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Research Article

Association of acyl-CoA-binding protein (ACBP) single nucleotide polymorphisms and type 2 diabetes in two German study populations

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The human acyl-CoA-binding protein (ACBP) is a potential candidate gene of type 2 diabetes (T2D), since it plays a central role in determining the intracellular concentration of activated fatty acids which contribute to insulin resistance. The aim of our study was to evaluate whether single nucleotide polymorphisms (SNPs) of the ACBP gene are associated with risk of T2D. Genotyping of eight SNPs (rs2084202, rs3731607, rs8192501, rs8192504, rs2244135, rs2276596, rs8192506, rs2289948) was performed in 192 incident T2D subjects and 384 matched controls of the European Prospective Investigation into Cancer and Nutrition-Potsdam cohort. A putative promoter SNP (rs2084202) of splice variant ACBP 1c showed decreased risk of T2D (odds ratio (OR) 0.63, 95% CI 0.41 – 0.96). The haplotype, that contained the mutant base of rs2084202 showed similar evidence for the association with disease risk as single SNP rs2084202. In a second population-based study, Cooperative Health Research in the Augsburg Region of 226 individuals with T2D and 863 control subjects a borderline significant association between rs2084202 and T2D (OR 0.72, 95% CI 0.51 – 1.01) was observed. In summary, we obtained evidence from two Caucasian study populations that the minor allele of ACBP rs2084202 might be associated with reduced risk of T2D.

Keywords: Acyl-CoA-binding protein / Association study / Haplotype / Single nucleotide polymorphism / Type 2 diabetes

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1 Introduction

The human acyl-CoA-binding protein (ACBP) is a highly conserved polypeptide with multiple physiological functions [1]. The protein was first isolated from rat brain as a diazepam-binding inhibitor (DBI) [2]. Since then it has also been described as a regulator of insulin release from pancreatic cells [3, 4], as a potent cholecystokinin-releasing peptide in the intestine [5, 6], and as a mediator in corticotropin-dependent adrenal steroidogenesis [7]. *In vitro* and *in vivo* studies suggest that ACBP acts as a pool former and transporter of acyl-CoA esters [8, 9]. Thereby, ACBP med-

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Abbreviations: ACBP, acyl-CoA-binding protein; **BMI,** body-mass-index; **EPIC,** European Prospective Investigation into Cancer and Nutrition; **KORA,** Cooperative Health Research in the Augsburg Region; **OR,** odds ratio; **SNP,** single nucleotide polymorphism; **T2D,** type 2 diabetes; **WHR,** waist–hip ratio

iates the transport of acyl-CoAs to enzymes such as acyltransferases. These enzymes are involved in fatty acid oxidation and lysophosphatidic acid synthesis in the mitochondria as well as the synthesis of phospholipids and cholesteryl ester in the endoplasmatic reticulum. ACBP also controls the intracellular level of unbound acyl-CoAs that interact with regulatory sites of enzymes and intracellular signaling proteins [10]. Besides its localization in the cytosol ACBP is present in the nucleus [11, 12], where a physical and functional interaction of ACBP with hepatocyte nuclear factor 4α has been previously described [13]. These data suggested a gene-regulatory role of ACBP. Although the ACBP gene is described to be a housekeeping gene, *in vitro* and *in vivo* studies showed that its expression is modulated by insulin, androgens [14, 15], feeding status [16], and status of cell differentiation [17]. In addition, the promoter of the human ACBP gene is activated by *PPARγ* and *SREBP-1* [18–20], which are the key regulators of genes involved in lipid metabolism. The responsiveness to lipogenic transcription factors and its involvement in lipid metabolism pathways imply ACBP as a functional candidate gene for type 2 diabetes (T2D). Genetic variations in this gene and the corresponding promoter may alter binding properties or expression levels of the protein and thereby modulate the level of free acyl-CoA. This cytotoxic metabolite influences the activity of insulin-mediated signal transduction pathways. To address our hypothesis, we systematically screened all exons including adjacent splice sites, and a promoter region including 877 bp upstream to the transcription initiation site of ACBP splice form 1c (M15887) for sequence variants. Single nucleotide polymorphisms (SNPs) were analyzed in two independent German study populations (European Prospective Investigation into Cancer and Nutrition (EPIC), Cooperative Health Research in the Augsburg Region (KORA)). Here, we present evidence that the minor allele of ACBP SNP rs2084202 is associated with decreased risk of T2D.

