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ESPS Peer-review Report
Name of Journal: World Journal of Gastroenterology
Ms: 224
Title: Predomiant mucosal expression of 5-HT4(+h) receptor splice variants in pig stomach and colon
Reviewer code: 00001781
Science editor: x.z.huang@wjgnet.com
Date sent for review: 2012-08-17 20:46
Date reviewed: 2012-09-20 01:59

CLASSIFICATION	LANGUAGE EVALUATION	RECOMMENDATION	CONCLUSION
[Y] Grade A (Excellent)	[Y] Grade A: Priority Publishing	Google Search:	[ ] Accept
[ ] Grade B (Very good)	[ ] Grade B: minor language polishing	[ ] Existed	[ ] High priority for publication
[ ] Grade C (Good)	[ ] Grade C: a great deal of language polishing	[ ] No records	[ ]Rejection
[ ] Grade D (Fair)	[ ] Grade D: rejected	BPG Search:	[Y] Minor revision
[ ] Grade E (Poor)		[ ] Existed	[ ] Major revision
		[ ] No records	

COMMENTS

COMMENTS TO AUTHORS:
Summary This manuscript describes the expression of pig 5-HT4 receptor +h variant in the stomach and colon. The significance of this work is the use of LMPC and end-point PCR to allow the positive identification of the specific HTR4 variant (+ exon h) at discernable functional cell types relevant to serotonin response. This study also discussed several technique modifications and improvements and even artifacts in details, which is quite helpful for wider application of similar interests.
Comment
1. Need to better define h-exon structure and function as study rationale Pig HTR4 is less familiar to most readers than its human and rodent counterparts despite the authors have published an extensive work in Physiol Genomics. How is the cryptic exon h (42 bp) within exon 4-5 important needs to be addressed in the introduction. Comparison to its human HTR4 isoforms in terms of GI tissue distribution/physiology and 5-HT drug sensitivity would make the aim of the study better received. homo_sapiens:5
AAGGAAAGGA-GTCTAAACMAAGGCCTSGGCCAGGATTTTCATGYRGTATGTCAGAACAAGT
ATTCCTTAGGATTCTGAAAGAAATGCCG E R S L N K G L
G Q D F H V sus_scrofa:2
CAGGAAAGGACATCCAAACCAAGAC-TGGGCCAGGATTTGCATGTGGTATGTGAGAACACG
AGTTCCTTGGGCTTCTGAAGGAAATGCTT
E R T S K P R L G Q D L H V Fig 2A does depict other C-term isoforms but the ECL-2 sequence corresponding to h-exon (14 a.a.) is not included or should be added to Fig 2B.

2. Need to better distinguish 5-HT stained cell types MAB352 staining is primarily to identify 5-HT immunopositive cell types that would include mucosal EC cells, MMP neurons and/or blood-derived platelets. EC cells needs other endocrine marker such as chromogranin A (ChgA) as defined in Fig 3. Likewise, it would be desirable to include ChgA or TPH1 PCR in Fig 8C to demonstrate that there are indeed EC cells in pooled LMPC since GAPDH is a general house-keeping gene regardless its overamplification produced artifact band in Fig 8A.

3. Which C-terminal variants are in combination with up-regulated h-exon? Authors discussed the possibility of h-exon may coexpress with a, b, m, and r exons. Is any particular isoform dominant or all about the same in the GI mucosa at increased level?

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ESPS Peer-review Report
Name of Journal: World Journal of Gastroenterology
Ms: 224
Title: Predomiant mucosal expression of 5-HT4(+h) receptor splice variants in pig stomach and colon
Reviewer code: 00028211
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CLASSIFICATION	LANGUAGE EVALUATION	RECOMMENDATION	CONCLUSION
[ ] Grade A (Excellent)	[Y] Grade A: Priority Publishing	Google Search:	[Y] Accept
[ ] Grade B (Very good)	[ ] Grade B: minor language polishing	[ ] Existed	[ ] High priority for publication
[Y] Grade C (Good)	[ ] Grade C: a great deal of language polishing	[ ] No records	[ ]Rejection
[ ] Grade D (Fair)	[ ] Grade D: rejected	BPG Search:	[ ] Minor revision
[ ] Grade E (Poor)		[ ] Existed	[ ] Major revision
		[ ] No records	

COMMENTS

COMMENTS TO AUTHORS:

This manuscript investigated 5-HT4R distribution including splicing variants in porcine gastrointestinal tissue. Main conclusion is that h-exon containing 5-HT4(+h)R expressed higher than h-exon deleted 5-HT4(-h)R in mucosal region in both colon and gastric fundus. They used microdisssection and pressure catapulting (LMPC) and extracted RNA quality was evaluated by the Experion automated electrophoresis system. Obtained results are clear and quantitative, and the following discussion is logical and reasonable.