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Commentary: Excessive Iodine Promotes Pyroptosis of Thyroid Follicular Epithelial Cells in Hashimoto's Thyroiditis Through the ROS-NF-κB-NLRP3 Pathway

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A Commentary on

Excessive Iodine Promotes Pyroptosis of Thyroid Follicular Epithelial Cells in Hashimoto's Thyroiditis Through the ROS-NF-κB-NLRP3 Pathway
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It is well-known that iodine is an indispensable element for thyroid hormone synthesis, and both deficiency and excess in iodine cause thyroid dysfunction. The article mentioned above used a human normal thyroid cell line, Nthy-ori 3-1, to study the effect of excessive iodine on thyroid cell death (1), where cell death was induced by incubating the cells with extremely high concentrations of iodine ($\geq 10^{-2}$ M). The same combination of Nthy-ori 3-1 cells and very high concentrations of iodine (up to 5×10^{-2} M) has also been previously used for studies on iodine-induced thyroid cell death, ER stress induction, etc. (2, 3).

I would like to raise two issues about this article. First is about the cell's capability of iodine uptake. The previous articles report that Nthy-ori 3-1 cells do not incorporate iodine (4, 5). Nthy-ori 3-1 cells were derived from HTori-3 cells which are immortalized human thyroid cells by transfection of simian virus 40 T-antigen (https://web.expasy.org/cellosaurus/CVCL_2659), and can grow without TSH. Thus, it is easy to assume that these cells are rather de-differentiated. In contrast, rat functional thyroid cell lines, FRTL5 and PCCL3, both of which are spontaneously immortalized cell lines and their growth is totally TSH-dependent, a characteristic of differentiated thyroid cells, take up iodine. We recently confirmed that PCCL3 cells incorporated ^{131}I in a dose-dependent manner, reaching the peak at 30 min in the presence of TSH (2 mU/mL), which was completely abolished by 5 μM $NaClO_4$ (perchlorate), a competitive inhibitor of sodium-iodine symporter (NIS), confirming NIS-mediated iodine uptake (6). Thus, I am afraid that the effect of iodine on Nthy-ori 3-1 cell death observed in (1) does not appear to be exerted by iodine incorporated into the cells.

Second is the concentration of iodine used. The physiological concentration of iodine in human sera is typically $2–6 \times 10^{-8}$ M (5–15 μg/L) (7), and pretreatment with 10^{-6} M or more iodine suppresses subsequent iodine uptake and expression of NIS (i.e., Wolff-Chaikoff effect) in FRTL5 cells (8) and porcine thyroid cells in suspension culture (9). 10^{-3} M iodine is generally used as an excessive dose in FRTL5 and PCCL3 cells, which suppresses the expression of other thyroid differentiation genes such as thyrotropin receptor, thyroglobulin, and thyroid peroxidase

TABLE 1 | Summary of iodine uptake and cytotoxicity of iodine in FRTL5/PCCL3 cells and Nthy-ori 3-1 cells.

	Iodine uptake	References	Cytotoxicity of iodine for 24 h	References
FRTL5	Yes	(8)	Yes, $\geq 3 \times 10^{-2}$ M	(15)
Nthy-ori 3-1	No	(4, 5)	Yes, $\geq 10^{-2}$ M	(1)

(10–12), and induces pendrin expression/iodine efflux (13). Thus, the concentration of iodine used in (1) is 10^6 times higher than the physiological serum concentration, 10^4 times higher than that required for the Wolff-Chaikoff effect, and $\geq$10 times higher than the excess dose used in FRTL5 and PCCL3 cells.

Despite of a clear difference in the ability of iodine uptake between FRTL5 and Nthy-ori 3-1 as mentioned above, cell death can only be induced by very high doses of iodine ($\geq 10^{-2}$ M) in both cells (1, 14). The ability of iodine uptake and cytotoxicity of iodine are summarized in the **Table 1**. In addition, human thyroid cells cultured in the presence of 1 mU/mL TSH are resistant to iodine-induced cell death with up to 3×10^{-2} M iodine for 24 h (15). Therefore, thyroid cells used in all the above reports are relatively resistant to a high dose of iodine, irrespective of their capability of iodine uptake. It is at present unknown as to how extremely high concentrations of iodine in the culture medium exert its cell killing effect in Nthy-ori 3-1 cells. Of interest, a recent paper has described the impairment of reproductive function in male rats by excess iodine, which is thought to be attributed to the aberrant expression of NIS in the testis (16).

Altogether, although it is understandable that one prefers to use the thyroid cells of human origin rather than of rodent origin, I would like to express my concern that Nthy-ori 3-1 cells may not be suitable for studies on the *in vitro* effect of iodine on thyroid cell function and/or survival.

AUTHOR CONTRIBUTIONS

YN has written the manuscript.

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Conflict of Interest: The author declares that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.