the nutrient content of the diets fed to minimize nutrient excretion. However, these models require the pigs' compositional growth to be parameterized (Schinckel and de Lange, 1996)

The aim of this review is to compare different methods for *in vivo* quantitative estimation of protein deposition in pigs and other animals. The main criteria for the involving of *in vivo* methods for quantitative estimation of chemical body composition are: a) the accuracy of estimation; b) the financial costs and c) the possibilities for exploitation in field conditions. The chemical analysis post mortem is the most reliable, but it is also the most cost and labor - intensive method of estimation in cases of large numbers of animals. From statistical point of view, the best estimate of the accuracy of estimation respectively prediction equations of composition is the percentages RSD for percent substance (Schinckel et al., 2007; Schinckel et al., 2010b).

The D_2O-dilution technique is relatively economical, since it allows the calculation of the composition of the empty body along with good opportunities for measurements under field conditions with high accuracy. The ultrasonic analysis has been shown to provide acceptable estimation of the protein deposition rate, where the two-dimensional B-mode scans (R^2=0.36 – 0.68) are superior to the one-dimensional A-scan gauges (R^2 = 0.11 – 0.20). The bioelectrical impedance analysis supplies an accuracy of R^2 = 0.81 for the estimation of the fat-free mass of a live animal. The magnet resonance tomography (MRT) has been shown to accurate in the both, indirect (R^2 = 0.89 - 0.97) and direct (R^2 = 0.62) prediction of total body protein. The total body electromagnetic conductance TOBEC accurately predict fat-free lean and protein mass (R^2 = 0.94), showing good prediction capacity, when used in line speeds in combination with growth modeling with optical probe. Corresponding to the stated criteria, the ultrasonic analysis, the bioelectrical impedance, and the D_2O-dilution technique are the most economical and acceptability accurate methods that can be applied under field conditions. The MRT, the X-ray computer Thomography (R^2 = 0.92 - 0.98), TOBEC, and the measurements of the isotope K^{40}, are precise, but too expensive under the most conditions, since their implementation is possible only through specially instrumented equipment, only selection in nucleus farms).

Key words: estimation of body composition, nitrogen retention, pig, accuracy, cost, feasibility

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Abbreviations: BIA – Bioelectrical Impedance Analysis; CT – Computer Tomography; DXA –Dual X-ray Absorptionmetry; MRT – Magnet Resonance Tomography; TOBEC – Total Body Electromagnetic Conductance

Introduction

The selection practice for increased lean growth rate in swine includes the measurement of daily body weight gain (g/day), daily feed intake (g/day), and carcass lean percentage (Schinckel and Richert, 1998; Hermesh et al., 2003). However, the traits feed intake (g/day) and daily live weight gain (g/day) describe the growth of the muscle mass indirectly, whereas the protein deposition rate is a more direct indicator of the muscle growth. Carcass lean percentage largely determines the commercial carcass value. The ingestion of nutrients and energy from fodder, through the feed intake (g/day), is brought into line with the internal impetus for protein growth (Webster, 1993; Barea et al., 2010). Therefore, the actual protein deposition (g/day) is an essential parameter of the compilation of complex growth models (Susenbeth, 1990; Schinckel, 1994 a,b; Susenbeth and Nickl, 1994). Otherwise, the protein deposition is an index for the animal welfare in the sense of the physical and social well-being of animals (Williams et al., 1997; Wellock, 2004).

The inclusion of the trait protein deposition capacity into the practical swine breeding should be gradually carried out by the following means: 1) an estimation of the genetic parameters related with the protein deposition as derived by Hermesch et al., (2003); 2) the identification of the quantitative trait loci's combined with the protein deposition rate (g/day), as assessed by Duthie et al. (2010); 3) assessment of appropriate breeding strategies for increasing of selection efficiency concerning the protein growth capacities of the pig (Knap et al., 2003). Therefore, a quantitative determination of the protein deposition rate (g/day) at a defined economy suited supply of energy and nutrients during the growth are required (Hermesh et al., 2003).

As reviewed and defined by (Hendrick, 1983) as "subjective" methods for quantitative description of the characteristics of "Slaughter animals are routinely evaluated on the basis of dressing percentage, live weight and carcass grades, but neither are precise estimators of composition". One method to estimate composition growth rates is to serially slaughter animals at a series of target live weights and determine the chemical composition of different separated tissues by a chemical analysis (Wagner et al., 1999; Schinckel et al., 2001, Schinckel et al., 2010 a, b). This methods yields the most accurate determination of the body composition and at the same time it is the most expensive and labor-intensive determination (Schinckel and de Lange, 1996; Schinckel et al., 2001, Figure 1). In addition, the dissection of the carcass and the fabrication of the cuts have some level of human error.

Using the "objective methods" for research and description of the bodies of living animal, based on scientifically developed techniques and measurements, referred here as *in vivo* methods, an accurate estimation is achieved. Thus, the methods of quantitative determination of the protein deposition rate in swine can be divided into direct or invasive determination post mortem and indirect or noninvasive determination *in vivo*.

Objective of study is to compare accuracy of the different methods for estimation of protein deposition rate *in vivo*, and their feasibility of application in selection of

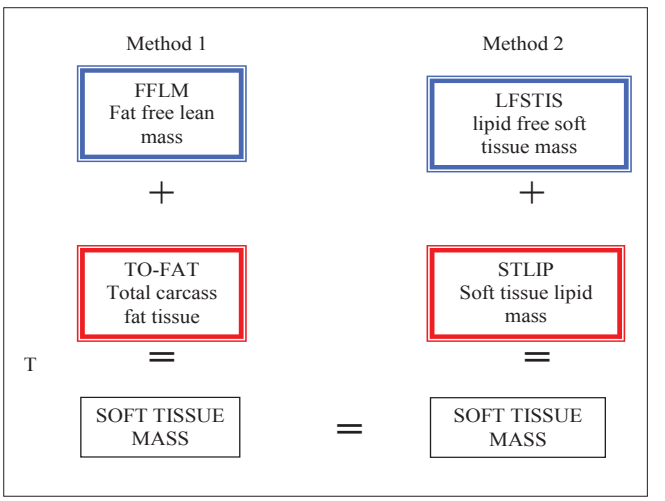

Fig. 1. Methods of separation of the swine carcass into fractions according to (Schinckel et al. 2001).

pigs according this trait. As a basis for representing and estimation of the accuracy are used statistical methods, estimating the confidence bands between the empirical data set and the results of the model-estimation, and characterizing the accuracy of each model (Schinckel and de Lange, 1996).

In vivo determination methods

In general, it is an aim in the breeding practice that any potential breeding animal remains unscathed (Hedrick, 1983). Within the framework of growth examinations, the *in-vivo*-methods allow a multiple determination of the body composition of the breeding animal, without disturbing considerably its pattern of growth. Therefore, the noninvasive methods to determine the body composition are desired for use in selection programs. The alternatives noninvasive methods currently used on pigs for the estimation of the protein in the body include live-animal ultrasonic analysis, N-balance, D_2O-dilution, TOBEC (Total Body Electromagnetic Conductance), X-ray computer tomography (CT), Magnetic Resonance Tomography (MRT), dual energy X-ray absorptiometry (DXA), Bioelectrical Impedance Analysis (BIA), and measurements of the isotope K^{40}.

According to the manner of the protein quantity determination, the *in-vivo*-methods can be split into two groups: 1) methods measuring the protein content in the body (g) and 2) methods obtaining the protein content in the body (g) or the protein deposition rate (g/day) like a derivate from mathematical model using particular numerical *in vivo* measurements like variables (Table 1).

N-balance technique

This method is based on the quantitative determination of the difference between the nitrogen ingestion and its excretion from animals. The nitrogen balance technique is a common method in human medicine and in animal nutrition science. It was used to determine the nutrient characteristics of different foodstuffs, to estimate the influence of differing nutritional regimes on individuals and to ascertain the need of proteins and amino acids in humans (Food and Nutritional Board, 1989) and in agricultural animals (Susenbeth and Keitel, 1988; Susenbeth, 1995; Ilchev 2010a; Ilchev 2010b; Ilchev and Ganev, 2011a, b; Ilchev, 2012).

The nitrogen balance is influenced by a wide range of factors, such as: the level of intake of nitrogen, energy, carbohydrate and fat in the ration; amino acid content and digestibility in the rations and it adequacy to amino acid requirement of animal; age and genotype of animals, accuracy of determinations of nitrogen and other factors (Manat and Garcia, 1992; Ilchev 2010b; Ilchev and Ganev, 2011; Ilchev, 2012). The utilization of the N-balance method in the examinations of Noblet et al. (1987) resulted in an overestimation of 12 % of real nitrogen deposition in the pig body. Quiniou et al. (1995) reported a difference ranging between 5.8 and 7.0 % in order to determine the protein deposition at low and high crude protein content of the diets.

Thyu et al. (2010) reported high probability (P) of determination of the nitrogen deposition by nitrogen balance technique (g/d) amounting to 0.97, SE 2.51 (kg) for the four different means 12.3 g/day, 12.4 g/day, 13.7 g/day, 12.3 g/day for the different foods. The obvious differences in the accuracy of this method of

Table 1
Splitting of the In-vivo-methods for estimation of protein deposition rate (g/day) according to the manner of determination of the protein in the body

Manner of determination of the protein content in the body	Methods of determination of body composition
I. *Direct* determination of the protein content in the body and calculation of the protein deposition (g/day)	· N-balance technique · D_2O-dilution technique · Measurements of the isotope K^{40}
II. *Indirect* determination by ascertaining the *lean portion* and derivation of the protein deposition (g/day)	· Magnet Resonance Tomography · X-ray computer Tomography · Ultrasonic Analysis · TOBEC · Bioelectrical Impedance Analysis

estimation of nitrogen deposition require an additional comparison with nitrogen deposition calculated by a slaughter technique and chemical analysis (as so-called reference method).

Deuterium oxide dilution technique

This method for the estimation chemical composition of body is based on the knowledge of and the interrelation between the total fat-free body substance (protein, ash, water), with water content in the body, which is not affected by the body fat content on the one side, and the total body mass on the other side. The Deuterium oxide dilution technique facilitates the quantitative determination of the total body water content, (kg). The determination of the body water transpires through administering a unique marker (D_2O) and by the taking of a blood sample as soon as the marker has been blended with the body water. The body water volume can be calculated from the ascertained marker concentration and the administered marker quantity, which afterwards will facilitate the calculation of the fat free body substance. The marker disperses readily into the body fluid, is not changed by the metabolism, does not have any harmful side effects, leaves the body unchanged, and is economical (42 € /l, 50 % D_2O solution).