2 Materials and methods

2.1 EPIC study Potsdam (EPIC-Potsdam)

The EPIC-Potsdam study is a population-based, longitudinal study comprising a total of 27 548 people from the area of Potsdam, Germany. Baseline examinations, including anthropometric measurements, blood sampling, a selfadministered food frequency questionnaire, and a personal interview on lifestyle habits and medical history was conducted between 1994 and 1998 [21]. During the first follow-up, on average 2–3 years after recruitment, 192 incident cases of T2D were confirmed by the primary care physician as described before [22]. Cases were matched with two control subjects each by age and sex. Gender distribution of the final study population ($n = 576$) was 59% male and 41% female subjects with a mean age of 55.5 years (35–65 years). All study participants had given their informed consent and the genotype assessment was agreed to by the local ethic committee. Body weight, height, and fat percent were determined at recruitment by means of standardized procedures as described previously [21]. Detailed information on drug use was obtained at baseline and comprised all medications being taken during the previous 4 wk on the level of medication name. Sports activities were calculated from hours *per* week in summer and winter. HbA1c was determined using enzyme immunoassay (Dako Diagnostika, Hamburg, Germany).

2.2 KORA

The KORA Survey 4 (S4) studied a population-based sample of 4261 subjects aged 25–74 years during 1999–2001 [23]. Each study participant signed a consent form to participate in genetic studies. All study methods were approved by the ethics committee of the Bavarian medical association. The sampling design followed the guidelines of three previous surveys in the same region as part of the multinational World Health Organization (WHO)-MONICA (Monitoring Trends and Determinants of Cardiovascular Disease) study. In the age range 55–74 years, 1653 people participated in a standardized interview followed by biochemical and clinical analyses. An oral glucose tolerance test and biochemical and immunological analyses were performed as described previously [24]. Acute infections (fever) or gastrointestinal illness were an exclusion criterion for the oral glucose tolerance test. Diabetes was diagnosed according to 1999 WHO criteria. After the exclusion of all subjects with selfreported type 1 diabetes, humoral autoimmunity to glutamic acid decarboxylase, or diabetes onset in the context of pancreatitis, a total of 226 individuals with T2D were available for analyses. There were 863 normoglycemic control subjects randomly selected after matching for age and sex. Among the diabetic patients, 120 were newly detected and did not yet receive antidiabetic treatment; among the other 116, 33% were under insulin treatment and 57% took oral antidiabetic agents [24].

2.3 Genetic analyses

Six exons including the untranslated region and exon–intron boundaries, and the putative promoter region of the ACBP gene (M15887) were analyzed by terminator cycle sequencing, using Big Dye chemistry on an ABI 3700 capillary DNA sequencer (Applied Biosystems, Foster City, CA, USA) in 47 genomes of unrelated diabetic subjects from the Metabolic Intervention Cohort Kiel (MICK). Amplifications were performed using a "Touch-Down" PCR by decreasing the annealing temperature three times by 2°C, starting 2°C above the specific primer Tm. Genotyping of the EPIC-Potsdam subjects was performed with the TaqMan system (ABI, Foster City, CA, USA), fluorescence was

measured with ABI Prism 7900 HT sequence detection system. Genotyping of the KORA S4 study subjects was performed in the Genome Analysis Center (Munich-Neuherberg, Germany) using the Mass-ARRAY system (Sequenom) as described previously [25]. Genotyping error rate was <1% in 210 replicates. All sequences of primers and assay probes are available on request.

Figure 1. ACBP gene structure according to Nitz *et al.* [20] and location of genotyped SNPs.

