



Effect of Partial Replacement of Maize with Maize cob - Molasses (*Mcmolas*) Meal on the Haematology and Biochemistry of Weaner Pigs

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Abstract

This experiment was conducted to determine the effect of partial replacement of maize with Maize cob-Molasses (MCMOLAS) meal in the diet of weaner pigs on their haematological and some biochemical indices on sixteen composite breed of pigs with an average weight of 8.12 kg \pm 0.5, the experiment lasted for a period of 84 days. MCMOLAS meal was prepared by mixing milled maize cobs and molasses in a ratio of 4:1 and pre-digested with an enzyme (cellulase) at 0.5 g/kg and ensiled in air tight container for 7days, sundried and stored for the feeding trial. Proximate composition of the MCMOLAS meal was determined before its inclusion in their diets. Four (4) pigs were allocated to each dietary treatment at 0, 15, 30 and 45% of MCMOLAS meal partially replacing maize in a completely randomized design (CRD). Results showed that the values obtained for haematological indices measured varied significantly ($P < 0.05$) among the dietary treatments except for lymphocyte (LY) and monocyte (MO), RBC, Hb, WBC, GR, MCHC all increased as the level of MCMOLAS meal increased in the diet, whereas, MCH, MCV, MPV, PCT, PLT and PDW decreased, HCT differed across the treatments. Most biochemical indices (TP, ALB, GLO, HDL, LDL and CHOL) were not statistically ($P < 0.05$) influenced by the MCMOLAS meal except glucose which decreased as the level of MCMOLAS meal increased. It was concluded that MCMOLAS meal can be incorporated at 15% into the diet of pigs without adverse effect on their health and well-being.

Keywords: Enzyme, Biochemistry, Haematology, pigs, MCMOLAS meal.

Introduction

Majority of pig farmers are confronted with the challenge of limited range of feed ingredients that can be utilized in formulation of pig diets, hence the need for research on the use of fibrous, easily accessible, cheap and unconventional or local feedstuffs in order to increase feed ingredients available to the farmers. According to Fetuga (1977), one of the major obstacles to livestock improvement in Nigeria is that of inadequate and unbalanced feed, this has immensely affected the production level of livestock because feed accounts for about 70 – 80% of the overall cost of animal production (Oyediji, 2001; Uchegbu *et al.*, 2009). The major energy sources in Nigeria are either maize or guinea corn, palm oil or groundnut oil is sometimes

used to boost energy content of pig feeds, (Perez, 1997), as a result of this limitation there is need to seek for other major energy sources of feed for pigs so as to make available enough animal protein source for human consumption as pork is one of the most veritable sources of animal protein to human (Apata and Ojo, 2000; Udofia *et al.*, 2007). There is also the need to reduce the competition between man and animal for grains, which led to efforts being made to use maize cobs as an animal feed, it has been discovered that an average of 180 to 200kg of maize cobs are available per ton of grains (Božović *et al.*, 2004) this may indicate that a large quantity of this feed resource is available. Maize cobs is fibrous in nature and pigs lack microbes in their digestive system

that can aid its digestion hence the need to pre-digest the maize cobs with the use of an exogenous enzyme and molasses.

Exogenous enzymes are now also commonly used in pig diets to disrupt anti-nutritional factors including fiber since pigs lack the digestive enzymes appropriate for fibre digestion. (Omogbenigun *et al.*, 2004; Jones *et al.*, 2010). Kenzyme[®] is an enzyme that has been used in chicken reared for broilers and layers, turkey and pigs. It contains five (5) active substances viz: endo-1, 3 (4) beta-glucanase with enzyme Activity (2350 u/g), endo-1, 4-beta-glucanase (cellulase) with E.A (18000u/g), alpha-amylase with E.A (400u/g), endo-1,4-beta-xylanase with E.A (35000u/g) and bacillolysin (protease) with E.A (1700u/g). Molasses value as a feed is based mostly on its sugar content (about 50%). In comparison with the carbohydrates in concentrated form, molasses contains a small amount of protein, but it provides also a certain amount of non-protein, non-sugars which have some nutrient value especially for ruminants (Senthilkumar *et al.* 2016). The objective of this study was to determine haematological and biochemical indices of pigs fed diets with *MCMOLAS* Meal as partial replacement for maize.