Foot and Greenhalgh (1970) ran D_2O-dilution analysis on sheep with high accuracy. Schields et al. (1983) conducted an experiment (n= 73) to evaluate the efficacy of the deuterium oxide (D_2O) dilution procedure for in vivo estimation of body composition in swine at 18, 36, 52, 80, 100, 120, 145 kg live weight. The D_2O pool measurements accurately predicted chemically determined components (Table 2) with residual coefficients of variation below 5% and $R^2 > 0.90$.

The use of dilution of deuterium oxide (D_2O) to estimate body water in cattle, sheep and pigs is reviewed bibliographycaly by Trenkle (1986), showing that the calculation of body composition from total body water can vary depending upon fill of the digestive tract, and that the applyied multicompartmental models more accurately describes the dilution of D_2O in water from blood after infusion of labeled water, but has not significantly improved estimates of body composition in all experiments.

The repeatability (R^2) represents the accuracy of the estimations of the body composition (empty body water, the body protein, the body ash, and the body fat) of multiparous Holstein-cows (n = 83) with the help of the D_2O dilution technique, covers a range from 0.82 till 0.87 and respectively 0.81 till 0.87, 0.49 till 0.66, and 0.71 till 0.87 (Crooker et al., 1998). Results by Rozeboom et al. (1994) indicate that prediction equations involving D_2O poll, live weight, and (or) back fat thickness as an independent variables are accurate ($R^2 = 0.99$ for Yorkshire*Landrace and $R^2 = 0.87$ for Duroc*Yorkshire*Landrace) in estimating body composition *only* in animals originating from the population, in which the equations were derived. Thus, loss of accuracy and precision of prediction equations can occur when the target population differs from the source population in age, genetics, nutritional background, and physiological state.

Table 2
Predicted chemically determined empty body components according to Schields et al. (1983)

Empty body component	Estimation model
Weight	Live weight
Water	Total D_2O space
Fat (EBF)	EB H_2O live weight
Protein (EBP)	(89.56-0.9(EBH_2O/(EBWT-EBF))
Ash (EBA)	EBWt- ∑(EB H20 + EBP + EBF).
%protein (fat –free)	- 0.91 [% water (fat-free)l + 89.56.

EBF = empty body fat, EBP = empty body protein, (EBA) = empty body ash, EBWT = empty body weight

Susenbeth (1984) used D_2O dilution technique during an examination of 42 castrated male German Landrace pigs with a live weight between 16 and 124 kg in order to determine the protein and fat deposition (Table 3). The results showed an overestimation of the body water content in a range from 0.8 until 3.19 % according to the live weight at the time of taking the blood sample.

The calculation of the composition of the body water in empty body through assistance of D_2O (Appendix, equations 4 to 9), resulted in an error expressed by

Table 3
Calculated overestimation of the total body water content by help of the D_2O-dilution analysis (Susenbeth, 1984)

Test No.	Body mass, kg				Overestimation of the water content of the body alive by D_2O at the time of:					
	At the time of the D_2O administering		empty body		D_2O administering, %		Blood specimen collection, %		Empty body, %	
	mean	sd	mean	sd	mean	sd	mean	sd	mean	Sd
1	17.2	1.16	16.4	1.06	1.71	0.57	1.6	1.73	1.6	0.89
2	39.4	4.17	37.3	3.53	2.74	0.7	2.8	2.81	2.7	0.67
3	66.2	5.67	63.9	5.31	2.83	1.03	3.1	0.62	3.2	0.62
4	88.1	5.79	85.6	5.37	2.88	0.62	3.1	0.68	3.0	0.69
5	116.0	6.85	112.6	6.44	0.81	0.89	1.0	0.75	0.9	0.69

sd = standard deviation

the standard deviation in % of the empty body mass of 1.1 % in relation to the body water, of 1.7 % to the fat-free body substance and to the body fat, and of 1.0 % to the body protein according to Susenbeth (1984).

Rozeboom et al. (1994) conducted an experiment on 58 gilts to evaluate oxide dilution technique and ultrasonic measurements (10^{th} rib back fat thickness) and live weight at slaughter to evaluate oxide dilution technique and ultrasonic measurements (10th rib back fat thickness) and live weight at slaughter to predict the body composition. The average prediction errors were respectively: for the 37 (Duroc*(Yorkshire*Landrace) gilts: 2.37 (%) for the empty body weight; 2.36 (%) for empty body water; 1.07 (%) for empty body protein and 2.76 (%) (For empty body fat and for the other 21 crossbreed gilts: 2.03 (%) for the empty body weight; 1.66 (%) for empty body water; 0.47 for empty body protein and 2.89 (kg) for empty body. Cross-validation by applying of the prediction equations to the data of the other crossbreed animals in the experiment, and to empirical data in the bibliography, resulted in larger prediction errors. However, these results indicate that prediction equations using D_2O space, live weight, and (or) backfat thickness are accurate only in animals physiologically resemble the population in which the equations were derived.

Ultrasonic technique

Animal compositional growth can evaluated with the use of ultrasonic measurements in two ways: firstly, by direct inclusion of ultrasonic measurements in the prediction models of protein deposition rate (Whittemore, 1993); and secondly, by estimation of the lean mass (kg) and the derivation of the protein deposition as a function of lean mass (2.55 g fat free lean per 1g body protein), as described by Schinckel and De Lange (1996) and National Research Council (1998).

According to the bibliographical review made by Moeller (2002), the present developments within ultrasonic technology two variants have to be distinguished: 1) The one-dimensional or the linear measuring impulse echo sounder (sonar) method (A-mode Scan) and 2) the two-dimensional or surface reflecting sectional view method (B-mode Scan). The ultrasonic analysis, which has been practiced for years, led to an essential improvement in the prediction of the body composition of pigs in contrast to the considerably differing results from any other animal. Improved image resolution and quality allowed the lateral and bottom boundaries of the ultrasound image to be more accurately defined and easier to interpret, resulting in improved accuracy and repeatability of the technique (Moeller, 2002). The successful use of the ultrasonic method in the determination of the body composition of pigs can be explained by the repeatability of serial measurements, its high correlation coefficients (r) between the ultrasonic measurements and the real distinctive traits of the carcass **(**Table 4**),** and the accuracy described by the coefficient of determination (R^2) and the remaining standard error (RSE).

The choice and the number of the ultrasonic measurements involved as independent variables of the estimation model, exert an important influence on the ac-

Table 4
Repetition of ultrasonic measurements (Moeller, 2002)

Author	Device	*Accuracy* Repetition (W)/
Krieter et al. (1990)	COMBISON (two-dimensional)	0.53 - 0.74
Busemann (1991)	RENCO (one-dimensional)	0.61 - 0.88
Von Felde (1996)	RENCO (one-dimensional) COMBISON (two-dimensional)	0.19 - 0.91 0.27 - 0.97
McLaren et al.(1989)	Johnson and Johnson Ultrasound 210 DX B-mode scanner	Average R^2= 0.58 AVBF, R^2= 0.39 TRIB, R^2= 0.64 LEA, R^2= 0.53 LNGN, R^2 = 0.76
Moeller and Christian (1990)	ALOKA 500V (Corometrics Medical Systems, Wallingford, CT)	BF 10, r = 0.69-0.82 LMA, r = 0.57-0.68
Schinckel et al. (2010)	An ALOKA 500V (Corometrics Medical Systems, Wallingford, CT	BF, R^2=0.58-0.90gilts BF, R^2=0.64-0.91barrows LD, R^2=0.77-0.87 gilts LD, R^2=0.75-0.90 barrows

AVBF = average carcass backfat; TRIB = 10th rib carcass backfat; LEA = 10th rib loin eye area; LNGN = estimated lean gain/d of age, BF 10 = backfat thickness at 10th rib; LMA = lion muscle area

curacy of the prediction. The estimation models of the muscle lean portion (%, or kg), which already had been described in the literature (Terry et al., 1989; Orcutt et al., 1990; Sather et al., 1991; Gresham et al., 1992a,b, 1994; Liu and Stouffer, 1995; Higbie, 2002) considering the ultrasonic measurements of the back-fat thickness and the *M. longissismus dorsi* reflecting the choice of the variables, provide a coefficient of determination (R^2) in a range from 0.27 till 0.95 and a remaining standard error (RSE) from 0.85 till 3.7.

The two-dimensional B scan devices are superior to the one-dimensional A-scan devices (Busemann, 1991; McLaren et al., 1989; Schinckel et al., 1994). The B Scan provided, on the one hand, the more accurate predictions of the meat concentration in the carcass (g/kg), related to the live weight (kg) and the muscle and fat area (measurements at 13th/14th rib at 100 kg live weight in 153 pigs) on the other hand. It facilitated, also, the prediction concerning the portion of valuable cuts (the sum shoulder, ham, neck and cutlet, kg) measurements at 120 kg live weight via the live weight means and the ultrasonic measurements of fat depth, fat area, muscle area and the computed ratio fat:muscle area at 13th/14th rib producing an coefficient of determination R^2 from 0.36 till 0.61 with a remaining standard error between 24.4 and 19.4 (mm) (Krieter and Kalm, 1991). The examinations of Krieter and Kalm (1991) supported the results of Sather et al. (1987) and McLaren et al. (1989), confirming that measurements at higher live weights (100 - 200 kg) and increasing back fat thickness are more reliable in their forecast and more accurate in their reiteration. Cisneros et al. (1996) compared transverse and longitudinal real-time techniques on live pigs as predictors of lean cut yield and fat-free lean content in the carcass, where small differences were evident, but stated that longitudinal placement of the probe was more accurate when taken anterior to the last rib location compared with a posterior last rib location.

The prediction of the carcass lean deposition (%) with the inclusion of the ultrasonic measurements of the ham thickness (Figures 2 and 3), back-fat thickness and loin eye thickness at last rib provides another appropriate alternative to accurately record (R^2 = 0.69, RSD =2.96, sm) the growth (Blicharski and Ostrowski, 1997).

