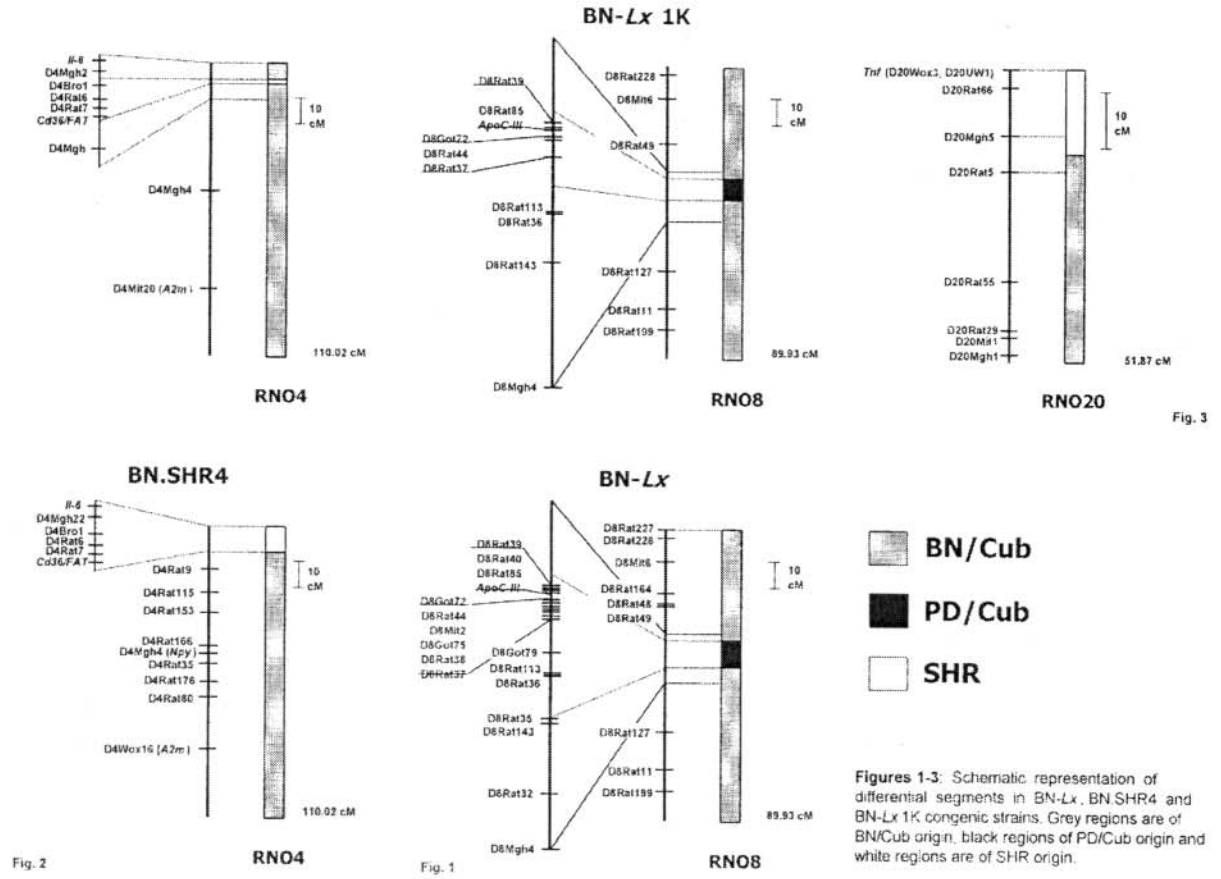

Figures 1-3: Schematic representation of differential segments in BN-*Lx*, BN.SHR4 and BN-*Lx* 1K congenic strains. Grey regions are of BN/Cub origin, black regions of PD/Cub origin and white regions are of SHR origin.

as described previously (Šedová et al., 2000a) in not fasted animals. The oral glucose tolerance test (OGTT) was performed after overnight fasting. Blood for glycemia determination was drawn from the tail at intervals of 0, 30, 60 and 120 min after intragastric glucose administration to conscious rats (3 g/kg TW, 30% aqueous solution). Commercially available analytical kits were employed to determine plasma glucose and serum triglyceride concentrations (Lachema, Brno, Czech Republic). Serum FFA were determined using an acyl-CoA oxidase-based colorimetric kit (Roche Diagnostics GmbH, Mannheim, Germany). At the end of the experiment, the rats were sacrificed and the weights of heart, liver, kidneys and epididymal fat pad were determined.

