PO1808

The Dual Endothelin Type A Receptor (ET_AR) and Angiotensin II Type 1 Receptor (AT_1R) Antagonist, Sparsentan, Protects Against the Development of Albuminuria and Glomerulosclerosis in the gddY Mouse Model of IgA Nephropathy

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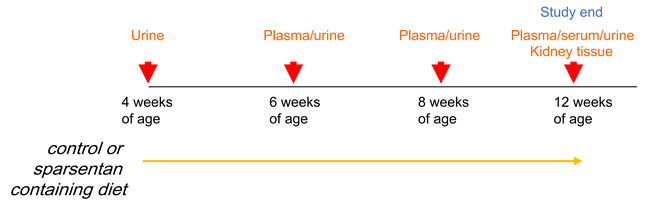
Background and Aims

gddY mice constitute an IgA nephropathy (IgAN)-prone mouse model that develops albuminuria between 4 and 8 weeks (wks) of age with glomerular IgA, IgG, and C3 deposits and glomerular injury.[1] A previous study in the ddY mouse model, the more genetically heterogeneous predecessor of gddY mice, using the endothelin type A receptor (ET_AR) antagonist, FR139317, resulted in amelioration of proteinuria and preservation of kidney function.[2] Treatment of ddY mice with the angiotensin II type 1 receptor (AT_1R) blocker, valsartan, resulted in significant protection from glomerulosclerosis (GS) without significant prevention of proteinuria.[3] Here we examined the effect of sparsentan (SP), a dual ET_AR and AT_1R blocker, currently in phase 3 trials for focal segmental glomerulosclerosis and IgAN, on the development of albuminuria and GS in gddY mice.

Methods

Study design: gddY mice at 4 wks of age were fed ad libitum with control (C) chow (n=5) or chow containing 900 ppm (n=10) or 1800 ppm (n=10) SP (approximately 180 and 360 mg/kg/day, respectively) for 8 wks. Albuminuria (U-Alb) was assessed at 4, 6, 8, and 12 wks of age, and plasma levels of SP were determined at 8 am and 4 pm at 6, 8, and 12 wks of age. Kidney tissue samples were taken at the end of the study at 12 wks of age for assessment of glomerulosclerosis, and serum was analyzed for content of IgA and for IgA galactose content.

Figure 1. Study design in gddY mice



Sample collection and analysis

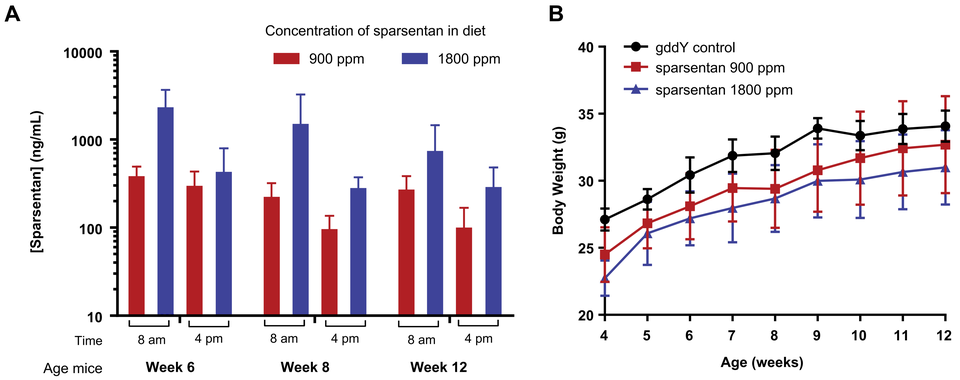
- Urine collected at 0, 2, 4, and 8 wks of treatment was analyzed for albumin using an Albumin ELISA (Exocell Inc., Philadelphia, PA, USA)
- At 12 wks of age, after 8 wks of treatment, kidneys were excised and processed for analysis. The extent of glomerular damage was assessed in 30 glomeruli per mouse using light microscopy in PAS-stained sections. Visual scoring of the percentage of glomerulosclerosis per glomerulus was as follows: 0=0%-<1%, 1=1%-<50%, 2=50%-<99%, 3=99%-100%. Data shown were taken from an unblinded assessment.
- Plasma was collected at 8 am and 4 pm at 2, 4, and 8 wks of treatment and was analyzed for sparsentan levels by Q[2] Solutions BioSciences, LLC (Indianapolis, IN, USA), using an LC/MS/MS method
- Serum was collected after 8 wks of treatment for determination of IgA content. Serum IgA levels were measured by ELISA (Bethyl Laboratories, Montgomery, TX, USA), and galactosylation of IgA was determined with an ELISA-based assay using biotinylated lectin-binding ricinus communis agglutinin (RCA)-I, as previously described[4]