Jiancheng et al. (2010) showed at that *that post mortem* ham measurements in the pigs could be used accurately to predict the lean portion in the carcass.

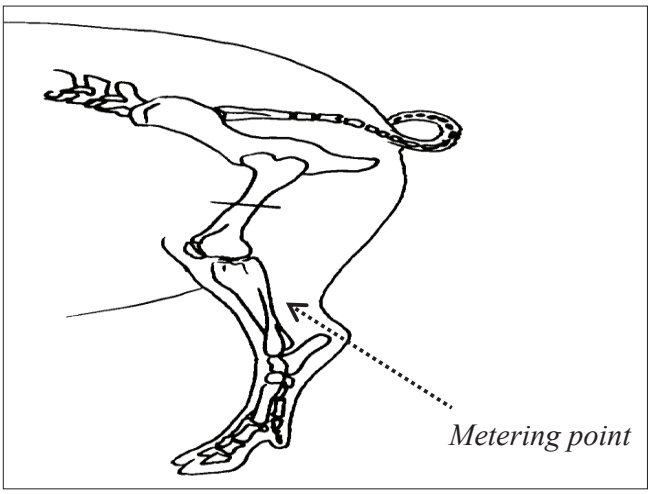

Fig. 2. Measuring point in order to capture the ham depth on the femur by ultrasonic measurements
Source: Blicharski and Ostrowski (1997)

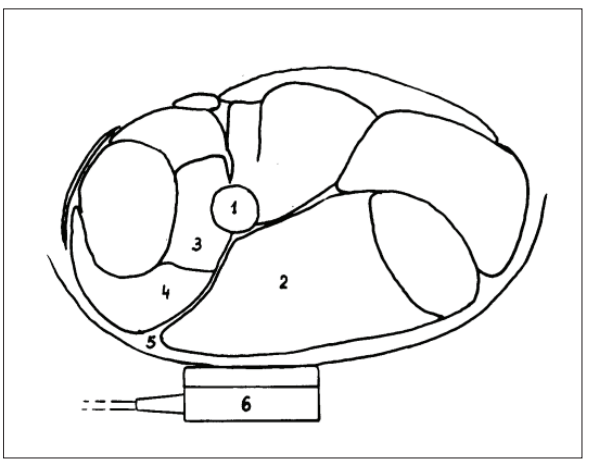

Fig. 3. Metering point on ham - cross-section: 1-femur, 2-Musculus biceps femoris, 3-Musculus vastus intermedius, 4-Musculus vastus lateralis, 5-fat and skin, 6-head of the ultrasonic device
Source: Blicharski and Ostrowski(1997)

The carcass data of dissected lean and fat in the four primal cuts (ham, loin, Boston button and picnic shoulder) were used as dependent variables in establishing regression equations. Carcass lean weight (kg) was best predicted by hot carcass weight (kg), 10th rib back fat depth, and ham lean area (R^2 = 0.92). Ham lean area (cm^2) was related to carcass lean weight (kg) (r =0 .74, P <0.0001) and ham fat area to carcass fat (r = 0.81, P < 0.0001). The results of this study indicated video image analysis of ham cross-section slices combined with backfat depth at the 10th rib could be used for accurate estimation of total carcass lean (kg) or fat (kg).

Additionaly, Schinckel et al. (2010a) conducted an experiment post mortem on 203 barrows and gilts of 7 genetic populations (optical G-100 probe, and Giraldo OPTO-Electronic PG-200 probe) to predict the carcass percentage (%) of 5 alternative measures of carcass composition (fat-tissue-free lean, lipid free soft tissue, lipid-free lean, total fat tissue, and soft tissue lipid, Figure 1) using optical probe measurements (Hennesy grading probe), stated that Optical probe back fat and LM measurements can be used to predict the carcass composition. The optical probe back fat depths were slightly related to lean percentage (r = − 0.82 to − 0.88), highly related to total fat tissue percentage (r = 0.84 to 0.88), and the soft tissue lipid percentage (r = 0.86 to 0.87). Optical probe depths were weakly related (P < 0.05; r = 0.23 to 0.34) to measures of carcass lean percentage and total fat tissue percentage (r = −0.16 to −0.26). Fat-free lean percentage was predicted with residual SD (RSD) of 3.7% for equations including last rib midline back fat thickness, 2.4 to 2.7% for equations including optical probe back fat and LM depth, and 2.3% for ribbed carcass measurements. The carcass data of dissected lean and fat in the four primal cuts (ham, loin, Boston button and picnic shoulder) were used as dependent variables in establishing regression equations. Carcass lean weight (kg) was best predicted by hot carcass weight (kg), 10th rib back fat depth, and ham lean area (R^2 = 0.92). Ham lean area (cm^2) was related to carcass lean weight (r =0 .74, P <0.0001) and ham fat area to carcass fat (r = 0.81, P < 0.0001). The results of this study indicated video image analysis of ham cross-section slices combined with back fat depth at the 10th rib could be used for accurate estimation of total carcass lean (kg) or fat (kg).

Schinckel et al. (2009) demonstrated that live-animal ultrasonic measurements at back fat thickness and loin muscle area could be used to estimate carcass composition. The growth of 1,990 barrows and gilts of 3 sire and 2 dam lines was evaluated over 2 replicates. Pigs were weight and ultrasonic back fat and loin depth measurements were taken at approximate 28-d inter-

vals from 37 kg BW to target BW of 113, 127, or 141 kg. Carcass back fat and loin depths were measured with an optical probe. The ultrasonic back fat depths were affected ($P < 0.01$) by replicate, dam line, and the interactions of replicate × BW and dam line × BW. The ultrasonic loin depths were affected ($P < 0.05$) by replicate, sire line, and replicate × BW and sire line × BW interactions. Carcass back fat and loin depths were affected ($P < 0.01$) by replicate, sire line, dam line, target BW, sex, and the sire line × sex interaction. Pigs of different sire and dam lines have different rates of back fat and muscle growth, indicating differences

Schinckel et al. (2010b) studied the effect of birth BW on pig growth in 1,932 barrows and gilts. The pigs were weighed and measured ultrasonically at approximately 28-d intervals from 74 to 58 d of age. Equations fitting ultrasonic loin depths at 46.7, 64.6, 83.5, and 102.5 kg BW included significant ($P < 0.05$) linear, quadratic, and cubic birth BW variables. At market BW, pigs with birth BW of 1.0 kg had 1% less predicted lean than pigs with birth BW of 2.0 kg. The average daily gain had linear quadratic relationships ($P < 0.001$) to birth BW in the gilts and linear-quadratic- cubic relationships ($P < 0.02$) to birth BW in the barrows. Regression equations predicted that increasing the birth BW of pigs with below-average birth BW had a greater impact on increasing the average daily gain than increasing the birth BW of pigs with average or above-average birth BW. No relationships ($P > 0.60$) were found between daily feed intakes and birth BW. Pigs with birth BW less than 1.1 kg had greater feed: gain, more days to achieve 125 kg, and decreased predicted percentage lean than pigs with birth BW greater than 1.1 kg.

Fortin and Elliot (1985) computed the relationships between back fat thickness and the chemical composition of the total body, whole carcass, non-carcass (offal and viscera), head, perinephric-retroperitoneal and subcutaneous fat depots, and meat (externally defatted and boned ham, loin, picnic and butt) from 48 Yorkshire boars slaughtered at an average live weight of 91 kg. These relationships were then used to estimate how a reduction in back fat thickness would be expected to influence the distribution and partition of protein and fat in the tot a l body and its components. A reduction in back fat thickness was accompanied by a decrease in the fat content of the subcutaneous fat depot and to a lesser extent of the perinephric-retroperitoneal fat depot (126.2 and 20.4 g/g of back fat, respectively), but did not change the quantity of fat in the inter-and intramuscular fat depots (estimated from the meat component) or the non-carcass component, which comprises the mesenteric and omental fat depots. Furthermore, the protein content of the whole carcass and more particularly of the meat component increased at a rate of 29.2 and 19.5 g, respectively, for each millimeter of back fat thickness reduction.

Moeller et al.(1998) recorded serial real-time ultrasonic measurements of backfat and loin muscle area to assess the rate of change per unit of live weight and growth rate at barrows (648) and gilts (459) representing eight major U.S. pure breeds of swine at average live weights of 67.4, 80.3, 93.4, and 104.9 kg. This study showed a need for breed- and sex-specific adjustment factors for rates of back fat and loin muscle deposition in swine. Moeller and Christian (1998) conducted a study (655 purebred barrows and 472 purebred gilts at four mean BW (67.4, 80.3, 93.4, and 104.9 kg) with the following objectives: 1) evaluation of the ultrasonic measurements of back fat and loin muscle area as indicators of corresponding carcass measurement; 2) to compare and evaluate the different measures of accuracy: a) bias, b) absolute deviation, c) percentage absolute deviation, and d) frequency distribution and a measure of variation*: standard error of prediction [SEP]* for the comparison of preslaughter ultrasonic and carcass measurements of back fat and loin muscle; and 3) to compare the accuracy of preslaughter ultrasonic back fat depth and loin muscle area for predicting carcass measurements across sex and breed, and related to the magnitude of the carcass measurements. Real-time ultrasonic measurements of 10th-rib backfat (BF10) and loin muscle area (LMA) were made by a single technician before and at slaughter (Table 5).

Results indicate that the magnitude of the carcass measurement affects both bias and accuracy of prediction for the both made *in vivo* real-time ultrasonic measurements. The SEP statistic is more consistent in evaluating accuracy of ultrasonic measurement than bias, absolute deviations, and percentage of absolute deviation. The degree of bias in LMA related to the size of

Table 5
Relationships between preslaughter measurements of loin muscle area measurement and of back fat measurement and the corresponding carcass measurements, *characteristics of the variation and of the accuracy*, and sex- and breed effects and their interactions (Moeller and Christian, 1998)

Indicators	Preslaughter ultrasonic measurement of back fat	Preslaughter ultrasonic loin muscle area
	depending on the sex	*depending on the sex*
Variation: standard error of prediction [SEP]	*Sex effects* without influencing *Breed effects* were important *Sex effects* without influencing	*Sex effects* were found to influence *Breed effects* were important *Sex effects* without influencing
Accuracy: bias absolute deviation percentage absolute deviation frequency distribution bias carcass measurement	*Overestimated* the carcass measurement by 0.57 mm for carcasses measuring < 24.1 mm and *underestimated* by 2.81 mm carcasses with BF10 > 30.3 mm	*Overestimated* the carcass by 2.35 cm^2 in carcasses measuring < 32.5 cm^2 and *underestimated* by 2.29 cm^2 in carcasses measuring greater than 37.9 cm^2.
Correlations to carcass meassurement	at weights > 63.5 kg are moderately correlated (r = 0.53 to 0.75)	at weights > 63.5 kg are moderately correlated (r = 0.53 to 0.75)
Residual correlations	test, sex, and breed effects BF10 (r = 0.69 to 0.82)	test, sex, and breed effects (r =0.57 to 0.68),

the loin muscle for a particular breed, but no trend for the degree of bias related to the average fat thickness of a particular breed.

