International Journal of Medical and Health Research
ISSN: 2454-9142
Received: 04-10-2018; Accepted: 06-11-2018
www.medicalsciencejournal.com
Volume 4; Issue 12; December 2018; Page No. 13-17

Estimation of reference intervals for liver function tests in apparently healthy population of Lucknow

Nikhil Gupta[1], Ritu Karoli[2*], Jalees Fatima[3], Prem Shanker Singh[4], Amit P Srivastava[5], GG Agarwal[6]

[1] Assistant Professor, Department of Medicine, Dr. R.M.L.I.M.S., Lucknow, Uttar Pradesh, India
[2] Associate Professor, Department of Medicine, Dr. R.M.L.I.M.S., Lucknow, Uttar Pradesh, India
[3] Professor & Head, Department of Medicine, E.L.M.C. & H., Lucknow, Uttar Pradesh, India
[4] Professor &Head, Department of Medicine, Dr. R.M.L.I.M.S., Lucknow, Uttar Pradesh, India
[5] Assistant Professor, Department of Medicine, E.L.M.C. & H., Lucknow, Uttar Pradesh, India
[6] Professor, Department of Statistics, Lucknow University, Uttar Pradesh, India

Abstract
Introduction: Liver function tests do vary with various factors like race/ethnicity, environmental, life styles, metabolic, physiological changes with advancing age and gender. Clinicians need to know these variations in reference interval when considering the presence of liver disease. There is scarce data of reference interval of liver function tests in north Indian population which have drastically different food habits, life style, exposure to environmental conditions as compared to the population of Southern and Eastern India.
Material and Methods: Apparently healthy medical and paramedical professionals and healthy volunteer donors coming to the blood bank were included in the study and studied for eight liver function parameters.
Results: 1302 subjects were screened for the study.265 subjects were excluded as 73 were diabetics (fasting $\geq$ 126mg/dl), 190 subjects had fatty liver detected on ultrasound and 2 were Hepatitis B positive. 1037 subjects were included. The upper and lower limit of Reference Interval (RI)(observed Vs reported) for Direct Bilirubin (0.3-0.9Vs 0.3-1.3, SGPT(19-53.5 Vs 7-41), SGOT(16-47.0 Vs 12-38), GGT(35-90 Vs 9-58), ALP(47.5-134.5 Vs 33-96)were different as compared to reported standard RI. The upper limit of GGT (64-92 vs 34- 68), SGPT (18-54 vs20- 52) and SGOT (16-48 vs17- 45.8) was greater in males than females. The upper limit of SGPT was higher in urban population than rural and ALP showed more wide variation in RI in rural than urban population.
Conclusion: The reference interval for most liver function tests determined in this study were different from the reported reference interval in literature. Larger community based studies are needed for determining reference intervals of our population which will provide valuable guidance for clinical practice and decision making.

Keywords: reference intervals, liver function tests, apparently healthy population of Lucknow

Introduction

Reference interval (RI) for biochemical and haematological parameters are used to aid physicians to interpret the results of clinical measurements. In establishing reference values, it is essential that population is well defined and properly selected to be representative of that population. The lower and upper limits of measurement are known to be more affected by the choice of sample population, standardization of sample collection, handling, analysis and statistical analysis [1, 2].

Liver function tests comprise a variety of individual tests that are used to evaluate how well the liver functions. Diseases like non-alcoholic fatty liver disease, chronic liver disease can just present with mild elevation of aminotransferases. Level of aminotransferases has diagnostic as well as treatment implications in liver diseases. So it becomes important to know about normal reference range in our set of population. Reference interval do vary with various factors like race/ethnicity, environmental, life styles, metabolic, physiological changes with advancing age and gender based formal changes [3] Clinicians need to know these variations in reference interval when considering the presence of liver disease (especially with mild derangement in transaminases). The expert committees in USA formulated some set of rules which stated that each manufacturer of diagnostic kit must provide reference intervals and the laboratories are instructed to validate the same and prove that they are applicable for their environment and also for their population [4].

There is scarce data of RI of liver function tests in north Indian population which have drastically different food habits, life style, exposure to environmental conditions as compared to the population of southern and eastern India [12] The aim of this study was to estimate reference ranges for liver function tests in the apparently healthy group of individuals of Lucknow and surrounding regions that would serve as the reference values for the reference population.