Data analysis

- The total score of the 30 glomeruli assessed for glomerulosclerosis per mouse was reported. Statistical analysis was performed by one-way ANOVA and post hoc Tukey's test.
- Analysis of albuminuria was conducted on log-transformed data
- Statistical analysis of albuminuria and body weight was performed by two-way ANOVA and post hoc Dunnett's test
- A nominal *p* value <0.05 was considered significant

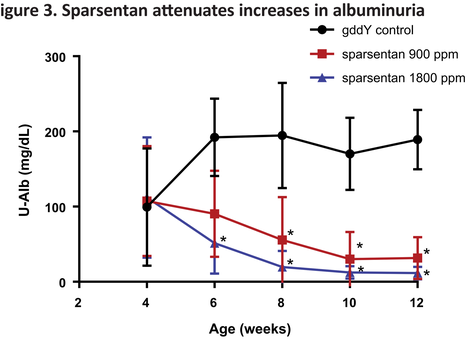
Results

Figure 2. Administration of sparsentan in chow to gddY mice results in efficacious plasma levels of sparsentan without effects on weight gain



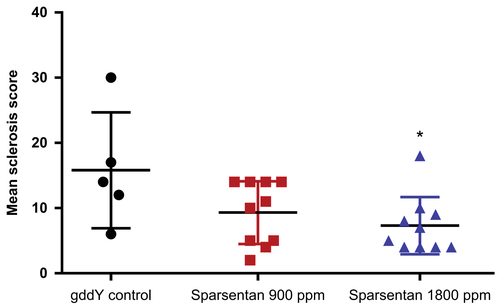
Data are shown as mean±SD. (A) Sparsentan concentration in plasma (n=5 at 6 and 8 weeks of age; n=10 at 12 weeks of age). (B) Weight gain from study start. gddY control: gddY mice fed regular chow.

Figure 3. Sparsentan attenuates increases in albuminuria



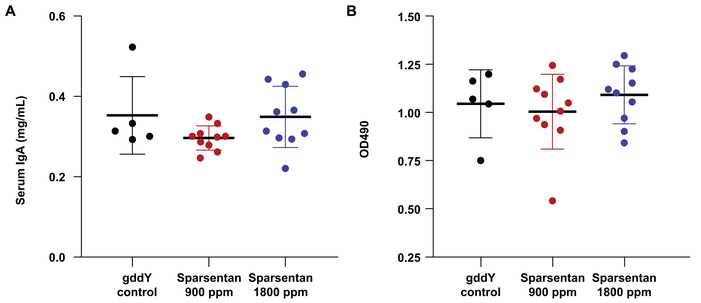
Data are presented as mean±SD. *p<0.05 vs gddY control.

Figure 4. Sparsentan prevents development of glomerulosclerosis in gddY mice



Data shown as mean±SD glomerulosclerosis score in 12-week-old gddY mice after 8 wks of treatment. Control mice are gddY mice fed regular chow.
*p<0.05 vs control gddY mice. Sparsentan 900 ppm was not significantly different from control mice.

Figure 5. Sparsentan does not alter serum total IgA or IgA galactose content in gddY mice



Individual and mean±SD values for (A) serum IgA, (B) galactosylated IgA (RCA-1 reactivity), in blood collected from mice at 12 wks of age (8 wks of treatment). OD 490 is the optical density at 490 nm. Control mice are gddY mice fed regular chow.

Conclusions

- Treatment of gddY mice with sparsentan from 4 to 12 wks of age attenuated the development of albuminuria and glomerulosclerosis associated with development of IgAN in the mice
- Sparsentan treatment had no effect on serum levels of total or aberrantly glycosylated IgA
- Results from this study, if translated to the clinic, may support the development of sparsentan as a new treatment approach for IgAN

References

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Acknowledgements

The authors would like to acknowledge Patricia Bedard for her help with statistical analysis. Editorial support was provided by Dolores Matthews, ELS, of MedVal Scientific Information Services, LLC (Princeton, NJ), and was funded by Retrophin, Inc.

Disclosures

CJ, KL, and RK: Employees of Retrophin, Inc., and may have an equity or other financial interest in Retrophin, Inc.The mouse gddY studies were conducted in the laboratories of Yusuke Suzuki, at Juntendo University, Japan, and were funded by Retrophin, Inc.

Data first presented at ERA-EDTA Virtual Meeting, June 6-9, 2020.