2.4 Statistical analyses

Allele and genotype frequencies were determined by gene counting. Hardy–Weinberg-equilibrium was assessed in controls with χ^2 test. In EPIC-Potsdam, the association between disease and each SNP was calculated by conditional logistic regression analysis assuming additive and dominant models of inheritance for each allele. For each polymorphism, the frequent allele was designated as "wildtype" allele "1" and the minor allele as "2". Under the additive inheritance model, subjects with two wildtype alleles (11) were coded as "0", those with one wildtype and one mutant allele (12) were coded as "1", and those with two mutant alleles (22) were coded as "2". Under the dominant model, subjects with two wild-type alleles were coded as "0" and those carrying one or two mutant alleles were coded as "1". Since mutant alleles were rare, no recessive models were applied. Multivariate logistic regression analysis was conducted for adjusting the body-mass-index (BMI), waist–hip ratio (WHR), sports activities (h/week), total energy (kJ/d) and alcohol intake (g/d), and other noncontinuous, covariates as there were: presence of comorbidities (hypertension or/and hyperlipidemia), smoking (current smoker, nonsmoker, and former smoker), and use of lipid-lowering drugs (statins, fibrates, CSE-inhibitors, and others). Data on biochemical and anthropometric measurements were missing in six T2D and nine control subjects of the EPIC-Potsdam study and therefore, analysis of covariance (ANCOVA) was performed in a final study sample of 186 cases and 377 controls. HbA1c values were log-transformed prior to analysis. Haplotype frequencies were estimated by the expectation-maximization (EM) algorithm using SASGenetics software (SAS Institute, Cary, NC). Individual haplotypes were estimated using haplotype trend regression method (HTR) described by Stram *et al.* [26]. The association between each haplotype and T2D was analyzed by conditional logistic regression analysis with adjustment for other covariates. Significance level was set at $p < 0.05$. Statistics were computed with the Statistics Package for the Social Sciences 11.5 (SPSS, Chicago, IL, USA), SAS software 9.1 (SAS Institute) and S-PLUS 6.2 PROFESSIONAL EDITION (Insightful).

3 Results

Sequencing using DNA from 47 unrelated German subjects confirmed eight common SNPs located in the ACBP gene: rs8192506, rs8192501, rs3091405, rs3091406, rs3795890, rs2084202, rs3731608, and rs3731607. There was no novel SNP discovered during resequencing. Based on their respected allele frequencies, functional implications and assay availability, four SNPs confirmed through sequencing and additionally four validated SNPs from NCBI dbSNP were chosen for genotyping in a nested case-control study of 192 incident T2D patients and 384 controls of the EPIC-Potsdam cohort. Figure 1 shows the ACBP gene structure and positions of genotyped SNPs. Genotype distributions in EPIC-Potsdam study subjects are given in Table 1. SNPs rs3731607, rs2289948, and rs2276596 showed significant deviations from Hardy–Weinberg equilibrium and therefore were excluded from further analysis.

As shown in Table 2, SNP rs2084202 was significantly associated with decreased risk of T2D (odds ratio (OR)

Table 1. Genotype frequencies of ACBP polymorphisms

dbSNP ID	Genotypes *n* (%)				*p*	MAF
	11	12	22	Total (*n*)	$HWE_{Controls}$	
rs2084202	407 (72.0)	139 (24.6)	19 (3.4)	565	0.11	0.156
rs3731607	376 (66.1)	160 (28.1)	33 (5.8)	569	0.01	0.199
rs8192501	544 (94.8)	30 (5.2)	0	574	0.58	0.026
rs8192504	568 (99.1)	5 (0.9)	0	573	0.95	0.004
rs2276596	362 (65.6)	159 (28.8)	31 (5.6)	552	0.02	0.200
rs2244135	389 (70.2)	146 (26.4)	19 (3.4)	554	0.29	0.166
rs8192506	528 (92.3)	44 (7.7)	0	572	0.54	0.039
rs2289948	380 (66.2)	161(28.1)	33 (5.7)	574	0.01	0.198

HWE, Hardy–Weinberg-equilibrium; MAF, minor allele frequency.

Table 2. Associations of ACBP rs2084202 and rs8192506 with T2D in EPIC-Potsdam

Genotype	Cases *N* (%)	Controls *N* (%)	Crude OR (95% CI)	Adjusted OR[a)] (95% CI)	Adjusted OR[b)] (95% CI)
rs2084202 A>G					
AA *vs.*	145 (77)	255 (69)	1.0	1.0	1.0
AG	39 (21)	97 (27)	0.68 (0.44–1.05)	0.69 (0.43–1.12)	0.57 (0.32–1.00)
GG	3 (2)	16 (4)	0.30 (0.09–1.08)	0.34 (0.09–1.32)	0.35 (0.08–1.56)
AA *vs.* AG + GG	42 (23)	113 (31)	0.63 (0.41–0.96)*	0.65 (0.40–1.04)	0.54 (0.31–0.94)*
rs8192506 A>G					
AA *vs.*	169 (89)	355 (94)	1.0	1.0	1.0
AG	21 (11)	23 (6)	1.89 (1.03–3.47)*	1.97 (0.96–4.05)	1.92 (0.87–4.24)

Conditional logistic regression analysis (sex and age were matching variables).
a) Adjusted for BMI.
b) Adjusted for BMI, WHR, sports activities, total energy and alcohol intake, smoking, presence of comorbidities (hyperlipidaemia and hypertension), and lipid-lowering medication; *$p < 0.05$.

