Introduction
It has been shown that diabetes mellitus (DM) patients are often associated with olfactory dysfunction [1,2]. It has been reported that axonal transport in the olfactory bulbs of streptozotocin (STZ)-treated diabetic mice is impaired [3]. In this study, diffusion tensor imaging (DTI) is used to assess axonal water diffusion abnormality in the olfactory system in type-I diabetic rats induced by STZ injections.

Materials and methods
Animal preparation: Eight-week old male Sprague-Dawley rats, weighing 202-278 g, received intraperitoneal injections of either 62 mg/kg STZ dissolved in sodium citrate buffer (0.01 M, pH 4.5) (STZ group, n=10) or the same amount of sodium citrate buffer (control group, n=10). The treated animals were considered to be diabetic if their fasting blood glucose levels were higher than 223 mg/dl on day 3 after injections. MRI: MRI studies were performed at 2 weeks after diabetes induction on a 7.0 T/20 cm Bruker Biospec scanner with a 72-mm diameter volume coil for RF transmission and a quadrature surface coil for signal detection. High resolution anatomical images were acquired from axial slices of the olfactory bulb (OB) with a RARE sequence, TR 5800 ms, TE_{eff} 40 ms, matrix size 512×384, FOV 3.5 cm×3.5 cm, slice thickness 0.58 mm, RARE factor 4 and 8 averages. A spin-echo echo planar DTI sequence was used with the following acquisition parameters: TR 2000 ms, TE 26 ms, matrix size 128×128 (zero-filled to 256×256), FOV 3.0 cm×3.0 cm, slice thickness 0.8 mm, Δ 14 ms, δ 3 ms (b=0 and 800 s/mm²) and 4 averages. The diffusion-weighting gradients were applied on 30 non-collinear directions for encoding scheme. Data processing: With the DTI mapping software (DTIstudio), the fractional anisotropy (FA) maps were obtained. The high resolution T₂ weighted images were used to define the regions of interest (ROIs) which were manually outlined on FA maps. To characterize the axonal transport in the ROIs of the olfactory bulbs quantitatively, four diffusion indices, including FA, mean apparent diffusion coefficient (MD), axial diffusivity (ADC_{∥}) and radial diffusivity (ADC_{⊥}) were calculated and compared between the diabetic and control groups with Student’s t-tests.

Results
Figure 1 showed a FA map (Fig. 1A) and T₂ weighted image (Fig. 1B) acquired from a control rat. The five histological layers (olfactory nerve layer: ONL, glomerulus layer: GL, external plexiform layer: EPL, granule cell layer: GCL and subependymal zone: SEZ) were outlined on the T₂ weighted image. The ONL and GL were included in the ROIs delineated on the FA map. Fig. 1C plotted the quantitative changes of FA (the lateral or medial average values), MD, ADC_{∥} and ADC_{⊥} (the lateral average values) of bilateral OB. Compared to the control animals, the STZ-treated rats showed diffusion abnormalities in both the lateral and medial OB, indicated by the significantly reduced FA. Correspondingly, statistically significant increases in both MD and ADC_{⊥} but no significant changes in ADC_{∥} were associated with hyperglycemia.

Discussion
This result suggests the microstructure of the OB appear to be damaged as early as 2 weeks after STZ injections. In non-myelinated olfactory nerves, axonal membranes may play a role in anisotropic diffusion of water molecules [4]. The contribution of the deficit in axonal membranes and/or the loss of the neural fibres should be considered in the lower FA, higher MD and ADC_{⊥} in the OB of the diabetic rats. In conclusion, the DTI technology can detect pathological abnormalities in axons in the OB, especially the ONL and GL, and provide further insights into mechanisms of olfactory dysfunction in diabetes.

Acknowledgements
This work is supported by grants from Natural Science Foundation of China (30870674) and Knowledge Innovation Program of Chinese Academy of Sciences.

References