Oxygen is the key factor associated with the difference between in vivo and in vitro effects of antioxidants

Antioxidants have received much attention as food factors and drugs. Fox et al. (1) reported that antioxidants induced DNA damage response and cell death in cultured cells. I believe that their study has exceptional importance; however, at the same time, we must be very careful in evaluating antioxidants on the basis of experiments performed under air exposure. The oxygen (O_2) concentration in water at 37 °C is $\sim 200 \mu M$, and O_2 also dissolves from the air above the culture medium. Antioxidants easily reduce O_2 and generate hydrogen peroxide (H_2O_2) almost quantitatively, as evidenced by the fact that H_2O_2 is industrially produced by the reaction between O_2 and anthrahydroquinone derivative. Resveratrol, genistein, and baicalein share the same structure with anthrahydroquinone in having two or more hydroxyl groups at the aromatic ring. Therefore, most antioxidants behave as prooxidants when exposed to air. Because the rate of H_2O_2 formation depends on the antioxidant, each antioxidant behaves differently. Many studies have shown that antioxidants, including polyphenols and even vitamin C, induced apoptosis in cultured cells. I think that H_2O_2 , having extremely high biological activity, is the key factor that explains these effects.

The antioxidant concentration used in the study by Fox et al. (1) was $92 \mu M$, which could produce nearly $100 \mu M H_2O_2$, at which many effects, including apoptosis, could be caused.

Therefore, some of the reported effects are possibly explained on the basis of the amount of H_2O_2 produced. Considering these points, I would like to propose that an in vitro study of antioxidant effects be performed to determine the contribution of H_2O_2 . As an example, experiments under low- O_2 concentration are necessary in the future.

On the other hand, O_2 concentration in the cells of animal tissues is far less than 40 mmHg (O_2 pressure in the vein); furthermore, O_2 concentration is 2–5% even in the arterial walls (2). Under low- O_2 concentration, the formation of H_2O_2 is largely delayed, thus enabling an antioxidant to function as an antioxidant. It is also well known that O_2 concentration in cancer cells is much less than that in normal cells. In addition, all antioxidants used in the study (1) are xenobiotics and their tissue concentration is assumed to be far less than $92 \mu M$. Therefore, the statement “Our findings thus raised the possibility that resveratrol, baicalein, and genistein could be better used as cancer drugs” in the discussion section of the study by Fox et al. (1) may be controversial.

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