Hinson et al. (2009) conducted an experiment, involving live ultrasonic measurements on back fat (10th rib, last rib) and 10th rib muscle area, showing high accuracy (P=0.98 and 0.54). However this accuracy is technician dependent, and the NSIF started programs for standardize ultrasonic measurement for these traits working out scan practicum. This study verified earlier reports that magnitude of the carcass measurement affects the accuracy of ultrasonic estimates.

X-ray computer tomography (CT) and Dual X-ray absorptiometry (DXA)

The *X-ray computer tomography* (CT) is a method to produce cross-section x-ray images. A CT-device consists of three units: the x-ray tube, the detector system and the computer. The digitalized scores of the obtained x-ray absorption profiles are subjected to a number of computational operations (convolution, rear projection) in the computer, converting and capturing the absorption score like an individual picture point of the image. In that way, a digital analog transmitter converts the calculated absorption scores into varying gray scales corresponding to their size. In this way the CT-imaged develops.

By Horn et al. (1997) the *accuracy* of the CT was estimated at a fundamental measuring rate R^2 of 0.92 for the ham-flesh (kg), of 0.91 for the whole ham (flesh and femoral bone, kg), of 0.90 for the total lean mass (kg), and of 0.92 for the fraction of subcutaneous fat (kg) and dermis from 30 live castrated males with a live weight of 100 kg.

The DXA technical originate from the area of nuclear medicine. The original procedure is referred to as *dual photon absorptiometry***,** and initially this technique used the radioactive isotope ^{153}Gd. Consequently, the radioactive sources had been replaced by an X-ray tube (XRT) and scintillation detector ($CdWO_4$ or NaI(Tl) scintillator), mounted on a scanning-arm (Figure 4), so that the examined object is exposed to a pencil X-ray beam scanning in a rectilinear fashion, giving the name of the DXA. The scintillation detector is related to a computer for dual-energy data processing, image display and image-diagnostics, where the body composition parameters can be derived from the image data. This method has found widespread clinical application (scan times are of the order of 2 to 5 minutes, depending on the examination) like an alternative to another,

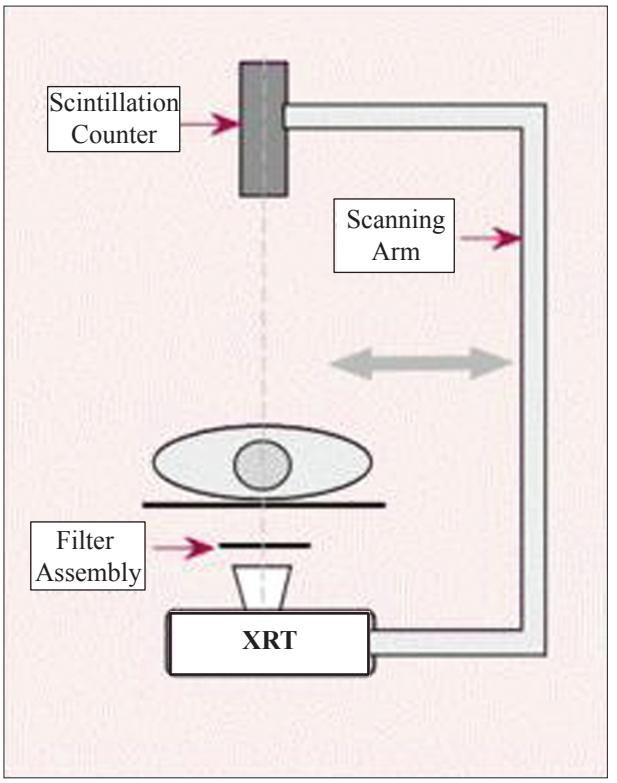

Fig. 4. DXA-Scanner Diagram (source: Wikibooks)

younger image technique, the (Quantitative) Computed Tomography (**QCT/CT**) in terms of accuracy, precision and radiation dose. Within the examinations of Mitchell et al. (1996), Pintauro et al. (1996), pigs with a live weight from 35 to 95 kg had been analyzed by *Dual X-ray Absorptiometry*. This method resulted in a residual variation coefficient from 2.5 to 4.7 % for the adipose tissue and from 0.6 to 2.3 % for the muscular tissue. The essential correlation between the muscular flesh and the fat (kg), estimated with DXA (muscle flesh portionDXA and fatDXA) and the fat, protein, and water content (% fatCHEM, % proteinCHEM, % waterCHEM) which had been determined by a chemical analysis, were confirmed by Mitchel et al. (1998) in examinations of 96 piglets with live weight between 5 and 27 kg and the relations described by the equations (1), (2) and (3):

%fatCHEM = 5.22+0.817*fatDXA;
r = 0.86, SEE = 2.3 (1)

%proteinCHEM= -7.8+0.256*% muscular flesh portionDXA; r=0.935, SEE=2.3 (2)

%waterCHEM=-5.2+0.808*% muscular flesh portionDXA; r=0.59, SEE=3.67 (3)

Mitchel and Scholz (2008), conducted study on 54 females and 43 barrows using dual energy X-ray absorptiometry (DXA) compared with slaughter analysis (at 30, 40, 50 and 60 kg) to measure energy and protein deposition in pigs, fed at four feeding regimen, calculated from DXA lean body mass measurements. The use of DXA for measuring total body fat and protein of pigs within this range in body size has been validated through slaughter and chemical analysis of pigs from this and other studies (Mitchell et al., 1996). Total body protein was calculated from the DXA lean values using the following equation:

*protein (g) = -1.062 + (0.2*DXA lean)*

During growth from 30 to 60 kg the mean value for carcass energy deposition measured by DXA was 251 MJ compared to 249 MJ by chemical analysis (R^2=0.94).

Marcoux et al. (2005) applied dual energy-X-ray-absorptiometry (DXA) to 95 half-carcasses of gilts from three genetic lines of widely varying composition and predicted the dissected lean ($R^2 > 0.85$), fat ($R^2 > 0.70$), bone ($R^2 < 0.66$) and weight of the major primal cuts and the overall carcass ($R^2 > 0.95$). The empirical data showed, that because of small decrease in the prediction error (less than 1.43%), it is not necessary to adjust the prediction equations between genetic lines. In conclusion of further experiments with DXA (Mitchell and Scholz, 2009), DXA can be used to replace the comparative slaughter technique for measuring energy and protein deposition in pigs.

Magnetic resonance tomography (MRT)

The basis for the magnetic resonance measurement is the magnetic qualities of atomic cores with odd proton numbers. Among organic substances, the most common element showing this quality is the hydrogen atom. The MRT procedure facilitates a differentiation of soft tissue without any contrast medium, while the skeleton only appears as a “molding” on the image,

since base on its solid molecular bond, the substance of bones does not reflect any signals (resonance). The estimation of the portion of the individual kind of tissue is carried out by the cluster-analysis. First, the individual scanning elements are assigned to a gray scale range, then the quantity of fat and muscular flesh (kg, or %) is estimated by gradual regression analysis (Scholz et al., 1993; Baulein, 1997). The recent devices provide algorithms for image segmentation and analysis, reconstruction and visualization, and removal of distortions and artifacts for increased image quality and analysis. The MRT-scanned images can facilitate the prediction of the body protein content (kg) as a function of the lean increase, measured *in vivo*.

MRT has been used on several domestic animals: dogs (Thiet and Baulain, 1992), chickens (Janzen et al., 1989), growing lambs (Streitz, 1995), turkeys (Schulte Spechtel et al., 1997), peking ducks, mulards, geese, and musk ducks (Wiederhold,1996), and pigs (Henning, 1991; Scholz et al., 1993; Kallweit et al., 1994; Kastelic, 1997; Baulein et al., 1996, 1998; Westendarp, 1999). Furthermore, an appropriate use seems to be direct prediction of total body protein (kg) (R^2= 0.62) using as independent variables: the volumes (as a percent of body weight) of jowl fat, back fat, shoulder muscle and ham muscle, gained by the MRT and the body weight (kg) as briefly described recently by Mitchel et al. (2001). This approach obtains the total body fat using the fat volume from the 10-cm section of *M. longissimus dorsi* and the fat: muscle ratio from the 15 cm section of the ham (R^2 = 0.90). The prediction of the body composition of live pigs by MRT results in a relatively high *fundamental measuring rate* within a range from 0.89 until 0.96 (Baulain et al., 1996), as shown in Table 6, whereas the MRT-measurement of the carcass at slaughter shows more favorable results (Griep, 1991).

The muscle lean values ascertained by MRT measurement show high correlation coefficients in relation to the muscle flesh portion (Westendarp, 1999). The accuracy of the prediction by this method depends on the live weight of the animal and on the choice of the metering point, which should differ (Baulain et al., 1996). The MRT was carried out at certain metering points (5th/ 6th cervical, 13 th/14th thoracic, 2nd/3rd lumbar vertebra, cranial from the 1st lumbar vertebra and at the caudal end of body weight) of jowl fat, back fat, shoulder muscle and ham muscle, gained by the MRT and the body weight (kg) as briefly described recently by Mitchel et al. (2001). This approach obtains the total body fat using the fat volume from the 10-cm section of *M. longisimus dorsi* and the fat:muscle ratio from the 15 cm section of the ham (R^2 = 0.90). The accuracy of the prediction by this method depends on the live weight of the animal and on the choice of the metering point.