Materials and Methods

The study was conducted at the piggery unit of University of Benin Teaching and Research Farm, Ugbowo Campus Benin City, Edo State, Nigeria. The campus lies between Latitudes 6 and 30¹ N of the Equator and longitude 5^o40¹ and 6^oE of the Greenwich meridian in the rain forest zone, with mean monthly temperature of 27.6^oC. The area has an average annual rainfall and relative humidity of 2162 mm and 72.5% respectively (NAA, 2018).

Production and Proximate Composition of *MCMOLAS* Concentrate

The maize cob was collected from dry maize sellers at New Benin market in Benin-city, and broken into smaller sizes using a hammer and then milled at NIKSEG feedmill with industrial milling machine to granulated size. The milled maize cobs were treated with cellulase (Kenzyme[®]) at the rate of 0.5 g of kenzyme to 1kg of maize cobs, the enzyme treated maize cob was mixed with molasses sourced from a distributor in Ikire, Osun state. It was hand mixed in a ratio of 4:1 maize cob to molasses on a clean surface. It was then packed into a large bucket and compressed in order to remove all air before covering with polythene and finally the bucket cover. It was left to ferment for 7 days and on the 7th day, the *MCMOLAS* meal was opened, sun dried, stored in a sack and a sample taken to the laboratory for proximate analysis.

Feeding Trials and Animal Management

Four diets were formulated with *MCMOLAS* meal replacing 0%, 15%, 30% and 45% of maize in the growing diet on a weight for weight basis. Diet 1 represented the control while diets 2, 3 and 4 contained the varying levels of *MCMOLAS* at 15, 30 and 45%, respectively. The ingredients and determined composition of the experimental diets are presented on Table 2. Sixteen (16) five weeks old piglets (composite breeds) with the weight range of 8.12 – 8.62 kg purchased from a reputable pig farmer was used for the experiment. The pigs on arrival were allowed an adjustment period of two weeks during which they were fed uniform diet containing 3353 kcal/kg on DM basis and CP of 20.55%. They were later housed individually on concrete floor pens designed for pigs. Prophylactic dose of Ivomectin[®] and Oxytetracycline[®] as well as Multivitamin were also administered. After the adaptation period, the growing pigs were randomly assigned to four dietary treatments each replicated four (4) times, on weight equalization basis with each animal

constituting a replicate and fed differently *ad libitum*, with clean drinking water also provided. The pigs were closely monitored for any signs of ill - health. The experimental design adopted was Completely Randomized Design (CRD). The data collected was subjected to a one way analysis of variance using Genstat (2009) and the Separation of means was done using the Duncan Multiple Range Test (Duncan, 1955) at 5% level of probability.

Blood collection

At the end of the feeding trial, blood samples were collected from two (2) randomly selected pigs from each treatment. Five milliliters (5ml) of blood was withdrawn from the ear and thigh veins of the pigs through the use sterile hypodermic needle and syringe. Three milliliters (3ml) was collected in a labelled sample bottle containing speck of ethylenediamine tetra acetate (EDTA) to prevent the blood from coagulation and the remaining two milliliters (2ml) into a labelled plain bottle for blood serum analysis.

Results

The result of the proximate analysis of maize cobs, molasses and *MCMOLAS* are presented in Table 1. The values obtained from maize cobs were $3.98 \pm 0.03\%$, $28.80 \pm 0.10\%$, $3.34 \pm 0.06\%$, $3.27 \pm 0.06\%$ and $53.33 \pm 1.79\%$ for Crude Protein (CP), Crude Fibre (CF), Ether Extract (EE), Ash and Nitrogen Free Extract (NFE) respectively.

The values obtained for Molasses showed $2.47 \pm 0.02\%$, $0.00 \pm 0.00\%$, $0.25 \pm 0.02\%$, $14.53 \pm 0.06\%$ and $59.96 \pm 0.07\%$ for CP, CF, EE, Ash and NFE respectively while the values for the combination of ground maize cob and molasses which is *MCMOLAS* were $3.69 \pm 0.02\%$, $19.80 \pm 0.10\%$, $1.0 \pm 0.02\%$, $1.57 \pm 0.06\%$ and $63.84 \pm 0.30\%$ for CP, CF, EE, Ash and NFE respectively.

