tein measurements in black bears have not been reported. This study investigates the concentrations of CHOL and lipoproteins in denning and non-denning black bears. Serum CHOL concentrations were measured in 1052 serum samples drawn from wild black bears in both the denning and non-denning states. Non-denning bears were immobilized with ketamine (5 mg/kg) and xylazine (2 mg/kg) whereas denning bears were immobilized with ketamine (12 mg/kg) or tiletamine and zolezapam (4.5 mg/kg). Serum total lipids (TLIP), CHOL and lipoproteins were measured in ten denning bears (4 males, 3 females and 3 lactating females with cubs). CHOL and lipoproteins also were measured in a non-denning female. Denning bears were fasting whereas non-denning bears were trapped with high fat content foods and were non-fasting. Non-denning bears had significantly lower CHOL levels than denning bears. CHOL levels in denning bears were higher in early winter (Nov/Dec) than late winter (Feb/Mar). Males had higher CHOL levels than females during winter, but not during summer. Yearlings had lower CHOL levels than adults in winter. In the subpopulation of ten denning bears, TLIP were 1247 ± 454 mg/dl (range 639–2167); triglycerides (TRIG) were 351 ± 180 mg/dl (range 111–680); CHOL were 376 ± 162 mg/dl (range 175–695); HDL were 111 ± 35 mg/dl (range 82–185); LDL were 129 ± 66 mg/dl (range 47–228); and VLDL were 53 ± 22 mg/dl (range 22–87). HDL, LDL and VLDL levels were not ascertained in three bears because the precipitation technique was not possible and triglycerides were too high. The CHOL/HDL ratio was 2.8 ± 1.1 (range 1.7–4.8). Values at the lower range for TLIP, TRIG, CHOL, LDL and VLDL in this subgroup were found in two of the lactating females, who also had high HDL concentrations. Values for the non-denning female were TRIG 318, CHOL 298, HDL 210, LDL 24, VLDL 64 and CHOL/HDL 1.4. This bear recently had consumed 10 kgs of bacon. *Conclusions:* This study confirms previous reports that CHOL levels are high in black bears, especially during denning. Further, it demonstrates that, although CHOL, LDL and VLDL concentrations were in the range associated with development of human AHD, the HDL levels were much higher than observed in most human studies. In fact, the CHOL/HDL ratio was low to low-normal in all bears, including the bear which had consumed a high fat meal. These data imply that a high HDL concentration, along with a low CHOL/HDL ratio, confers protection against development of AHD in black bears despite high serum lipid concentrations. Further investigation could yield information of therapeutic benefit for man.

MOLECULAR BIOLOGY/RECEPTORS/HORMONES

901-88 Rapid Genetic Screen for Common β-Myosin Heavy Chain Mutations Causing Familial Hypertrophic Cardiomyopathy

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Hypertrophic cardiomyopathy (HCM), an autosomal dominant disease and the most common cause of sudden death in the young, is diagnosed by echocardiographic detection of cardiac hypertrophy. Unfortunately, the hypertrophy is usually not detectable until adolescence, which may be too late since sudden death often occurs as the first symptom. The availability of a clinical genetic assay to detect mutations would provide a definitive diagnosis, independent of clinical features and can be made at or before birth. Computer analysis showed most of the known mutations in the βMHC gene involve a restriction enzyme recognition site which can be exploited as a rapid genetic screening test. Accordingly, we employed polymerase chain reaction (PCR) to amplify the involved region of the gene followed by enzyme restriction digestion for 15 known mutations in 122 families with HCM. Fourteen families were positive and 108 negative. Ten families carried one of the three mutations known to have a high incidence of sudden death with 26 offspring having the mutation and at risk for sudden death while 49 did not have the mutation and are not at risk of developing HCM. Four families had one of the three known benign mutations with 23 offspring having the mutation and 21 without. The mutations were confirmed by sequencing and shown to be co-inherited with the disease. This test provides for a definitive genetic diagnosis and identifies those with the mutation and at risk for HCM and those without who will not develop HCM. Genetic counseling can be given prior to development of symptoms and those with mutations having a high incidence of sudden death are candidates for electrophysiology testing and possible implantation of cardiac defibrillators or amiodarone therapy. The test requires only a blood sample, is simple to perform and results are available in 48 hours. Over 25 of the βMHC mutations have now been shown to be amenable to rapid genetic diagnosis using PCR and restriction enzyme digestion.