The MRT was carried out at certain metering points (5th/6th cervical, 13th /14th thoracic, 2nd/3rd lumbar vertebra, cranial from the 1st lumbar vertebra and at the caudal end of the coccygeal bone) in 237 pigs, so that the attained accuracy considered and registered the flesh and fat surfaces at any individual metering point with a live weight of 20, 50 and 90 kg. The best accuracy was obtained with a live weight of 20 kg at the metering point 13 th/14th thoracic vertebrata with R^2 = 0.91 for the lean quantity and R^2 = 0.42 for the lean portion. In relation to the fat quantity, it was ascertained R^2 = 0.84. Whenever the live weight is larger, the metering

Table 6
Accuracy of the prediction of the body composition of pigs by Magnet Resonance Thomography according to Baulein et al. (1996)

Trait	Live weight 20 kg N=43			Live weight 50 kg N=40			Live weight 90 kg N=40		
	R^2	SEE	SEE/sd	R^2	SEE	SEE/sd	R^2	SEE	SEE/sd
lean quantity (g)	0.91	190	0.32	0.96	265	0.20	0.89	612	0.36
fat quantity (g)	0.89	90	0.35	0.97	150	0.19	0.91	374	0.34
lean portion (%)	0.55	1.46	0.73	0.83	0.92	0.46	0.87	1.19	0.38
fat portion (%)	0.68	1.06	0.59	0.80	0.97	0.46	0.89	1.01	0.36

SEE = estimation error. SEE/sd = estimation error/standard deviation

point breast (13 th/14th thoracic) is more meaningful, since it determines $R^2 = 0.93$ for the flesh quantity and respectively 0.86 for the fat quantity. Upon increasing the live weight to 90 kg, the metering point femoral thigh facilitates the most accurate prediction of the flesh quantity $R^2 = 0.79$ (Baulain et al., 1996). Besides the relatively high accuracy in the determination of the body composition, the MRT also needs substantial financial expenditures in its instrumental equipment and to the maintenance costs.

K^{40} measurements with the help of Electronic Meat Measuring Equipment, (EMME)

The radioactive isotope K^{40} is a component (0.012 %) of the total body potassium. In a living body, a concentration of 68.1 mmol K^{40} /kg muscle mass can be measured (Forbes et al., 1961). The estimation of the protein mass can be based on the direct metering of the released gamma rays (1.46 MeV) of the isotope. Domermuth et al. (1976) carried out measurements of the K^{40}– isotope with the whole body counter device (*Pacard instrument company, Chicago, Illinois*) in order to determine the protein and meat mass of pigs, as described by Tumbelson et al. (1968). This experiment was conducted using a whole body counter (0.61 m diameter hemicylinder), which had a 2-pi liquid scintillation 6-module detector (length = 2 m) and 2-channel spectrometer. Each module consisted of a 16-guage (1.52 ram.) stainless steel tank with a 0.41 m diameter photomultiplier tube 7 (PMT). Each tank contained approximately 105 I. of scintillation fluid (solution: diphenyloxazole (PPO) plus 1,4-bis-2-(4-methyl-5-phenyloxazolyl)-benzene (dimethyl-POPOP, in toluene equivalent solvent). The whole mechanism may be moved vertically and horizontally to measure-induced radioactivity in man and experimental animals ranging in size from 1 to 635 kg.

The prediction of the protein mass R^2 score of 0.80, was ascertained in when the K^{40}-measurements and the live weight were used as independent variables, where included in a prediction equation.

Bioelectric impedance analysis (BIA)

The principle of the BIA- metering system is founded on the electrical properties of cellular membranes and the intra and extra-cellular water. Lukaski (1985) and Segal et al. (1988) used the bioelectrical impedance analysis on humans.

The recent *in vivo* investigations conducted by Berg and Marchello (1994), Slanger et al. (1994) on lambs and the experiments on beef and cattle by Marchello and Slanger (1994), Marchello et al. (1999a), Marchello et al. (1999b), strengthened the assumptions about high accuracy (Table 7) of estimation, approving BIA as a rapid and accurate nondestruktive technique for monitor animals with greater genetic potential.

In order to assess the applicability to BIA on pigs Swantek et al. (1992) ran examinations on 92 living interbred animals (Du*Ha) with a live weight of 104 kg and on 99 slaughtered animals by a body impedance analysis device (BIA, Model BIA 101, RJL Systems, Detroit, MT) of 800 mA alternating current (Figure 5).

The fat-free mass of the live animal (FFM-L) is computed by regression equations in view of the circumstances of the resistance (RS), the distance between the metering points (L), the conducting capacity (LF), the ratio Vol1 = L2/RS, and the live weight, as an independent variable (pattern 1) with a maximum definable rate of 0.81 (Table 7).

By supplementing the base model (live weight) with the variable resistance and L-length, an accuracy of 0.79 is estimated in relation to the fat-free mass. The BIA facilitates a fast and exact determination of the protein deposition within a period of several years under favorable financial and working conditions. The precise accuracy of the prediction of the fat-free mass of the living animal (FFM-L) according to the first model was confirmed in examinations by Swantek et al. (1999) on 72 crossbred animals (Duroc x Hampshire x Yorkshire) with a live weight of 50, 70, 90, 110, and 130 kg (12 gilts and 12 barrows at each live weight range, Table 8). Predicted fat-free mass (PredFFM) was calculated using the following *multiple regression equation:*

Predi FFM=0.486(live weight)-0.88(resistance) + 0.48(length) +0.86(reactance) +7.959

The correlation coefficients between the actual fat free mass (ActFFM) and PredFFM were stated to vary from 0.66 to 0.91 overall. Correlation coefficients at slaughter weight (90 kg) were 0.94 ($P < 0.02$). It was

Table 7
Accuracy of estimation using *in vivo* BIA in lambs, beef and cattle as referred in the literature

Author	Animal *n*	Accuracy of estimation	
Berg and Marchello (1994)	lambs n=98	fat-free mass for live lambs 77.7% [RMSE] = 1.97 kg) fat-free soft tissue for live lambs 78.6% (RMSE = 1.78 kg)	
Slanger et al. (1994)	lambs n=53	total weight of retail-ready cuts from cold carcass measurements $R^2 = 0.97$	
Marchello and Slanger (1994)	beef cows n=33	<u>total skeletal muscle</u> Live data $R^2 = 0.90$, Exsanguinated carcass data $R^2 = 0.96$, hot carcass data $R^2 = 0.94$ cold carcass data $R^2 = 0.92$	<u>total skeletal fat-free muscle</u> Live data $R^2 = 0.87$ Exsanguinated carcass data $R^2 = 0.93$ Hot carcass data $R^2 = 0.90$ cold carcass data $R^2 = 0.80$
Marchello at al. (1999a)	Beef n=56 Pork n=64	beef trim : $R^2 = 0.80$, Mallows's C(P) = 5.1, and root mean square error = 6.64. fat percentage in beef: $R^2 = 0.84$ and R2 = 0.95. pork trim $R^2 = 0.77$, Mallow's C(P) = 5.0, and root mean square error = 6.2. fat percentage in ground pork $R^2 = 0.87$ and $R^2 = 0.96$	
Marchello at al. (1999b)	Beef Cattle n=50	Mass of saleable products (kg) live carcass $R^2 = 0.80$; Mass of saleable products (kg) in Hot carcass $R^2 = 0.95$ Mass of saleable products (kg) cold carcass $R^2 = 0.93$	

concluded that PrediFFM was underestimated by the prediction equation at all slaughter weights, but the PrediFFM was highly correlated to ActFFM, except for the 110-kg gilts (r = 0.68, P = 0.15) and the 130-kg barrows (r = 0.65, P = 0.16). Thus, overall, the bioelectrical impedance methodology may be used to accurately assess compositional changes of finishing pigs weighing 50 to 130 kg. However, bioelectrical impedance has not been proved to assess body composition of pigs below 50 kg and above 130 kg live weight. Swantek et al. (1999) used the live weight, the length, the reactance and resistance measurements in regression equations and estimated the fat free lean mass with high accuracy amounting to $R^2 = 0.978$ (cp =5, RMSE =2.83). As known, the body weight is accounting for 95% of the variation in FFM, and the addition of length, resistance, and reactance to the equation improved the prediction of FFM ($R^2 = 0.978$, $P < 0.02$).

In order to achieve improvement in the sence of accuracy, Altmann et al. (2004) investigating lambs suggested the use of the impedance at direct current and at a very high frequency and wide frequency range,

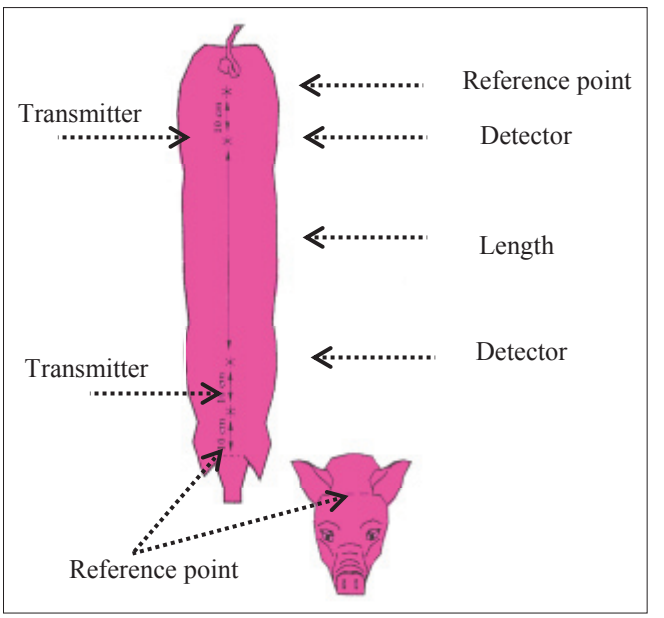

Fig. 5. Points of the bioelectrical impedance measurements on the back of the pig: The cranial point is the base line between the ears and the caudal point is the second caudal vertebra according to Swantek et al. (1999)

Table 8
Accuracy of the prediction of the fat-free mass of a living animal (n = 72) according to Swantek et al. (1999)

Live weight, kg	50		70		90		110		130	
Sex	M	F	M	F	M	F	M	F	M	F
n	12	12	6	6	6	6	6	6	6	6
Relevant FFM, kg	37.0	37.5	50.9	50.8	62.4	63.2	71.0	75.8	80.1	89.9
Predicted FFM-L, kg	30.9	30.2	45.4	45.4	58.2	58.6	66.7	71.1	77.4	83.2
Probability*	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.04	0.32	0.001
Correlation Coefficient**	0.66	0.74	0.74	0.74	0.94	0.94	0.96	0.68	0.66	0.91

Probability * = the probability which is identical with the predicted and actual scores; correlation coefficient** = Pearson's correlation coefficient, n = number of experimental animals. M = male; F = female

extrapolated from the impedance spectrum (*impedance spectroscopy*), compared to the *fixed impedance at 50 kHz*, used in the investigations on BIA described above. Altmann et al. (2004) made evident on lambs, that the *impedance spectroscopy* and time domain-based impedance measurements yield the same result. Investigations on swine should be designed and undertaken to prove the accuracy of the *impedance spectroscopy*.