Table 3. Mean values (SEM) of anthropometric and metabolic variables according to ACBP rs2084202 and rs8192506 polymorphisms in EPIC-Potsdam

	rs2084202						rs8192506		
	AA	AG	GG	p_{trend}	AG + GG	*p*	AA	AG	*p*
Subjects (*N*)	405	139	19		158		520	43	
Men (%)	57	63	74		65		60	53	
Age (years)[a)]	55.6 (0.3)	55.0 (0.6)	58.2 (1.6)	0.74	55.4 (0.5)	0.75	55.4 (0.3)	56.8 (1.0)	0.18
BMI (kg/m^2)[b)]	28.14 (0.22)	27.73 (0.37)	27.94 (1.01)	0.42	27.76 (0.35)	0.36	27.98 (0.19)	28.64 (0.67)	0.34
WHR[b)]	0.908 (0.003)	0.909 (0.006)	0.903 (0.015)	0.96	0.908 (0.005)	0.93	0.907 (0.003)	0.921 (0.010)	0.16
Waist circumference (cm)[b)]	93.95 (0.58)	93.28 (1.00)	92.22 (2.71)	0.42	93.15 (0.94)	0.47	93.53 (0.51)	96.03 (1.79)	0.18
Body fat (%)[b)]	30.37 (0.28)	30.47 (0.47)	30.90 (1.28)	0.89	30.46 (0.44)	0.86	30.32 (0.24)	31.39 (0.85)	0.23
HbA1c (%)[c)]	5.1 (5.0, 5.2)	5.3 (5.1, 5.5)	4.9 (4.4, 5.4)	0.41	5.2 (5.0, 5.4)	0.16	5.1 (5.0, 5.2)	5.4 (5.1, 5.8)	0.07

Mean values (SEM).
a) Adjusted for sex.
b) Adjusted for sex and age.
c) Adjusted for sex, age, and BMI.

0.63, 95% CI 0.41–0.96) in the dominant inheritance model calculated. For rs8192506, the heterozygote genotype was significantly associated with increased risk of T2D in an unadjusted model (OR 1.89, 95% CI 1.03–3.47). For both SNPs, stepwise logistic regression analyses revealed that BMI did not change the OR markedly, although risk estimates became borderline significant (Table 2). When we adjusted for additional confounders, we obtained a significant association between rs2084202 and T2D (OR 0.54, 95% CI 0.31–0.94). As shown in Table 3, ANCOVA tests of ACBP polymorphisms (rs2084202, rs8192506) and BMI, WHR, waist circumference, body fat percent, and HbA1c values revealed no significant associations. Haplotypes were calculated from five ACBP SNPs (rs2084202, rs8192501, rs8192504, rs2244135, and rs8192506) and association was tested by logistic regression analysis. Haplotype II (Table 4) was significantly associated with reduced risk of T2D (OR 0.32, 95% CI 0.12–0.83).

ACBP polymorphism rs2084202 and rs8192506 genotypes were assessed in a second nested case-control sample taken from the KORA. A total of 226 patients with T2D and 863 normoglycemic subjects were included in the association analyses. In KORA, rs2084202 showed a borderline association with T2D (OR 0.72, 95% CI 0.51–1.01) (Table 5).

4 Discussion

ACBP was originally described as a diazepam-binding inhibitor. On the strengths of this function, plasma ACBP concentration was considered as a biological marker of epilepsy [27] and neuropsychiatric disorders [28]. Recently,

Table 4. ACBP haplotype frequencies and associations with T2D in EPIC-Potsdam

No.	Haplotype	Frequency		OR (95% CI)[a)]
		Cases	Controls	
I	1-1-1-1-1	0.783	0.757	1.39 (0.63–3.08)
II	2-1-1-2-1	0.118	0.172	0.32 (0.12–0.83)*
III	1-1-1-1-2	0.054	0.029	4.43 (0.86–22.92)
IV	1-2-1-1-1	0.022	0.027	1.35 (0.17–10.74)

Only the four most common haplotypes are given; the order of polymorphisms relates to the 5′ to 3′ direction of the gene and includes all SNPs which were in Hardy–Weinberg-equilibrium.
a) Adjusted for covariates described in legend of Table 2; *$p<0.05$.