Materials and Methods

Study Area and Population

The present study was conducted in department of medicine, Era's Lucknow Medical College, Lucknow. It is a tertiary care

medical college which caters to the population of Lucknow and its surrounding districts. Study subjects were from rural and urban areas of Lucknow and its surrounding districts. It was a cross sectional study and data was collected for 1 year between June 2015 to June 2016.

Ethical Consideration

Ethical approval was obtained from ethical committee of Era' Lucknow Medical College. Written and informed consent was taken for participating in the study from the subjects.

Inclusion and Exclusion Criteria

Study subjects were apparently healthy medical and paramedical professionals and healthy volunteer donors coming to the blood bank of Era's Lucknow medical College between 18 to 60 years. People on any form of medication or oral contraceptives, history of alcohol intake >20 gm/day, body mass index>30Kg/m [2], pregnancy, on excessive exercise, known hypertension, diabetes mellitus, coronary artery disease, chronic liver disease, chronic renal failure, chronic respiratory insufficiency, HIV, Hepatitis B, Hepatitis C positive were excluded from the study. Fasting serum samples of 1302 subjects were taken out of which 265 subjects were excluded and 1037 patients were finally included in the study. Information regarding age, sex, address, disease if any, dietary habits, physical activity, smoking or alcoholic habit, anthropometric parameters were obtained in respect to each subject.

Sampling and Laboratory Analysis

Fasting venous samples were taken after 8 hrs of fasting. Analysis of samples was done after proper standardization of the instruments with the help of calibrators and controls. Samples which were lipemic, hemolytic or icteric were not considered in the study. Samples were analysed for eight liver function parameters, fasting blood sugar and triglycerides level.Total bilirubin and direct bilirubin were analyzed by Diazo (Walter and Gerarde) method [5] Serum glutamate oxaloacetate transferase (SGOT) and Serum glutamate pyruvate transferase (SGPT) were done by IFCC without Pyridoxal Phosphate (L.aspartate/L. Alanine/oxoglutarate) method [6, 7]. Gamma glutamyltransferase (GGT) was analysed by IFCC recommended GCNA to glycylglycine conversion method.[8] Alkaline phosphatise (ALP) was estimated by ALP-AMP method], Totalprotein and albumin by Biuret and Bromocresolgreen method [9-11] All tests were performed on Transasia EM-360 fully automatic biochemistry analyzer. To ensure the reproducibility and repeatability of the testresults, the laboratory participated in established external quality assessment programs by CMC, Vellore and Erba Mannheim, Germany and a comprehensive internal quality control program. The quality control check was done every day.

Statistical analysis

Data were double entered into a Microsoft Excel database, compared, and corrected for data entry errors then imported into Statistical Package for Social Sciences (SPSS).Outliers were identified in the data by calculating the first quartile (*Q25*), the median (*Q*.50) and third quartile (*Q*.75). The interquartile range (IQR) was calculated by subtracting the first quartile from the third quartile (*Q*.75 - *Q*.25). Any data observation which lay more than 1.5 × IQR lower than the first quartile or 1.5 × IQR higher than the third quartile was considered an outlier and deleted from data. The above exclusions for some parameters led to different sample sizes for each parameter. The lower limit of the reference interval was 2.5th percentile of the distribution (variable) and upper limit of the reference interval was 97.5th percentile of the distribution.

Results

Study subjects were apparently healthy individuals coming to blood bank for blood donation. In the present study, 1302 subjects were screened and265 subjects were excluded as 73 were diabetics (fasting ≥ 126mg/dl), 190 subjects had fatty liver detected on ultrasound and 2 were HBsAg positive.

Out of 1037 subjects who were included in the study, 783 were males and 254 were females ranging from 18-60 years. Median age of males and females was 30 and 32 years respectively. Reference ranges for eight liver function biochemical parameters were estimated. The reference values were constructed using 2.5th and 97.5th percentiles as lower and upper limits. The upper and lower limit of Reference Interval (RI)(observed Vs reported) for Direct Bilirubin (0.3-0.9Vs 0.3-1.3, SGPT(19-53.5 Vs 7-41), SGOT(16-47.0 Vs 12-38), GGT(35-90 Vs 9-58), ALP(47.5-134.5 Vs 33-96) were different as compared to reported standard RI while the RI(observed vs reported) for total bilirubin, total protein and albumin were nearly equal (Table 1).