Total body electrical conductivity – TOBEC

The TOBEC-Methodology was used originally for predicting of human body composition. The measurements are based on the electrical conductivity of the body, respectively, of the carcass. Lean body components (muscle tissue) conduct electricity to a higher degree than "insulative" tissues such as the adipose layers. For the measurements is used equipment as shown in Figure 6, introducing low electrical current into the subject of interest, which emerged from the subject and is measured at that moment. A subject of low conductivity would allow little current to pass through the subject and to be quantificated. Impedance is the inverse to conductance. It is the stopping and (or) slowing of current. So a subject with low electrical conductivity impedes current to greater degree. The TOBEC device is built from a solenoid (tube of coiled wire) and measuring sub-compartments. When the electricity passes through the solenoid, an electromagnetic field (EMF) is created. This field induces current up through the subject, and the device senses the current leaving the subject. The changes in the current indicate whether the subject is highly conductive (high water content like muscle tissue) or not, and the measurements of the current-changes are compiled to characteristics of the body composition.

Fig. 6. The TOBEC device. *Author*: A. P. Schinckel

The Total Body Electrical Conductivity (TOBEC), B - mode ultrasonic and bioelectrical impedance are potential live animal measurements that can be taken repeatedly at different live weights (Schinckel and De Lange, 1996). Berg et al. (1994) showed high correlations between the carcass characteristics and the TOBEC measurements (Figure 6, Table 9).

Schinckel (1994) carried out examinations by the TOBEC - technique on slaughtered animals in order to predict their body protein and their body fat. In relation to the prediction of the empty body protein portion and the fat portion, the definition rates varied within a range of R^2 = 0.84 - 0.86 and, respectively R^2=0.92 - 0.94. Gu et al. (1992) stated in large experiment 181 barrows from five genetic populations R^2 values for the fat free

Table 9
Correlations (r) between the TOBEC measurements and carcass traits according to BERG et al. (1994) (n = 48)

TOBEC measurement	TOTLN	% LEAN	HAMLN	LOINLN	SHLN
PEAK	0.85	0.59	0.80	0.61	0.77
TOBEC (D 7.5-2.5	0.87	0.64	0.87	0.60	0.76
TOBEC (D10-27.5)	0.88	0.67	0.86	0.60	0.76
TOBEC(A130-172.5)	0.86	0.54	0.79	0.60	0.79

TOTLN = weight of total dissected carcass lean; % LEAN = TOTLN/chilled carcass weight; HAMLN = weight of dissected ham lean; LOINLN = weight of dissected loin lean; SHLN = weight of dissected shoulder lean;
PEAK = phase absorbance peak value; TOBEC (D 7.5-25) = difference in phase absorption curve heights at 7.5 and 25 % of the scan curve; TOBEC (D 10-27.5) = difference in phase absorption curve heights at 10 and 27.5 % of the scan curve; TOBEC (A 130-172.5) = area under the curve at 130 and 172.5 % of the s

Table 10
Accuracy of prediction of body composition in swine by TOBEC technology as reviewed in the bibliography

Author	N	Dependent variable	Independent variables	R^2	RMSE
Berg et al. (2002)	54	total carcass lean (kg)	warm TOBEC	0.95	1.31
		carcass lean (%)	warm TOBEC	0.87	1.88
		Ham lean (kg)	warm TOBEC	0.84	0.75
		Loin lean (kg)	warm TOBEC	0.85	0.78
		Boston butt lean	warm TOBEC	0.72	0.50
		Picnic lean	warm TOBEC	0.73	0.46
Higbie et al. (2002)	32	fat free lean (kg)	warm TOBEC	0.91	0.81
		fat free lean (kg)	chilled TOBEC, chilled temperature, carcass length	0.93	0.73
		fat free lean (kg)	Ham TOBEC, chilled carcass side weight, ham temperature, ham butt-face thickness	0.95	0.65

R^2 = coefficient of determination; RMSE = root mean square error

lean in the range from 0.64 to 0.93 for different carcass measurements. TOBEC measurements alone an especially in combination with carcass back fat measurements and warm carcass weight (kg) like independent variables in regression equations resulted in equations with the greatest R^2 values and smallest genetic population biases. Overall, high coefficients of determination have been reported for TOBEC measurements in recent investigations Table 10.

The recent works from Shelton et al. (2003) estimating the carcass composition at 24 barrows with high accuracy for the following prediction equations, involving TOBEC *(TOBEC; model MQI-27: Meat Quality Inc., Springfield, IL*) measurements:

protein *(kg) =[1.13269 + (0.17516 × left side cold carcass weight, kg) + (0.00232 ×* ***PMA****)*
+ (−0.05636 × 10th-rib fat depth,cm)
+ (0.02049 × temperature, °C)
*+ (0.00022369 ×****LMA****, cm²)*
+ (−0.01552 carcass length, cm)] × 2,
where R^2 = 0.91 and RMSE = 0.12
fat *(kg) = [−1.81944 + (0.41374 × left side cold carcass weight, kg)*
+ (−0.11177 × ***PMA****)*
+ (0.08470 × 10th-rib fat depth, cm)
+(−0.00809 × ***LMA****, cm²)*
+ (−0.28161 ×temperature, °C)] × 2,
where R^2 = 0.85 and RMSE = 0.22,

and:
***PMA**= TOBEC measurement, phase maximum average;*
***LMA**=area of M. longissimus dorsi*

Discussion

The practical selection for improved pork production efficiency has resulted in substantially changed pig populations (Pomar et al., 2003; Knap et al. 2003; Van der Steen, 2005). For the improvement of the future selection strategies, a knowledge of the biological mechanisms, as the basis of the changes occurring as result of past selection, is needed, and genetic tests of the biological traits underlying the post-weaning performance traits have to be carefully analyzed (Schinckel, 1999).

The traits per cent lean, feed intake, feed conversion and daily live weight gain, characterize the common target of *the trait growth capacity* indirectly. The ingestion of energy from nutrients for the animal, corresponding to daily feed intake (g/day), is directed toward the *impetus for protein growth* (Webster, 1993). Therefore, the biological trait protein retention (g/day), underlying the growth performance, has to be genetically evaluated. This requires the quantitative determination of the protein deposition rate (g/day) in the genetic population of interest.

Muscle lean growth depends on a large spectrum of endogenous and exogenous factors, which interact with each other (Wellock et al., 2003a; Wellock et al., 2003b; Wellock et al., 2004). The endogenous factors (sex, genotype, live weight and age) pre-determine genetically the potential for visceral organ growth and for muscle growth, and thus, for protein deposition. In general, during growth, the exogenous factors (pen density, climate, energy and nutrient supply, immunizations, diseases) cause a limitation of this genetic limit for growth and in this way modulate *the actual rate of protein growth.*

When the limiting factors are only the endogenous, the achieved actual muscle growth rate, and respectively the actual protein deposition rate, are the muscle growth capacity, and respectively *the protein deposition capacity.* Thus, the protein deposition is constantly related to the visceral organ growth, and the muscle growth, when all other factors are non-limiting. Although, based on this theoretical ground and available experiments in the bibliography, it is to be considered that, the trait of actual muscle lean and the achieved actual total protein deposition in the empty body have a great degree of relationship to each other, however may not be always an adequate pattern of the growth capacity.

Moreover, health problems, discrepancies from the nutrient requirements and non-optimal stocking conditions can affect the achievement of the maximum muscle gain and respectively the maximum protein deposition (g/day) largely than the visceral organ growth. Therefore, the experimental estimation of protein deposition capacity of a given genotype must to be carried out under non limiting environmental conditions. Additionally, model-simulations, qualifying the magnitude of the environmental factors (social, physical, and nutritional environments) on pig food intake and performance are discussed by Wellock et al. (2003c). Wellock et al. (2004) extended the model, involving the between-animal-variation in growth potential and on performance, initial state, and ability to cope when the animals were exposed to environmental stressors. The feasibilities for *in-vivo*-implementation as a target into the practical use in selection programs (the high accuracy, the economical utilization and the opportunities for use under field conditions) are the common reasons to use the noninvasive methods for determination of the body composition in the pig.

In general, the *ideal method* to determine the body composition should fulfill the following conditions: universality, guarantee of no reduction of the retail price of animals ready for the slaughter, no contact with radioactive and toxic substances, as little labor as possible, simple technical equipment, possibility of a flexible labor transfer to unskilled personnel in relation to the routine examinations and shortened operating time (Susenbeth, 1984; Trenkle, 1986; Forrest et al., 1989).

The measurement method should accurately predict the actual differences between pigs and genetic populations of pigs in composition. In this sense, the measurement devices must describe the animals´ characteristics adequately, i.e. there must be obtained a high degree of relationship between the true and predicted genetic

population means. This means that the measurement instruments must be able to predict the real variation between the genetic population's means (Schinckel and de Lange, 1996). There is a basis of individual pig accuracy (how accurately the body composition can be estimated), which is described through the R^2 (repeatability) and the residual standard deviation (RSD). According to Schinckel et al. (2010b) and Schinckel et al. (2007), the best estimate of the accuracy of estimation of composition is really the percentages RSD for percent fat-free lean.

Protein deposition is highly correlated to carcass lean growth, within *non- limiting environmental conditions*. On the basic of this relationship, the prediction related to lean quantity through a coefficient, computed on the base of empirical data, is obtainable (protein deposition rate (g/day) as a function of the fat-free lean gain on the basis of the assumption of 2,55 g fat-free lean per 1 g body protein the live weight range 20 to 120 kg (Susenbeth and Keitel, 1988; NRC, 1998).