Table 5. Associations of ACBP rs2084202 and rs8192506 with T2D in KORA

db SNP ID	Genotype	Cases *N* (%)	Controls *N* (%)	Crude OR (95% CI)	Adjusted OR (95% CI)[a)]
rs2084202	AA *vs.*	161 (75)	581 (68)	1.0	1.0
	AG	48 (22)	242 (28)	0.72 (0.50–1.02)	0.75 (0.51–1.09)
	GG	6 (3)	28 (4)	0.77 (0.31–1.90)	0.89 (0.35–2.29)
	AG + GG	54 (25)	270 (32)	0.72 (0.51–1.01)*	0.76 (0.53–1.09)
rs8192506	AA *vs.*	193 (96)	610 (96)	1.0	1.0
	AG	7 (4)	26 (4)	0.85 (0.26–1.99)	0.62 (0.24–1.62)

a) Adjusted for sex, age, and BMI; *$p = 0.06$.

multiple missense mutations in the ACBP gene were described in individuals with schizophrenia but no significant association with the disease was found [29]. An involvement of ACBP in the pathogenic mechanisms of peroxisomal deficiency disorders was also postulated [30]. In addition, ACBP is a prominent candidate for T2D because of its central role in determining free intracellular acyl-CoA concentrations that have been reported to inversely correlate with insulin sensitivity in muscle [31, 32]. According to the assumption that the alteration of acyl-CoA-binding properties, or expression levels of ACBP might be involved in the etiology of T2D, we examined the association of common and putative functional variants of the gene with T2D risk. We found a moderate but consistent association between one ACBP SNP (rs2084202) and T2D in two independent German study populations (EPIC-Potsdam and KORA). In both studies, carriers of the minor allele (A) showed lower risk of T2D compared to homozygote carriers of the major allele (GG). The obtained OR in the range of 0.5–0.7 and borderline significances are explained by limited statistical power on one hand, but on the other hand, might demonstrate the additional involvement of other (genetic or environmental) factors influencing gene-disease association. It is important to mention that we did not correct for multiple testing which obviously could lead to false-positive results by chance. Since we replicated our results in an independent study, a false-positive finding seems to be unlikely. However, replication studies in larger populations are desirable in the future. Associations between rs2084202 and related traits of the disease (*i.e.*, BMI, body fat percent) were not found. Also, adjustment for these confounders did not change the risk estimates markedly. These findings suggest that the effect of rs2084202 on T2D might not be related to body weight or fat mass. In fact, a direct function of the polymorphism on insulin resistance could be expected. SNP rs2084202 is located in the promoter region of splice variant ACBP-1c [20], 466 bp upstream from the translation start. We hypothesize that the A>G substitution of this promoter SNP may increase the transcriptional activity of this PPARγ-dependent variant which is highly expressed in adipocytes. This could lead to decreased free acyl-CoA concentrations in insulin sensitive cells such as hepatocytes or adipocytes and thereby reduce the risk to develop insulin resistance. Of course, our hypothesis has to be tested in future studies.

In conclusion, we obtained the first evidence from two Caucasian study populations that the minor allele of ACBP rs2084202 might be associated with reduced risk of T2D. The functional significance of this finding has to be unravelled in future studies.

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5 References

[1] Kragelund, B. B., Knudsen, J., Poulsen, F. M., Acyl-coenzyme A binding protein (ACBP), *Biochim. Biophys. Acta* 1999, *1441*, 150 – 161.

[2] Guidotti, A., Forchetti, C. M., Corda, M. G., Konkel, D. *et al.*, Isolation, characterization, and purification to homogeneity of an endogenous polypeptide with agonistic action on benzodiazepine receptors, *Proc. Natl. Acad. Sci. USA* 1983, *80*, 3531 – 3535.

[3] Chen, Z. W., Agerberth, B., Gell, K., Andersson, M. *et al.*, Isolation and characterization of porcine diazepam-binding inhibitor, a polypeptide not only of cerebral occurrence but also common in intestinal tissues and with effects on regulation of insulin release, *Eur. J. Biochem.* 1988, *174*, 239 – 245.

