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LETTER TO THE EDITOR–REPLY

Response to: letter to the editor: culture and maintenance of taste cells in vitro

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The first issue raised by them is culture term of the cell lines. To my knowledge, "clonal cell lines" synonymously means "immortalized cell lines". The cell lines reported in our paper can be maintained "for ever" in culture by serial passages.

In general, clonal or immortalized cell lines are established from tumors. Introduction of oncogenes such as *c-myc* to normal cells also gives rises to immortalized cell lines. And occasionally long-term culture of cells of rodents spontaneously gives rise to immortalized cell lines. In addition, we found that clonal cell lines could be easily established by limited dilution from various tissues of *p53*-deficient mice (references were cited in our report). These cell lines are generally subjected to the first analysis of cell types: whether they are epithelial or fibroblastic when established from non-neural tissues, and whether they are neuronal or glial when established from neural tissues. The second analysis examines whether they express specific markers of their original tissues. For instance, when a cell line is established from a cerebellum, it is examined whether it expresses neurofilaments and then examined whether it expresses markers of specific neurons of cerebellum. The analyses may or may not be followed by neuronal physiology such as action potential. The first report of establishing cell lines does not always demonstrate almost all known phenotypes. Instead, later studies by the original or other researchers reveal many interesting features of the cell lines.

We have been analyzing the cell lines and have preliminary results showing that some of the cell lines respond to tastant stimuli in calcium imaging (unpublished observation). It is the bottle neck in molecular and in vitro study of taste buds that they are small in size containing small number of cells and, to make it worse, they are scattered in the tongue epithelium. Therefore, results of "comparable immunostaining in CD-1 mice" were not included in the report. Alternatively, we established clonal cell lines of the tongue epithelium and showed immunocytochemical differences between taste bud cell lines and tongue epithelial cell lines (Nishiyama et al. 2011 accepted).

Finally, the clonal cell lines were established from isolated taste buds, and our report demonstrated that they expressed some of specific markers of the taste bud. Therefore, they are clonal cell lines of taste buds or derived from taste buds.

Ozdener MH and Rawson NE raised several issues on our recent report published in In Vitro Cell Dev Biol Animal, 47:333–340 (2011).

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Reference

Nishiyama M.; Yuki S.; Fukano C.; Sako H.; Miyamoto T.; Tomooka Y. Attempt to develop taste bud models in three-dimensional culture. Zool. Sci. (2011, accepted)