Table 2 depicts liver function test parameters according to different partitioning criteria. The upper limit of GGT (64-92 vs 34- 68), SGPT (18-54 vs20- 52) and SGOT (16-48 vs17-45.8) was greater in males than females while there was no difference between males and females in rest of the parameters. The upper limit for SGPT (18.0-57.0U/L) was highest in 18-28 years group and decreased in >39 years group onwards. The upper limit of SGO Twas nearly same in all age groups with a little decrease in 49-60 years group(14.5-45 U/L).The upper and lower limit of alkaline phosphatise (57.4-141.0U/L) was more in the oldest age group (49-60 yrs). The upper limit of SGPT was higher in urban population than rural and ALP showed more wide variation in RI in rural than urban population. In overweight and obese group upper value of SGPT was higher. RI for ALP, SGPT, and SGOT was higher innon-vegetarian group than vegetarian group. Smokers had higher RI for GGT, ALP and upper value of SGPT. The upper limit of SGPT and SGOT was higher in impaired fasting glucose group and higher triglycerides group.

Table 1: Comparison of observed vs reported RI

Variables	Extreme Lower Cases (No.)	Extreme Upper Cases (No.)	No. of cases	Observed RI	Reported RI	Reported RI (Harrison)
SBT(mg/dl)	0.2(5)	1.0(30)	1002	0.3-0.9	0.0-1.0	0.3-1.3
SBD(mg/dl)	0.05(0)	0.50(65)	972	0.1-0.4	0.0-0.2	0.1-0.4
GGT(U/L)	33.0(15)	117.5(0)	1022	T- 35.0-90.0 M-64.0-92.0 F 34.0-68.0	M- 15.0-85.0 F- 5.0-55.0	T- 9.0-58.0
ALP(U/L)	28(0)	145(19)	1018	T-47.5-134.5, M-47.2-135.5, F- 45.6-134.0	M-53.0-128.0 F- 42.0- 98.0	T-33.0-96.0
SPR(g/dl)	6.1(0)	8.5(0)	1037	7.0-7.8	6.4-8.3	6.7-8.6
SAL(g/dl)	2.7(0)	5.1(0)	1037	3.4-4.6	3.5-5.2	3.5-5.5
SGPT(U/L)	5(0)	62(16)	1021	T-19.0-53.5, M-18.0-54.0, F-20.0-52.0	M- 0.0-45.0 F - 0.0-34.0	T-7.0-41.0
SGOT(U/L)	11(0)	52(22)	1015	T-16.0-47.0, M-16.0-48.0, F- 17.0-45.8	M 0.0-35.0 F 0.0-31.0	T-12.0-38.0

Table 2: Liver function test parameters according to different partitioning criteria