[4] Borboni, P., Condorelli, L., De Stefanis, P., Sesti, G., Lauro, R., Modulation of insulin secretion by diazepam binding inhibitor and its processing products, *Neuropharmacology* 1991, *30*, 1399 – 1403.

[5] Herzig, K. H., Schon, I., Tatemoto, K., Ohe, Y. *et al.*, Diazepam binding inhibitor is a potent cholecystokinin-releasing peptide in the intestine, *Proc. Natl. Acad. Sci USA* 1996, *93*, 7927 – 7932.

[6] Li, Y., Hao, Y., Owyang, C., Diazepam-binding inhibitor mediates feedback regulation of pancreatic secretion and postprandial release of cholecystokinin, *J. Clin. Invest.* 2000, *105*, 351 – 359.

[7] Besman, M. J., Yanagibashi, K., Lee, T. D., Kawamura, M. *et al.*, Identification of des-(Gly-Ile)-endozepine as an effector of corticotropin-dependent adrenal steroidogenesis: stimulation of cholesterol delivery is mediated by the peripheral benzodiazepine receptor, *Proc. Natl. Acad. Sci. USA* 1989, *86*, 4897 – 4901.

[8] Mandrup, S., Jepsen, R., Skott, H., Rosendal, J. *et al.*, Effect of heterologous expression of acyl-CoA-binding protein on acyl-CoA level and composition in yeast, *Biochem. J.* 1993, *290*, 369 – 374.

[9] Rasmussen, J. T., Faergeman, N. J., Kristiansen, K., Knudsen, J., Acyl-CoA-binding protein (ACBP) can mediate intermembrane acyl-CoA transport and donate acyl-CoA for beta-oxidation and glycerolipid synthesis, *Biochem. J.* 1994, *299*, 165 – 170.

[10] Knudsen, J., Neergaard, T. B., Gaigg, B., Jensen, M. V., Hansen, J. K., Role of acyl-CoA binding protein in acyl-CoA metabolism and acyl-CoA-mediated cell signaling, *J. Nutr.* 2000, *130*, 294S-298S.

[11] Elholm, M., Garras, A., Neve, S., Tornehave, D. *et al.*, Long-chain acyl-CoA esters and acyl-CoA binding protein are present in the nucleus of rat liver cells, *J. Lipid Res.* 2000, *41*, 538 – 545.

[12] Helledie, T., Antonius, M., Sorensen, R. V., Hertzel, A. V. *et al.*, Lipid-binding proteins modulate ligand-dependent trans-activation by peroxisome proliferator-activated receptors and localize to the nucleus as well as the cytoplasm, *J. Lipid Res.* 2000, *41*, 1740 – 1751.

[13] Petrescu, A. D., Payne, H. R., Boedecker, A., Chao, H. *et al.*, Physical and functional interaction of Acyl-CoA-binding protein with hepatocyte nuclear factor-4 alpha, *J. Biol. Chem.* 2003, *278*, 51813 – 51824.

[14] Swinnen, J. V., Esquenet, M., Heyns, W., Rombauts, W., Verhoeven, G., Androgen regulation of the messenger RNA encoding diazepam-binding inhibitor/acyl-CoA-binding protein in the human prostatic adenocarcinoma cell line LNCaP, *Mol. Cell. Endocrinol.* 1994, *104*, 153 – 162.

[15] Swinnen, J. V., Esquenet, M., Rosseels, J., Claessens, F. *et al.*, A human gene encoding diazepam-binding inhibitor/acy1-CoA-binding protein: transcription and hormonal regulation in the androgen-sensitive human prostatic adenocarcinoma cell line LNCaP, *DNA Cell. Biol.* 1996, *15*, 197 – 208.

[16] Bhuiyan, J., Pritchard, P. H., Pande, S. V., Seccombe, D. W., Effects of high-fat diet and fasting on levels of acyl-coenzyme A binding protein in liver, kidney, and heart of rat, *Metabolism* 1995, *44*, 1185 – 1189.

[17] Hansen, H. O., Andreasen, P. H., Mandrup, S., Kristiansen, K., Knudsen, J., Induction of acyl-CoA-binding protein and its mRNA in 3T3-L1 cells by insulin during preadipocyte-to-adipocyte differentiation, *Biochem. J.* 1991, *277*, 341 – 344.