As reported in the literature reviewed above, the daily lean growth rate can be estimated from the *in vivo* measurements. Thus, this derivation approach needs an additional estimation of accuracy for each *in vivo* method used. If the accuracy of derivation of the protein quantity is needed to be evaluated and validated, a chemical analysis must be accepted as the reference method.

Via second approach of the determination of the magnitude of protein deposition, the protein quantity estimations, conducted at slaughter, are allowed to be used in future predictions, in relation to the various *in vivo* measurements on the same animals, which had shown high correlations to the protein content or the lean in the carcass, as shown by Schinckel et al. (2001).

A third approach is to use the relationship between the total body protein content and the carcass measurements post mortem. It has to be taken into account that some carcass measurements tend to overestimate the lean content of the genetic populations with lowest lean percentage and to underestimate it for the leanest lines, as statistically considered by Gu et al. (1992).

From a practical point of view, in the all approaches, the protein determinations are made under specific non-limiting environmental conditions, related to a particular genotype. This results in the application of the gained estimation models only to the given conditions.

The chemical analysis of physically dissected and ground whole bodies

By slaughter based on a step-by-step plan the animals are slaughtered at certain phases in their lives at particular live weight of interest, as reported by Walstra (1980), De Greef (1992), Bikker (1994), Susenbeth (1995), Wagner et al. (1999) and the chemical body composition is ascertained by a chemical analysis as proposed by (AOAC, 1990). Chemical analysis yields the most accurate determination of the body composition and at the same time it is also the most expensive and labor-intensive determination (Schinckel, 1994 b; Wagner, 1999). Fortin et al. (2004) and Schinckel et al. (2010) showed that the optical probe at slaughter (Henessy grading probe) is an accurate invasive method for carcass measurements.

The accuracy for the chemical composition of the separate tissue fractions of the carcass half depends on the practiced accuracy of the fine dissection of the carcass, and the pattern of separation of the carcass into *fractions and subfractions from interest*. The common aim is to get live weight and over target information before, during and after the live weight range from laboratory animals, which do not suffer under stress. The accuracy of estimation for lean mass, and respectively-protein mas, is higher, when the live weight of interest is greater (Schinckel et al., 2001). In this respect, further research is needed in order to universalize the estimation models for large groups of variable groups of animals and to adjust for sex (Hicks et al. 1998) and genetic population (Gu et al., 1992) biases.

The *in vivo* methods

Analyzing the reviewed bibliographical data, and the data collected in Table 1, it can be concluded that in relation to the accuracy of any other methods, the first group can be classified as follows: the ultrasonic technique with $R^2 = 0.11 - 0.68$, MRT with $R^2 = 0.62$, the K^{40}-metering with $R^2 = 0.80$, the N-balance technique with an overestimation variability from 5 till 20 %, and, and BIA $R^2 = 0.81$. In addition to of the precise

accuracy of the D_2O-dilution technique and the K^{40}-measurement, these approaches can be done quickly and any side effects are insignificant. A direct relation of the bioelectrical measurements and the body protein content, obtained by chemical analysis in the prediction model, would result in a high accuracy of prediction. An essential peculiarity of the determination of body protein by the D_2O-dilution technique, is, that it requires a long operational time and conditions, which is comparable with the N-balance technique.

D_2O-dilution technique

The D_2O-dilution technique shows additional advantages through low financial expenses and the possibility to transfer the laboratory work to personnel without any special skilled training. The application of the marker D_2O *per os* allows a simplification of the laboratory work through this method under field conditions and reduces significantly the costs, although, the biases of estimation by the D_2O dilution technique are obvious, due to the manner of application and concentration of the marker, the different approach to the calculation of the amount of body water, the involvement of the animals waste products during the measurement, and the time interval between the blood sampling and slaughter. Additionally, endogenous, genotype and sex depending factors, such as the quantity of the total body protein, total body fat and growth intensity, effect the accuracy. This bright spectrum of factors implies the need for further research in order to estimate the accuracy of this method and adjust the biases. Landgraf et al. (2006) conducted an experiment at Pietrain swine (n = 172) at target body weights of 30, 60, 90, 120, and 140 kg and stated the accuracy of the D_2O dilution technique.

N-balance technique

On closer examination, for a practical land utilization of the N-balance technique for determination of protein deposition, essential labor and financial expenditures have to be considered (150 € /animal). In comparison to slaughter analysis, the N-balance technique results in a smaller standard deviation of the retention means, when calculated over a limited period (< 3 months), as stated by Everts and Dekker (1994) for sows.

Electromagnetic technology, TOBEC

The high accuracy of prediction of carcass component composition and total carcass composition by electromagnetic technology has been collaborated by the recent experiments of Swan et al. (2001) (n=64, crossbred growing-finishing barrows at weight range of 40-115 kg.), where the prediction of the lean content of hams by TOBEC resulted in an $R^2 = 0.80$. Lean prediction of loins by TOBEC resulted in an $R^2 = 0.66$. The R^2 - values from the regression analysis predicting protein, moisture, protein + moisture, and fat-free soft tissue composition of the skinless bellies were R^2=0.67, R^2=0.68, R^2=0.71, and R^2=0.78, strengthening the results from Williams et al.(1994). These results are similar to the results obtained by Berg et al. (1994) and Berg et al. (1997) made on lambs. Prediction equations involving TOBEC measurements at the ham at slaughter (n=260, body weight amounting to 84.3 kg) were worked out by Apple et al. (2004) to calculate ham fat-free lean composition, and could not be presented because they are the intellectual property of Cargill Red Meat Sector.

It is to conclude, that the electromagnetic prediction of lean tissue weights possessed higher R^2 than equations predicting proportional yield (%) with these same parameters. This information can be utilized to establish a component pricing system, allowing for payment of the actual lean value associated with ham, loin and shoulder lean (Berg et al., 2001; Hermesh et al., 2003).

In favor of that outlined above, the TOBEC-measurement of the warm (r = 0.95) and chilled carcass (r = 0.93) are highly correlated with the fat free lean in the carcass, as reported by Higbie et al. (2002) in collaboration of the investigations by Berg et al.(1994), where high correlations between the warm TOBEC measurement and carcass lean (r = 0.93) and between the chilled carcass and carcass lean (r = 0.86) were indicated. Moreover, the TOBEC – means of the untrimmed ham were also found to be highly correlated with the fat free lean (r = 0.95). This mutual relationship explains the high accuracy of prediction ($R^2 = 0.80$) of the ham lean by the primal weight and the TOBEC – measures, as reported by Swan et al. (2001). According to the same author, the accuracy of prediction of the loin lean amounted to $R^2 = 0.66$. As shown in Table 9, the varie-

gated TOBEC measurements are highly correlated with the total carcass lean, the lean in the ham, and the lean in the shoulder, when measured at line speeds.

Similarly, middle to high correlations between the optical grading probe measurements (fat depth 3rd from the last rib) and the total carcass lean (0.42), total fat (0.83) and fat depth, measured by hand (0.91), were stated. Alone the optical probe has a small predictive capacity compared to TOBEC (R^2 = 0.22-0.67, RMSE = 2.92 - 4.45 for the total carcass lean) as stated by Berg et al. (1994). A combination of both in a model would allow the achievement of high prediction accuracy. In this respect, the estimation of carcass value through optical probe has to be carried out by an experienced operator, in order to minimize the measurement errors (Boland et al., 1995).

In general, measurement errors in the carcass, when used to develop prediction models, determine the accuracy of the model, and thus the practical application (Lofgren et al., 2000). Additionally, the involvement of measurements from partial dissection of the carcass would be biased, if the distribution of the lean in the primary dissected cuts varies (due to maturation or the effect of a major gene such as the stress gene). Besides the advantages of this technique: the potential implementation on living animals, low maintenance costs, no side effects in relation to the labor personnel and to the examined animals (the electromagnetic field is lower than 1/100 of that of a TV set), the costs of the TOBEC appliance are about 115,000.00 €.

The *in-vivo*-methods described above are comparable in their accuracy with the video image analysis (VIA) *post mortem*, as presented by Jiancheng et al. (2010), where video-images of ham cross-sections were recorded on 71 pork carcasses (in weight range from 72 to 119 kg) in order to predict with the help of regression equations the pork carcass lean and fat composition from: a) video image analysis (VIA) of ham cross-sectional area measurements; and b) 10th rib back fat depth; and c) and hot carcass weight. Carcass data of dissected lean and fat in the four primal cuts (*ham, loin, Boston button and picnic shoulder*) were used as *dependent variables.* Carcass lean weight was best predicted by hot carcass weight, 10th rib back fat depth, and ham lean area with R^2 = 0,92, whereas ham lean area was related to CLKg (r = 0.74; P < 0.0001) and ham fat area to CFKg (r =0.81; P <0.0001). The results of this study indicated video image analysis of ham cross-section slices combined with backfat depth at the 10th rib (mm) could be used for accurate estimation of total carcass lean or fat (kg) composition.

Bioelectrical impedance analysis

As stated in the studies of Swantek et al. (1992) and Swantek et al. (1999), the bioelectrical impedance analysis shows a high accuracy for the estimation of the fat – free mass *in vivo* and further allows for the calculation of the protein content (kg) and protein deposition (g/day). This high accuracy is comparable with that obtained by Velazco et al. (1999) amounting to R^2 = 0.99, SEE = 0.63 at age of 3 months and R^2 = 0.99, SEE = 0.26 at 6 months, R^2 = 0.97, SEE = 0.62 at 9 months and R^2 = 0.77, SEE = 0.28 at 12 months for 53 Holstein steers. Similarly, BIA predicts the fat content of pork with high accuracy as stated by Marcello et al. (1999). In each of the presented prediction models, different impedance information was taken as predictors (as independent variables) and the reported accuracy was obtained.

Therefore, further investigations into the evaluation of the best predictor at particular live weights of interest have to be conducted, and the group biases estimated. Another manner of prediction is visible in the direct relationship between the chemical estimated protein in the carcass and the BIA – measurements in combination with the live weight in the basic model. Altmann et al. (2004) made questionable that investigations on swine should be designed and undertaken to prove the accuracy of the *impedance spectroscopy*.