		Total Bilirubin	Direct Bilirubin	GGT	ALP	SGPT	SGOT	Protein	Albumin
Total N=1037	N=RI	1002	972	1022	1018	1021	1015	1037	1037
	Lower	0.3	0.1	35.0	47.5	19.0	16.0	7.0	3.4
	Upper	0.9	0.4	90.0	134.5	53.5	47.0	7.8	4.6
Male N=783	N=RI	756	757	783	766	770	766	783	783
	Lower	0.3	0.1	64.0	47.2	18.0	16.0	7.0	3.4
	Upper	0.9	0.4	92.0	135.0	54.0	48.0	7.8	4.6
Female N=254	N= RI	246	215	239	252	251	249	254	254
	Lower	0.3	0.1	34.0	45.6	20.0	17.0	7.0	3.4
	Upper	0.9	0.4	68.0	134.0	52.0	45.8	7.8	4.6
Age (18-28) YearsN=448	N= RI	432	440	445	435	441	438	448	448
	Lower	0.3	0.1	36.0	42.8	18.0	15.0	7.0	3.45
	Upper	0.9	0.4	90.0	134.0	57.0	48.0	7.8	4.6
Age (29-38) YearsN=354	N=RI	339	338	349	349	349	347	354	354
	Lower	0.3	0.1	34.0	50.0	19.0	16.0	7.0	3.4
	Upper	0.9	0.4	92.0	137.0	52.3	47.3	7.8	4.6
Age (39-48) Years N=180	N=RI	177	154	174	179	177	176	180	180
	Lower	0.3	0.1	35.0	47.0	19.5	18.0	7.0	3.6
	Upper	0.9	0.4	90.0	134.0	49.0	47.6	7.8	4.6
Age (49-60) YearsN=55	N=RI	54	40	54	55	54	54	55	55
	Lower	0.34	0.1	34.0	57.4	15.4	14.5	7.0	3.48
	Upper	0.9	0.4	92.0	141.0	49.8	45.0	7.8	4.6
Rural N=312	N=RI	299	304	308	302	305	304	312	312
	Lower	0.3	0.1	42.2	37.7	18.0	15.6	7.0	3.4
	Upper	0.9	0.4	90.0	138.9	52.4	48.4	7.8	4.6
Urban N=725	N=RI	703	668	714	716	716	711	725	725
	Lower	0.3	0.1	34.0	49.0	19.0	16.0	7.0	3.4
	Upper	0.9	0.4	90.0	134.0	55.0	47.0	7.8	4.6
BMI<23 Kg/m^2 N=371	N=RI	358	348	367	366	370	366	371	371
	Lower	0.3	0.1	34.0	46.2	18.0	15.2	7.0	3.4
	Upper	Upper	Upper	Upper	Upper	Upper	Upper	Upper	Upper
BMI 23-<25 N=350	N=RI	343	338	344	340	346	345	350	350
	Lower	0.3	0.1	35.6	49.5	19.0	15.7	7.0	3.6
	Upper	0.9	0.4	90.8	134.0	52.3	47.0	7.8	4.6
BMI≥25 N=316	N=RI	301	286	311	312	305	304	316	316
	Lower	0.3	0.1	35.0	44.7	18.7	17.6	7.0	3.4
	Upper	0.9	0.4	92.0	136.0	56.0	47.4	7.8	4.6
Veg N=762	N=RI	735	721	750	750	761	762	762	762
	Lower	0.3	0.1	35.0	45.8	18.0	16.0	7.0	3.4
	Upper	0.9	0.4	90.0	134.0	43.0	40.0	7.8	4.6
Non-veg N=275	N=RI	267	251	272	268	260	253	275	275
	Lower	0.3	0.1	36.7	49.7	20.0	17.4	7.0	3.4
	Upper	0.9	0.4	92.0	137.0	58.5	50.0	7.8	4.6
Smoker N=164	N=RI	160	148	164	147	164	160	164	164
	Lower	0.3	0.1	54.3	52.1	17.1	17	7.0	.4
	Upper	0.9	0.4	92.0	143.0	56.5	48	7.8	4.6
Non-smoker N=873	N=RI	842	824	858	871	857	855	873	873
	Lower	0.3	0.1	34.5	46.0	19.0	16.0	7.0	3.6
	Upper	0.9	0.4	88.0	134.0	53.1	47.0	7.8	4.6

FBS≤100 mg/dl N=856	N=RI	829	803	842	839	842	837	856	856
	Lower	0.3	0.1	35.1	46.0	18.0	16.0	7.0	3.4
	Upper	0.9	0.4	90.0	135.0	52.0	47.1	7.8	4.6
FBS (101-125) mg/dl N=181	N=RI	173	169	180	179	179	178	181	181
	Lower	0.3	0.1	34.0	48.5	19.0	15.0	7.0	3.4
	Upper	0.9	0.4	92.0	134.0	57.0	48.1	7.8	4.6
Triglycerides ≤150 N=700	N=RI	680	646	692	685	692	691	700	700
	Lower	0.3	0.1	35.0	46.0	18.3	16.0	7.0	3.4
	Upper	0.9	0.4	90.0	135.0	51.0	47.0	7.8	4.6
Triglycerides>150 N=337	N=RI	322	326	330	333	329	324	337	337
	Lower	0.3	0.1	35.3	49.0	19.0	16.0	7.0	3.4
	Upper	0.89	0.4	91.5	134.0	56.8	48.0	7.8	4.6

Discussion

In clinical practice, laboratory reference ranges are an important tool for identifying abnormal laboratory results and for guiding patient management decisions. Liver function tests are one of the commonest laboratory parameters used in day to day patient care and also used to detect or to monitor treatment responses in patients with liver diseases.