[18] Helledie, T., Grontved, L., Jensen, S. S., Kiilerich, P. *et al.*, The gene encoding the Acyl-CoA-binding protein is activated by peroxisome proliferator-activated receptor gamma through an intronic response element functionally conserved between humans and rodents, *J. Biol. Chem.* 2002, *277*, 26821 – 26830.

[19] Swinnen, J. V., Alen, P., Heyns, W., Verhoeven, G., Identification of diazepam-binding Inhibitor/Acyl-CoA-binding protein as a sterol regulatory element-binding protein-responsive gene, *J. Biol. Chem.* 1998, *273*, 19938 – 19944.

[20] Nitz, I., Doring, F., Schrezenmeir, J., Burwinkel, B., Identification of new acyl-CoA binding protein transcripts in human and mouse, *Int. J. Biochem. Cell. Biol.* 2005, *37*, 2395 – 2405.

[21] Boeing, H., Korfmann, A., Bergmann, M. M., Recruitment procedures of EPIC-Germany. European Investigation into Cancer and Nutrition, *Ann. Nutr. Metab.* 1999, *43*, 205 – 215.

[22] Spranger, J., Kroke, A., Mohlig, M., Hoffmann, K. *et al.*, Inflammatory cytokines and the risk to develop type 2 diabetes: results of the prospective population-based European Prospective Investigation into Cancer and Nutrition (EPIC)-Potsdam Study, *Diabetes* 2003, *52*, 812 – 817.

[23] Muller, S., Martin, S., Koenig, W., Hanifi-Moghaddam, P. *et al.*, Impaired glucose tolerance is associated with increased serum concentrations of interleukin 6 and co-regulated acute-phase proteins but not TNF-alpha or its receptors, *Diabetologia* 2002, *45*, 805 – 812.

[24] Rathmann, W., Haastert, B., Icks, A., Lowel, H. *et al.*, High prevalence of undiagnosed diabetes mellitus in Southern Germany: target populations for efficient screening. The KORA survey 2000, *Diabetologia* 2003, *46*, 182 – 189.

[25] Weidinger, S., Klopp, N., Rummler, L., Wagenpfeil, S. *et al.*, Association of CARD15 polymorphisms with atopy-related traits in a population-based cohort of Caucasian adults, *Clin. Exp. Allergy* 2005, *35*, 866 – 872.

[26] Stram, D. O., Leigh Pearce, C., Bretsky, P., Freedman, M. *et al.*, Modeling and E-M estimation of haplotype-specific relative risks from genotype data for a case-control study of unrelated individuals, *Hum. Hered.* 2003, *55*, 179 – 190.

[27] Ferrarese, C., Cogliati, T., Tortorella, R., Zucca, C. *et al.*, Diazepam binding inhibitor (DBI) in the plasma of pediatric and adult epileptic patients, *Epilepsy Res.* 1998, *29*, 129 – 134.

[28] Guidotti, A., Role of DBI in brain and its posttranslational processing products in normal and abnormal behavior, *Neuropharmacology* 1991, *30*, 1425 – 1433.

[29] Niu, N., Rice, S. R., Heston, L. L., Sobell, J. L., Multiple missense mutations in the diazepam binding inhibitor (DBI) gene identified in schizophrenia but lack of disease association, *Am. J. Med. Genet. B Neuropsychiatr. Genet.* 2004, *125*, 10 – 9.

[30] Breitling, R., Pathogenesis of peroxisomal deficiency disorders (Zellweger syndrome) may be mediated by misregulation of the GABAergic system *via* the diazepam binding inhibitor, *BMC Pediatr.* 2004, *4*, 5.

[31] Oakes, N. D., Bell, K. S., Furler, S. M., Camilleri, S. *et al.*, Diet-induced muscle insulin resistance in rats is ameliorated by acute dietary lipid withdrawal or a single bout of exercise: parallel relationship between insulin stimulation of glucose uptake and suppression of long-chain fatty acyl-CoA, *Diabetes* 1997, *46*, 2022 – 2028.

[32] Ellis, B. A., Poynten, A., Lowy, A. J., Furler, S. M. *et al.*, Long-chain acyl-CoA esters as indicators of lipid metabolism and insulin sensitivity in rat and human muscle, *Am. J. Physiol. Endocrinol. Metab.* 2000, *279*, E554 – E560.