Haematological responses of growing pigs to the experimental diets are as presented in

Table 3. All haematological indices were significantly affected by dietary treatments ($P < 0.05$). The result showed that the Red Blood Cell (RBC), Haemoglobin (Hb), White Blood Cell (WBC) and Mean Cell Haemoglobin Concentration (MCHC) values significantly increased as the levels of *MCMOLAS* increased in the diets as compared to the control ($P < 0.05$). The highest value for RBC (9.28×10^8 UL), Hb (14.40g/dl) and MCHC (34.20g/dl) obtained for growing pigs fed 45% *MCMOLAS* meal ($P < 0.05$), while that of WBC (24.60×10^8 UL) was obtained for those fed 15%.

Conversely, Heamatocrit (HCT), Mean Cell Haemoglobin (MCH), Mean Cell Volume (MCV), Mean Platelet Volume (MPV), Platelet Concentration (PCT), and Platelet (PLT) values were significantly reduced as the level of *MCMOLAS* meal in the growing pigs diets increased with the lowest value of MCH (15.56pg), MCV (45.40fl) and MPV recorded 6.27fl at 45% *MCMOLAS* dietary inclusion level ($P < 0.05$). The Lymphocyte (LY) and Monocyte (MO) of the growing pigs were not significantly influenced ($P > 0.05$) across the treatments. However, the Relative Density of Whole blood (RDW) and Granulocytes (GR) values of growing pigs increased as the dietary level of *MCMOLAS* increased ($P < 0.05$). The highest RDW (21.20%) and GR (66.50%) were recorded for 15% *MCMOLAS* inclusion level in the diets of growing pigs. On the other hand, Platelet density of whole blood (PDW) was significantly reduced as the dietary level of *MCMOLAS* meal increased with the highest (16.30fl) and lowest (10.20fl) value recorded for the control (0% *MCMOLAS*) and 30% *MCMOLAS* inclusion in the diets respectively.

The Triglyceride (TG) level of growing pigs blood was significantly decreased from 63.0 mg/dl for the control to 74.0 mg/dl for growing pigs fed 45% *MCMOLAS* meal ($P > 0.05$) as shown in Table 4. Conversely, the

blood glucose (GLU) reduced significantly ($P < 0.05$) from 175.7 mg/dl in growing pigs fed diets without *MCMOLAS* (Maize based diet) to 50mg/dl in growing pigs fed 45% *MCMOLAS* meal. However, blood total protein (TP), globulin (GLO), albumin (ALB), high density lipoprotein (HDL), low density lipoprotein (LDL) and blood cholesterol (CHOL) were not significantly ($P > 0.05$) affected by the inclusion level of *MCMOLAS* meal.

Discussion

The proximate composition of maize cobs obtained during this study showed that crude protein (CP) value was $3.98 \pm 0.03\%$, which was lower than the value reported by Wang (2009)., he reported that a good quality maize cobs should contain a CP of above 4%. This value is also lower compared to 4.19% reported by Abubakar *et al.* (2016). The slightly low value of CP could be attributed to differences in the state of maturity of the maize cobs, the more matured the maize cobs the less the value of CP likely to be obtained (Olanite *et al.*, 2010). Also, different species of maize cobs analysed, analytical procedures and accuracy could also affect results obtained. The Crude Fibre (CF), content of maize cobs used in this study was $28.80 \pm 0.10\%$ which was below the range of 31.65% - 40.06% quoted by Olanite *et al.* (2010) and 33.33% suggested by Abubakar *et al.* (2016).

This could be as a result of different cultivar of maize cobs used in this study compared to other studies. Ether extract (EE) also had $3.34 \pm 0.06\%$ which is lower to 4.72% quoted by Abubakar *et al.* (2016) also the Ash content of $3.27 \pm 0.06\%$ was higher and the nitrogen free extract (NFE) of $53.33 \pm 1.79\%$ was also higher than 48.56 quoted by Abubakar *et al.* (2016). Notably, the high NFE value of 53.33% suggests that the cultivar of maize cob used in this study is higher in energy value which is most essential for this study. According to Thomas *et al.* (2001), Johnson *et*

al. (2002) and Jensen *et al.* (2005), maize cob differs not only in composition as a result of cultivar but also due to planting season, type of farm (modern or traditional farm), maturity at harvest, location of maize plantation and mechanical processes used for grinding the maize cobs.