Genotypic analysis

Primers for the PCR detection of microsatellite markers used in this study were obtained from Research Genetics (Huntsville, AL) or synthesized in the University of California, San Francisco Biomolecular Resource Center according to published sequences. Further information about microsatellite markers and their polymorphisms among different rat strains can be found at: http://ratmap.gen.gu.se/ and http://www.resgen.com.

Statistical analysis

Comparison between two means were made by unpaired Student's *t*-test and between ≥ 3 means by one-way ANOVA followed by post-hoc (Tukey's) test. The null hypothesis was rejected whenever $P < 0.05$.

Results

Genotyping of RNO4, 8, 20 differential segments in BN congenic strains

BN-*Lx*. The **BN-*Lx*** strain differs from its progenitor **BN/Cub** in the introgressed segment of RNO8 of **PD/Cub** origin. The length of the differential segment of chromosome 8 was determined by PCR genotyping using mostly microsatellite markers or markers coding for real genes. Microsatellite markers of **PD/Cub** origin include: *D8Arb12, D8Arb23, D8Bord1, D8Mit2, D8Rat35, D8Rat36, D8Rat37, D8Rat38, D8Rat39, D8Rat40, D8Rat44, D8Rat84, D8Rat85, D8Rat111, D8Rat113, D8Rat156, D8Rat157, D8Rat158, D8Got72, D8Got75, D8Got79* and markers defining genes *ApoC-III* and *Sm22*. These primers span approximatelly 10–15 cM of chromosome 8 (Fig. 1). The length and the location of the differential segment was further verified by PCR genotyping, which showed that the primers of **BN/Cub** origin in the **BN-*Lx*** congenic strain are *D8Mit3, D8Mit4, D8Mit5, D8Mit6, D8Mgh1, D8Mgh4, D8Rat11, D8Rat16, D8Rat19, D8Rat20, D8Rat32, D8Rat41, D8Rat48, D8Rat49,*

D8Rat65, D8Rat71, D8Rat72, D8Rat75, D8Rat78, D8Rat103, D8Rat114, D8Rat124, D8Rat127, D8Rat135, D8Rat143, D8Rat149, D8Rat164, D8Rat199, D8Rat216, D8Rat227, D8Rat228 and *Mll*.

BN.SHR4. The length and location of the RNO4 differential segment in the **BN.SHR4** congenic strain was set by microsatellite markers listed in *Methods*. Except for eight RNO4 markers of **SHR** origin (*D4Bro1, D4Mgh22, D4Rat142, D4Rat6, Cd36/FAT, D4Rat7, D4Rat9* and *Il6*), the rest of the markers were found to be of **BN/Cub** origin. The differential segment spans approximately 10 cM of chromosome 4 (Fig. 2).

BN-Lx 1K. The lengths and locations of the RNO4, 8 and 20 differential segments in **BN-*Lx* 1K** congenic strain were set by microsatellite markers listed in *Methods*. On chromosome 4, only four markers were of **SHR** origin, *D4Rat6, D4Bro1, Cd36/FAT* and *D4Rat7*. On chromosome 20, the following markers were of **SHR** origin: *D20Wox3, D20Rat66* and *D20Mgh5*. Thus, the differential segment on chromosome 20 spans approximately 20 cM (Fig. 3). The differential segment on chromosome 8 was determined to span approximately 15 cM with markers *D8Rat39, D8Rat85, ApoC-III, D8Got72, D8Rat44* and *D8Rat37* being of **PD/Cub** origin.