The present study included apparently healthy individuals to assess the liver function test and to compare it with the reference range provided by the laboratory and the international standards. The reference interval (RI) for direct bilirubin, SGPT, SGOT, GGT and ALP were different as compared to reported standard RI probably because of difference in lifestyle, dietary habits, race. age and body mass index. Impact of these factors have never been related to liver specific parameters as a result of which normal reported RI remain unchanged for the past several years despite the population exposure to these factors. International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) recommends that the establishment of reference intervals requires a minimum of 120 individuals in each subgroup which was achieved in every subgroup in this study. The number of males was much greater than females possibly due to less frequent participation of females in blood donation. The upper limit of GGT (64-92 vs 34- 68), SGPT(18-54 vs20-52) and SGOT(16-48 vs17- 45.8) was greater in males than females which was consistent with other studies.[12-14] Sex differences in SGOT,SGPT have been known to exist due to differences in muscle mass. The differences by sex noted for GGT could be due to extra production from the prostate gland in males as compared to females who have no prostate gland.[15]The upper limit of SGPT increases with age up to 40 years and thereafter decreases [16]. This may be due to loss of liver cell integrity as age advances. In our study the upper limit of SGPT decreased in age greater than 38 years group but was increased in 18-28 years group which is in contrast to previous studies [12]. This may be because of increase in activity or more exercise in the youngest group. SGOT upper reference limits increase from childhood to young adult, but changes relatively little with increasing age in adults until after age 60 [16] In our study SGOT RI was nearly same in all age groups. Values of alkaline phosphatase are slightly higher in men than in women until late in life. Upper reference limits are slightly increased in older age group especially in females [16]. RI of alkaline phosphatase was higher in oldest age group in our study which was in agreement with previous studies who reported an increase of serum ALP with advancement in age [12].

In rural and urban partitioning the upper limit of SGPT was higher in urban population than rural and ALP showed more wide variation in RI in rural than urban population which was in concordance with previous study done in Jaipur [17] This difference could be attributed to different lifestyle habits. Reference interval of all the liver function parameters seems to be affected considerably by obesity but in our study only SGPT upper limit was higher in BMI > 23.0 kg/m^2 group. As per dietary habits SGOT, SGPT, ALP had higher RI in Non-Vegetarian group which is similar to previous study done by Purkins *et al*. who reported rise in SGPT by day 9 and day 12 to an average of 143% (95% CI 79, 231; $P < 0.001$) on high carbohydrate high calorie diet [18].

Smokers showed higher RI for upper value of SGPT, GGT and ALP. Previous studies showed controversial results whether smoking could affect aminotransferase activities (AST and ALT). Some investigators [19, 20]. Claimed AST increased by cigarette smoking as our results support, while other studies [21] argued that smoking did not influence AST or ALT Cigarrete smoke contains considerable number of free radicals which are highly reactive atom, which damage the biological membrane through lipid peroxidation. Lipid peroxidation, damage the biological cell membrane of the liver and serum aminotransferases are increased. Increased ALT level has been associated with reduced insulin sensitivity, adiponectin and glucose tolerance as well as increased free fatty acids and triglycerides which is in coherence with our findings [22]. The significance of this study is that there no recent data of reference interval of liver specific parameters in north Indian population. In view of increasing changes in the lifestyle of population this type of study is required as clinicians need to be aware of variation in RI according to different subgroups which will help in diagnosis and treatment.

The major limitation of our study was being done at a single Centre and was a hospital based study, so it may not be a representation of actual reference population. Larger sample size is required to validate the findings.

Conclusion

The reference interval for most liver function tests determined in this study were different from the reported reference interval. In the view of above variations seen in RI of various liver specific biochemical parameters, RI for different reference population should be established. Further larger studies are required to know the difference in reference intervals of different subgroups according to age, gender body mass index. This will provide us valuable guidance to improve

diagnosis and help in clinical practice and decision making to improve patient care.