However, the proximate composition of molasses was $2.46 \pm 0.02\%$ for the CP, this CP of molasses is fairly on the same range of 3-4% as reported by Olomu, (2011), but lower than 4% reported by Cleasby, (1963) the CF was $0.00 \pm 0.00\%$, EE was $0.25 \pm 0.02\%$ and Ash was $14.53 \pm 0.06\%$ which was similar with the value reported by Getahun *et al.* (2018). The NFE was $59.96 \pm 0.07\%$ which was slightly lower compared with 62.00% reported by Cleasby, (1963).

The energy concentrate, Maize cobs and Molasses (*MCMOLAS*) had a CP of $3.69 \pm 0.02\%$, CF of $19.80 \pm 0.10\%$, EE of $1.0 \pm 0.02\%$, Ash of $1.57 \pm 0.06\%$ with NFE of $63.84 \pm 0.30\%$, it was observed that the CF which was 28.80% in maize cobs has been reduced to 19.80% in *MCMOLAS*. This could be attributed to the action of the enzyme (Kenzyme[®]) used as well as the fermentation of the meal for a period of 7 days. However, the NFE value obtained for *MCMOLAS* (63.84) is of good quality for energy (Maize) replacement when compared to the value obtained for maize cobs.

The white blood cell (WBC) of the growing pigs was high at 15% and 30% *MCMOLAS* (24.60×10^8 UL and 24.47×10^8 UL) which is slightly above the value quoted by RAR (2009), Etim *et al.* (2013), ($7-20 \times 10^8$ /UL), and Coronado (2014)., ($11-22 \times 10^8$ /UL). The main functions of the white blood cell is to fight infections and protect the body by phagocytosis against invasion by foreign organisms and help in production, transportation and distribution of antibodies in immune respons (Kabir *et al.*, (2011),

Okunlola *et al.*, (2012), Soetan *et al.*, 2013). Iwuji and Herbert. (2012) and Isaac *et al.*, (2013), stated that animals with low white blood cells are exposed to high risk of disease infection, while those with high counts are capable of generating antibodies in the process of phagocytosis and have high degree of resistance to diseases and reduced susceptibility to local environmental and disease prevalent conditions. The high value recorded for WBC in this study may be due to the fact that tropical pigs are usually exposed to harsh environmental conditions and challenges of disease invasion, thus, the slight increase may be the pig's response to the challenges (Eze *et al.* 2010).

The hematocrit (HCT) value were within the range quoted by RAR (2009), Etim *et al.*(2013) and Coronado (2014) which indicated that the transportation of blood and absorption of nutrients is normal. The haemoglobin concentration (Hb) value was in line with the values reported by RAR (2009); Etim *et al.*, (2013) and Coronado (2014) which indicated that the pigs were not affected with anaemia as a result of the test ingredient. The mean corpuscular volume (MCV) at 45% *MCMOLAS* was lower than the values of 50-68 quoted by Coronado (2014) which may indicate a microcytic anaemia. mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) are within the ranges quoted by RAR (2009); Etim *et al.*, (2013) and Coronado (2014), though their values reduced with the increase in the level of *MCMOLAS* in the diets of the pigs. According to Peters *et al.* (2011), Packed cell volume (PCV), Hb and MCH are the main guide for evaluation of circulatory erythrocytes, and are principal in the diagnosis of anaemia which can also be used as an indication of the bone marrow capacity to produce red blood cells in mammals (Awodi *et al.*, 2005; Chineke *et al.*, 2006). High packed cell volume (PCV) value means either an increase in number of Red

Blood Cells (RBC) or reduction in circulating plasma volume (Chineke *et al.* 2006). Mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration stipulates the blood level conditions and a low level of these indicates anaemia (Aster, 2004).