Ultrasonic measurements

As reviewed in Table 4, the carcass measurements are highly correlated with the ultrasonic measures. Therefore, the live ultrasound measurements are good predictors of the actual carcass measurements and body composition. However, the prediction equations from the live animal serial ultrasound data will differ in accuracy, since there exist variations due to measurement errors (Lofgren et al., 2000). As known, the equations must be developed with the smallest possible level of

measurement error. Schinckel et al. (2007) showed that, level of genetic population biases increase as the level of measurement error increase (R^2 of the equations decrease, the RSD's values increase).

The area of application of the ultrasonic analysis dilates by considering the protein deposition capacity (g/day) of the modeling requirement for amino acids (Smith at al., 1996). The ultrasonic analysis shows a variable application for the purposes of protein deposition estimation, allowing the *in vivo* prediction of the protein mass in the pig according to two methods, as reviewed in the previous paragraph. Thus, the efficient involvement (n=469, R^2 = 0.62 – 0.64) of live ultrasonic measurements and live weight in the prediction of fat free lean of the body, as considered by Higbie et al. (2002), increases the flexibility of the calculation of the protein gain, as a function of the fat free lean gain.

Although there are well - known advantages to the ultrasonic analysis (the simple handling, fast work completion, mobility, modest costs, and the lack of any negative side effects in relation to the labor staff and the experimental animals), it is well to point out that high accuracy of the predictions, is not always obtained, depending on the metering devices, on the point of measurement and on the metering person (Krieter et al., 1990, Schinckel et al., 1994). The application of transverse and longitudinal measurements for the prediction of fat-free lean content in 80 pigs in the live weight range 108-148 kg did not result in any improvement of the accuracy of estimation, as concluded by Cisneros et al. (1996). In this study, the involvement of more than one scan measurement as an independent variable in the prediction - model did not increase the accuracy, as previously stated by Krieter and Kalm (1991).

The discussion of the application of ultrasonic analysis for estimation purposes allows one to point out that a reference estimation of the ultrasonic measurements is required, because of the fact that underestimations in leanest animals are indicated, as described by (Schinckel et al., 1994) for the loin eye area in large growth trail in 1993. This trend corresponds to the findings of Moeller and Christian (1998) of 655 purebred barrows and 472 purebred gilts for the back-fat at the 10th rib, where the ultrasound measurement overestimated the carcass measurement by 0.57 mm for carcasses measuring < 24.1 mm and underestimated by 2.81 mm carcasses with back-fat > 30.3 mm and overestimated the loin muscle area carcasses by 2.35 cm^2 in carcasses measuring < 32.5 cm^2 and underestimated by 2.29 cm^2 in carcasses measuring greater than 37.9 cm^2. The advancements in ultrasonic technology allow for the extension of the range of points for ultrasonic measurements used conventionally in the breeding practice (back-fat thickness at 13/14 rib, 10 cm behind the shoulder and 10 cm in front of the ham, 6 cm from the dorsal mid line; the loin eye depth at the same positions; loin eye area and fat area at 13 / 14 rib).

Schinckel et al. (2010a, b) stated that the addition of *in vivo* ultrasonic measurements (lion depth, and back fat thickness) in regression equations to the birth weight, pig's growth variables and lean percentage allowed prediction of the point of achievement adult live weight, and the corresponding feed intake pattern. The very recent investigations from Jiancheng et al. (2010) on ham in swine are collaborate the research from Blicharski and Ostrowski (1997), indicate high correlation between the post mortem measurements on ham and the in vivo measurements on ham. Future investigations about the accuracy of ultrasonic measurements on ham, and factors affecting the accuracy at this measurements point are needed. The experiments sited above are based on the initially findings by Schinckel et al. (2001) describe the technology of separation of the carcass into fractions and the different alternatives for in vivo prediction of carcass composition in swine via ultrasonic measurements at the back fat thickness and lion area.

Following the above presented high correlation coefficients in relation to the lean portion in the ham (kg) and the lean content of the carcass side (kg) and the significant correlation between the lean portion of the ham and the lean portion in the basic cuts on the one hand, and the relatively high correlation coefficient r_p = 0.67, obtained by Blicharski and Ostrowski (1997) between the *in-vivo* ultrasonic measurement of the ham thickness 5 cm above the knee and the muscular lean portion (%) in the carcass of the over side, it can be concluded that the inclusion of the ultrasonic measurement on the ham thickness (mean 6.65 cm, s = 1.16 cm) in the prediction of the lean and protein deposition rate is a justified alternative for extended growth recording.

The new gained experimental results by Schinckel et al. (2001), Gu et al., (2001) and Higbie et al. (2002) about the relationship between the serial in vivo ultrasonic measures Ultrasonic measurements: 1) the loin eye at the 10th rib; 2) for backfat depth, 7 cm off-midline; 3) at the 10th rib backfat depth and last rib) and the accuracy of prediction equations for lean mass (kg) or protein (kg), developed at different body weight ranges (carcass weight) are improving the growth modeling, especially involving the interrelations between the sex and backfat-thickness-measures (mm) at weight stage, and the interrelations between the genetic population and backfat-thickness-measures (mm) at weight stage. There are 8 different sets of accurate prediction equations for the fractions of the body (carcass lean (fat tissue-free lean; lipid-free soft tissue), dissected lean in the four lean cuts), fat (total carcass fat tissue), and lipid mass (soft tissue lipid), (Figure 1) as described by Schinckel et al. (2001), each of them developed and valid for very specific weight of interest. This could be explained with the interactions weight*sex, weight*genetic population for every ultrasonic measures, involved like independent variable in each of the 8 prediction equations, where the ultrasonic technic is applied at each 20 to 30 kg body weight intervals within the whole growth period, starting at 20 and finishing at 120 (152 kg) live weight.

Magnetic Resonance Tomography (MRT)

As stated by Baulein et al. (1996), the varying points of the MRT sectional measurements allow for varying accuracy amounting to R^2 = 0.89 - 0.91, SEE = 190 for the lean weight (g) and R^2 = 0.89 - 0.97, SEE = 90 for the fat weight (g). These results are comparable with these obtained by Mitchel et al. (2001), where the body lean (R^2 = 0.88) was computed upon the combining of the fat volume from the 10 – cm section of the *M. longisimus dorsi*, the fat: muscle ratio from the 15-cm section of the ham and the lean volume percentage from the 15-cm section of the ham. However, there is an obvious lower accuracy for the lean percentage as shown in Table 6**.**

Additionally, this variability of accuracy for the models should be addressed for the live weight range. Artifacts, due to the breathing of the animals, decreased the accuracy of estimation. Similarly, as shown above, biases occurred due to the involved volumes of well-defined regions of the back and ham in the estimation of the fat and protein content of the body at various weights of interest (Mitchel et al., 2001). This approach needs further development in order to improve the accuracy at various well-defined regions of the body in swine.

Conclusions

On the basis of the experimental results of *in vivo* prediction of the nitrogen retention rate or percent of muscular flash in the body of pigs and other farm animals it can conclude that the total body electrical conductance (TOBEC), the dual energy X-ray absorptiometry (DXA), magnetic resonance tomography (MRT), X-ray computer tomography (CT), and the radioactive isotope K^{40} measurements are the most accurate methods whit a coefficient of determination R^2 ranging from 0.85 to 0.98. Versa accuracy, simple data capture and relatively fast completion rates of those methods is expensive industrial instrumentation needed which holdback practical implementation in many cases. The MRT and TOBEC do not have any negative side effects with regard to the animal and the labor personnel, whereas to the CT and DXA has the small non-welcomed side effect of X-rays, which has to be taken into account.

The accurate prediction of nitrogen retention can be achieved also by bioelectric impedance analysis (BIA), with portable, simple and safe procedure, with tolerable expenses.

The D_2O-dilution technique is relatively accurate and shows additional advantages through low financial expenses and easy practical applications. However, results are influence by many factors, which need for further research in order to adjust the biases. This technique requires a long operational time and conditions, which is comparable with the N-balance technique.

The ultrasonic analysis have advantages as the simple handling, fast work completion, mobility, modest costs, and the lack of any negative side effects to the labor staff and the animals. However, the high accuracy of the predictions is not always obtained, depending on the metering devices, on the point of measurement and

on the metering person. Predictions are more accurate with two-dimensional device, but instruments are much more expensive than one-dimensional device.

Nitrogen balance method is easy to apply in experimental condition, but it need essential labor and time, and nitrogen retention is overestimated by 5 to 20%.

The all *in vivo* methods are continuing to be developed and improved in the instrument, manner of sampling and computer data processing.

Appendix 1
Calculation of the body composition of pigs by D_2O dilution technique

Water content of the body:
H_2O (kg) =D_2O in the animal (g)/ D_2O-content in the blood water (g/kg)-0.025*W (4)

Total content of the gastro-intestinal tract and the bladder (GIB):
GIB (kg) = (225-5.4N+812XF-60))*(1+0.0065(ITx-95))*W $^{0.75}$ /1000 (5)
N = fast period (days)
XF = crude fiber content of the dry matter, %
IT = feed intake converted to dry matter (g/Tag)/kg $W^{0.75}$

Total water in gastro- intestinal tract and the bladder (H_2O GIB)
H_2O GIB (kg) = 0.82 *GIB (kg) (6)

Water content of the empty body (H_2O e):
H_2Oe (%) = H_2O (kg) -
H_2O GIB (kg)*100/ W (kg) - GIB (kg) (7)

Fat free substance of the empty body (FFSe):
FFSe (%) = H_2Oe (%) / 82.43*W`-0.0219 (8)

Fat content of the empty body (XLe):
XLe (%) = 100 - FFSe (%)

Protein content of the empty body (Xpe):
XPe (%) = FFSe (%)*0.1402*W´0.0940 (9)

Ash content of the empty body (Xae):
XAe (%) = FFSe (%) *0.0307*W`0.0594 (10)

W = live weight (kg) in the time of D_2O – Application; We = empty body mass (W without the Total content of the gastro-intestinal tract and the bladder), W´ = We + 0.1 kg (Total content of the gastro-intestinal tract and the bladder) / kg $W^{0.75}$ (Susenbeth, 1984).

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Wikibooks: Basic physics of nuclear medicine/dual-energy absorptiometry

Received January, 23, 2012; accepted for printing June, 14, 2012.