Mutation	Prognosis	No. of Families
$Arg^{719}Trp$	malignant	4 (3.3% of total)
$Arg^{403}Gln$	malignant	3 (2.5% of total)
$Arg^{453}Cys$	malignant	3 (2.5% of total)
$Leu^{908}Val$	benign	2 (1.6% of total)
$Val^{606}Met$	benign	1 (0.8% of total)
$Gly^{741}Arg$	benign	1 (0.8% of total)

901-89 Enhanced Myocardial Relaxation In Vivo in Transgenic Mice Overexpressing the β_2-Adrenergic Receptor

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We have previously shown that the inotropic state of the intact mouse can be significantly increased with overexpression of the β_2-adrenergic receptor (β_2-ADR). However whether processes involving myocardial relaxation would also be influenced are unknown. Accordingly, transgenic mice that were created with cardiac-specific overexpression of the β_2-ADR (≅200 fold increase in β-ADR density and ≅2 fold increase in basal adenylyl cyclase activity) were studied using a 2F high fidelity micromanometer placed in the left ventricle (LV) of anesthetized intact transgenic (TG) (n = 7) and control (C) (n = 7) mice. Hemodynamic parameters measured were LV pressure, minimum first derivative of the LV pressure (LV dP/dtmin) and the time constant of isovolumic LV pressure decay (Tau) at baseline and after progressive doses of isoproterenol (ISO).

ISO	PSP (mmHg)		dP/dtmin (mmHg/s)		Tau (msec)	
	C	TG	C	TG	C	TG
basal	70 ± 3	70 ± 3	−3042 ± 196	−4766 ± 356	13.6 ± 1.5	8.2 ± 0.5
p*		ns		<0.001		<0.005
0.01 ng	90 ± 5	85 ± 5	−4233 ± 226	−6258 ± 628	13.4 ± 1.1	9.3 ± 0.7
p*		ns		<0.01		<0.01
1 ng	97 ± 5	79 ± 3	−5223 ± 655	−5434 ± 474	12.5 ± 1.4	9.9 ± 0.05
p*		<0.01		ns		ns

PSP = LV peak systolic pressure, Tau calculated using a mono-exponential model with zero asymptote, * ANOVA C vs. TG

In conclusion, overexpression of β_2-ADR resulted in a significant reduction in LV dP/dtmin associated with a shortening of Tau which was largely unaffected by ISO infusion. This suggests markedly enhanced myocardial relaxation in vivo which is most likely due to increased Ca^{2+} uptake into the sarcoplasmic reticulum.

901-90 In Vivo Genetic Engineering of Cardiac Cells: Intracoronary Administration of Antisense (AS) Oligonucleotides (ODN)

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We have previously documented that transfection of antisense ODN by a highly efficient Sendai virus (HVJ)-liposome delivery system can be utilized to modify lesion formation within the peripheral vasculature in vivo. In this study, we defined the feasibility of modifying cardiac cell gene expression via a catheter-based coronary infusion of AS ODN in rabbits. The coronary artery was cannulated via an over-the-wire approach from the carotid artery. Fluorescein (F)-labeled ODN were utilized to evaluate the cellular distribution and kinetics of ODN uptake within the myocardium after a single intraluminal bolus of HVJ-liposomes containing ODN. Cellular uptake of F-ODN was primarily localized in the microvasculature and significant staining was also observed in conduit vessels and cardiac myocytes. Immunohistochemical analysis verified prominent localization of F-ODN within the microvascular endothelium. Expression of F-ODN was observed within 10 minutes, peaked at 1 day, and remained evident for up to one week after transfection by the HVJ-liposome method. In contrast, F-ODN infused within liposomes without the viral particle exhibited transient expression that was undetectable within 3 days. These findings indicate that a single intracoronary bolus infusion of ODN within HVJ-liposomes is a reproducible methodology for delivery of AS ODN to targeted cells within the myocardium. Future studies will characterize the feasibility of using this approach to modify cardiac structure and function via regulating myocardial cell gene expression.