Reduction in glucose concentrations (GLU) with increase in inclusion level of fibre reflects the negative energy balance in the pigs. The inclusion of fibre in a diet is likely to have resulted in some nutrients not digested and absorbed, mainly due to accelerated transit time of feed along the gut (Wenk, 2001; Metzler and Mosenthin, 2008). Furthermore, in a study conducted by Antuszewicz, *et al.* (2005) with 13 male pigs weighing 39kg which were fed with five (5) different fibre sources diets at different time intervals, it was deduced from the study that the pigs fed diets containing molasses had a normal concentration of glucose in its blood. Glucose levels generally stipulate the efficiency of utilization of metabolizable energy (ME) in a diet (Fanim, 1991). The capacity of pigs to extricate energy yielding nutrient from fibrous material is important for improving the productivity of pig. The reduction in glucose level as the level of maize cobs increased, presumably indicated that pigs on higher level (30% and 45%) of maize cobs were not able to extract enough energy from the diets.

Conclusion

From the results of this study it can be concluded that *MCMOLAS* meal can be used to replace maize in the diet of pigs up to 15% without adverse effect on the hematological indices of the pigs.



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Table 1: Proximate composition of Maize cobs, Molasses and “MCMOLAS”

PARAMETERS	Maize cobs	MCMOLAS	Molasses
Moisture content (%)	8.39±0.01	10.10±0.30	22.80±0.10
Crude protein (%)	3.98±0.03	3.69±0.01	2.46±0.02
Crude fibre (%)	28.80±0.10	19.80±0.10	0.00±0.00
Ether extract (%)	3.34±0.06	1.00±0.10	0.25±0.02
Ash (%)	3.27±0.06	1.57±0.06	14.53±0.06
Nitrogen free extract (%)	53.33±1.79	63.84±0.30	59.96±0.07
Gross energy value(Kcal/kg)	2.62±2.25	2.83±2.12	2.56±2.32
Phosphorus (mg/kg)	1121.4±8.10	776.7±13.2	280.2±12.5
Potassium (mg/kg)	7.85±0.06	2.40±0.06	1.79±0.01
Calcium (mg/kg)	31.05±0.06	8.41±0.02	16.79±0.03
Magnesium (mg/kg)	218.50±0.20	97.51±0.09	272.30±0.20
Sodium (mg/kg)	54.340±0.04	31.07±0.59	50.97±0.76
Iron (mg/kg)	198.41±0.51	217.77±0.68	223.00±2.00



Table 2:

Composition of Experimental Diet (%)

Ingredient	Diet 1 (control)	Diet2 (15% <i>MCMOLAS</i>)	Diet3 (30% <i>MCMOLAS</i>)	Diet4 (45% <i>MCMOLAS</i>)
Maize	60.00	45.00	30.00	15.00
<i>MCMOLAS</i>	0.00	15.00	30.00	45.00
Groundnut meal	19.00	19.00	17.00	18.00
Soyabean meal	10.00	13.06	13.06	13.06
Wheat bran	7.35	4.29	6.29	5.29
Common salt	0.50	0.50	0.50	0.50
*Premix	0.20	0.20	0.20	0.20
Periwinkle	0.75	0.75	0.75	0.75
Bonemeal	2.00	2.00	2.00	2.00
Methionine	0.05	0.05	0.05	0.05
Lysine	0.15	0.15	0.15	0.15
Total	100	100	100	100
Determined composition				
Dry Matter (%)	88.34	85.10	85.48	85.63
Crude Protein(calculated)	20.55	20.79	19.36	18.93
Ether Extract (%)	18.30	14.09	9.90	5.78
Crude Fibre (%)	4.66	8.23	10.82	13.50
Ash (%)	5.19	7.91	4.36	7.46
NFE (%)	45.85	44.88	52.82	52.97
Energy(calculated) kcal/g	3353.00	3277.00	3148.00	3045.00
Calcium(mg/kg)	1.92	0.94	1.07	1.58
Phosphorous (mg/kg)	0.97	0.78	0.60	0.68

*Premix supplied the following per kg diet: vitamin A, 12,000 i.u; vitamin D₃, 1200 i.u.; vitamin E, 15.i.u; vitamin K, 1.8mg; thiamine, 0.8mg; riboflavin, 3.0mg; niacin, 18mg; calcium panthothenate, 10.5mg; biotin, 0.0375mg; vitamin B₆, 1.5mg; choline chloride, 75mg; folic acid, 0.3mg; vitamin B₁₂, 0.012 mg; vitamin C, 15mg; manganese, 15mg; zinc, 87.6mg; copper, 15mg; iron, 75mg; iodine, 0.5mg; cobalt, 0.54mg; selenium, 0.06mg.