References

1. Sasse Edward A, Doumas Basil T, Orazio Paul, Eckfeldt John H, Evans Susan A, Graham Gary A, *et al*. How to define and determine reference intervals in the clinical laboratory approved guideline second edition. NCCLS Document C28-A2. 2000; 20:13.
2. Solberg Helge Erik. The IFCC recommendation on estimation of reference intervals. The Ref Val Program. Clin Chem Lab Med. 2004; 42(7):710-4.
3. Horn Paul S, Pesce Amadeo J. Effect of ethnicity on reference intervals. Clin Chem. 2002; 48(10):1802-4.
4. TMalati. Whether western normative laboratory values used for clinical diagnosis are applicable to Indian population? an overview on reference interval.Indian Journal of Clinical Biochemistry. 2009; 24:111-22.
5. Doumas BT, Perry BW, Sasse EA, Straumfjord JV. Standardisation in bilirubin assays: Evaluation of selected methods and stability of bilirubin solutions. Clin Chem. 1973; 19:984-93.
6. Saris NE. Revised IFCC method for aspartate aminotransferase. Clin Chem. 1978; 24:720-1.
7. Bergmeyer HU, Scheibe P, Wahlefeld AW. Optimization of methods for aspartate aminotransferase and alanine aminotransferase. Clin Chem. 1978; 24(1):58-73.
8. Shaw LM, Stromme JH, London JL, Theodorsen L. IFCCmethods for the determination of enzymes part 4. IFCC method for gamma glutamyltransferase [(gamma glutamyl)-peptide; amino acid gamma glutamyltransferase, EC 2.3.2.2.]. Clin Chem Acta. 1983; 15F-338F.
9. Kingsley GR. The direct biuret method for determination ofserum proteins as applied to photoelectric and visual colorimetry. J Lab Clin Med 1942; 27:840-5.
10. Silverman LM, Christenson RH. Amino acids and proteins.In:Burtis CA, Ashwood ER, editors. Tietz textbook of clinical chemistry. 2nd ed. Philadelphia: W.B. Saunders, 1993, 696-8.
11. Doumas BT, Peters T. Serum and urine albumin: a progressreport on their measurement and clinical significance. Clin Chem Acta. 1997; 258:3-20.
12. Sultana F, Anitha D, Venkatesh T. Estimation of reference values in liver function test in health plan individuals of urban south Indian population. Indian J Clin Biochem. 2004; 19(2):72-9.
13. Ashavaid TF, Todur SP, Dherai AJ. Establishment of reference intervals in Indian population. Indian J Clin Biochem. 2005; 20(2):110-8.
14. Yadav D, Mishra S, Gupta M, Sharma P. Reference intervals ofcertain liver specific biochemical analytes in Indian population. Indian J Clin Biochem. 2011; 26(1):97-8.
15. SaathoffE S, Philine S, Vera K, Steffen G, Dennis H. Laboratory reference values for healthy adultsfrom Southern Tanzania. Trop. Med. Intern. Health J. 2008; 13(5):612-25.
16. Siest G, Henry J, Schiele F, Young DS. Interpretation of Clinical Laboratory Tests: Reference Values and Their Biological Variation. Foster City, CA: Biomedical Publications, 1985.
17. Yadav D, Mishra S, Gupta M, John PJ, Sharma P. Establishment of Reference Interval for Liver Specific Biochemical Parameters in Apparently Healthy North Indian Population. Indian J Clin Biochem. 2013; 28(1):30-7.
18. Purkins L, Love ER, Eve MD, Wooldridge CL, Cowan C, Smart TS, *et al*. The influence of diet upon liver function tests and serum lipids in healthy male volunteers resident in a Phase I unit.Br J Clin Pharmacol2003; 57: 199–208
19. Chan-Yeung M, Ferreira P, Frohlich J, Schulzer M, Tan F. The effects of age, smoking, and alcohol on routine laboratory tests. Am. J. Clin. Pathol. 1981; 75(3):320-26.
20. Alsalhen KS, Abdalsalam RD. Effect of cigarette smoking on liver functions: a comparative study conducted among smokers and non-smokers male in El-beida City, Libya. International Current Pharmaceutical Journal. 2014; 3(7):291-5.
21. Suriyaprom K, Harnroongroj T, Namjuntra P, Chantaranipapong Y, Tungtrongchitr R. Effects of tobacco smoking on alpha-2-macroglobulin and some biochemical parameters in Thai males. Southeast Asian J. Trop. Med. Public Health. 2007; 38(5):918-26.
22. James D, Tania SB, Sara ET, *et al*. Alanine Aminotransferase Levels and Fatty Liver in Childhood Obesity: Associations with Insulin Resistance, Adiponectin, and Visceral Fat. J Clin EndocrinolMetab 2006; 91:4287-94.