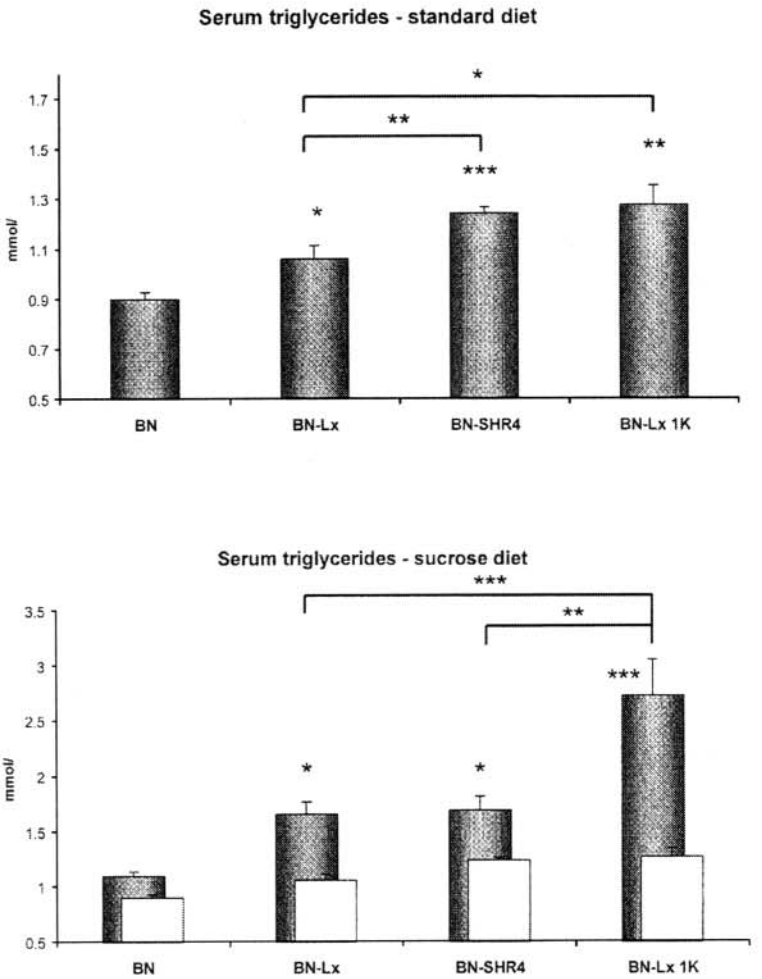

Fig. 4. Serum triglyceride levels (mmol/l) in not fasted animals after standard (upper figure) and high-sucrose (lower figure) diets. Values are given as mean ± SEM. Significant differences are shown as follows: *...P < 0.05; **...P < 0.01; ***... P < 0.001. Signs over the columns refer to comparisons with the BN/Cub strain. The white columns in the lower figure indicate the baseline triglyceride concentration (standard diet).

Phenotypic profiles of BN/Cub congenic strains

When fed standard diet, non-fasting serum triglyceride concentrations differed significantly among the strains (P < 0.01). The **BN/Cub** showed, as expected, the significantly lowest triglyceride levels (0.899 ± 0.027 mmol/l) when compared to all of the congenic strains. The **BN-*Lx*** congenic strain showed a significantly lower triglyceride concentration when compared to the **BN-*Lx* 1K** strain (1.058 ± 0.053 mmol/l vs 1.274 ± 0.079 mmol/l, respectively; Fig. 4).

After being fed high-sucrose diet for one week, all strains showed, as expected, a significant increase in triglyceride levels ranging from 21.59% in the **BN/Cub** strain to 136.60 % in the **BN-*Lx* 1K** triple congenic strain. At the same time, the differences in triglyceridemia among the strains were found to be very significant (P < 0.001), the **BN/Cub** strain exhibiting again the lowest concentrations and the **BN-*Lx* 1K** congenic strain the highest (Fig. 4). The serum concentrations of FFA (Fig. 5) were found to differ significantly among experimental strains after the high-sucrose diet (P < 0.0001) and followed a similar pattern, with **BN/Cub** showing the lowest (0.262 ± 0.019 mmol/l) and **BN-*Lx* 1K** the significantly highest (0.676 ± 0.029 mmol/l) values. No significant differences were found in total cholesterol levels both before and after high-sucrose diet (data not shown).

The oral glucose tolerance test showed significant differences between glucose levels at *time 0*, i.e. fasting glycaemia (P < 0.01) and *time 120*, i.e. 120 min after the glucose load (P < 0.0001) as well as in the area under the curve (P < 0.01), **BN/Cub** being the least glucose intolerant as compared to other strains. The course of OGTT in individual strains is depicted in Fig. 6.

The adiposity index, calculated as the weight of epididymal fat pad/100 g total body weight was, surprisingly, the significantly lowest in the **BN-*Lx* 1K** con-