Table 3: Haematological Response of Growing pigs fed diets containing levels of “MCMOLAS” as a partial replacement for maize.

Parameters	DIETS				SEM
	T1 (0% <i>MCMOLAS</i>)	T2 (15% <i>MCMOLAS</i>)	T3 (30% <i>MCMOLAS</i>)	T4 (45% <i>MCMOLAS</i>)	
RBC($10^8/l$)	7.74 ^c	8.89 ^{ab}	8.08 ^{bc}	9.28 ^a	0.24
HCT (%)	44.27 ^{ab}	44.80 ^{bc}	45.90 ^a	42.07 ^c	0.55
Hb (g/dl)	13.37 ^b	14.30 ^a	14.27 ^a	14.40 ^a	0.25
WBC($10^8/UL$)	16.13 ^b	24.60 ^a	24.47 ^a	17.93 ^b	0.60
MCH(pg)	17.37 ^a	16.07 ^b	17.67 ^a	15.50 ^b	0.24
MCHC (g/dl)	30.17 ^c	32.67 ^b	31.03 ^c	34.20 ^a	0.42
MCV(fl)	57.30 ^a	49.27 ^b	56.93 ^a	45.40 ^b	1.42
MPV (fl)	8.63 ^a	7.00 ^b	6.87 ^b	6.27 ^b	0.23
PCT (%)	0.55 ^a	0.25 ^b	0.23 ^b	0.22 ^b	0.02
PLT ($10^8/UL$)	642.00 ^a	353.70 ^b	326.70 ^b	336.00 ^b	30.60
RDW %	18.60 ^b	21.20 ^a	18.60 ^b	20.70 ^a	0.40
LY($10^8/UL$)	52.70 ^a	49.70 ^a	65.70 ^a	59.00 ^a	0.53
MO($10^8/UL$)	2.67 ^a	3.40 ^a	3.90 ^a	3.43 ^a	0.36
GR (%)	49.83 ^b	66.50 ^a	58.10 ^{ab}	58.57 ^{ab}	2.88
PDW (fl)	16.30 ^a	11.10 ^b	10.20 ^b	11.47 ^b	1.05

^{abc} means with different superscripts in the same row differ significantly ($p < 0.05$).

PCV/HCT= Packed cell volume (Haematocrit) RBC= Red blood cell MCHC= Mean cell haemoglobin concentration
 PLT= Platelet
 MPV= Mean platelets volume WBC = White blood cell MCV= Mean cell volume Hb = Heamoglobin
 MCH= Mean cell haemoglobin PCT= Platelet concentration MO = Monocyte GR = Granulocyte PDW= Platelet
 density in whole blood LY = Lymphocyte
 RDW= Relative density of whole blood

Table 4: Effect of *MCMOLAS meal* on some biochemical parameters of growing pigs

Parameters	DIETS				SEM
	T1 (0% <i>MCMOLAS</i>)	T2 (15% <i>MCMOLAS</i>)	T3 (30% <i>MCMOLAS</i>)	T4 (45% <i>MCMOLAS</i>)	
TG (mg/dl)	63.0 ^b	61.7 ^b	70.3 ^a	74.0 ^a	1.71
TP (g/dl)	6.30 ^a	7.20 ^a	6.67 ^a	6.87 ^a	0.29
GLO (g/dl)	3.20 ^a	3.53 ^a	3.10 ^a	3.37 ^a	0.27
GLU(mg/dl)	175.7 ^a	140.0 ^{ab}	80.0 ^{bc}	50.7 ^c	25.6
HDL(mg/dl)	43.7 ^a	54.0 ^a	49.7 ^a	43.0 ^a	4.68
LDL(mg/dl)	82.0 ^a	70.7 ^a	73.7 ^a	87.7 ^a	5.12
CHOL(mg/dl)	137.0 ^a	136.7 ^a	137.3 ^a	145.7 ^a	12.62
ALB (g/dl)	3.10 ^a	3.67 ^a	3.57 ^a	3.50 ^a	0.16

^{abc} means with different superscripts in the same row differ significantly (p<0.05).

TG = Triglyceride TP = Total protein GLO = Globulin GLU = Glucose CHOL = Cholesterol LDL = Lower density lipoprotein HDL = Higher density lipoprotein

ALB=Albumin