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Ultra-weak photon emission from biological samples: Definition, mechanisms, properties, detection and applications

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ABSTRACT

This review attempts to summarize molecular mechanisms, spectral and intensity properties, detection techniques and applications of ultra-weak photon emission. Ultra-weak photon emission is the chemiluminescence from biological systems where electronically excited species are formed during oxidative metabolic or oxidative stress processes. It is generally accepted that photons are emitted (1) at near UVA, visible, and near IR spectral ranges from 350 to 1300 nm and (2) at the intensity of photon emission in the range of several units to several hundreds (oxidative metabolic process) and several hundreds to several thousands (oxidative stress process) photons s^{-1} cm^{-2}. Current development in detection using low-noise photomultiplier tubes and imaging using highly sensitive charge coupled device cameras allows temporal and spatial visualization of oxidative metabolic or oxidative stress processes, respectively. As the phenomenon of ultra-weak photon emission reflects oxidative metabolic or oxidative stress processes, it can be widely used as a non-invasive tool for monitoring of the physiological state of biological systems.

1. Introduction

It is fascinating to observe that biological systems continuously emit very weak light without any external stimuli or additionally applied external luminophores and that the intensity of this weak endogenous light is modified by application of external stressors. This phenomenon, currently commonly termed as ultra-weak photon emission, is interesting even more because it is present virtually in all metabolically active systems from the level of bacteria [1,2], fungi [3], germinating seeds [4], whole plants [5–7], animal tissue cultures [8] and whole organisms [9] including human beings [10–13].

Obviously, one is curious about the physical and chemical mechanisms which underlie such general phenomenon such as the ultra-weak photon emission from biological systems. Understanding the mechanism for generation of electronically excited species together with the quantification of spectra, intensity, spatial and temporal distribution, and statistical parameters of photon signals from biological systems under various conditions can lead to potential application in non-invasive probing and diagnostics ranging from agriculture to biomedicine. To provide a state of the art introduction to the research of ultra-weak photon emission, we review molecular mechanisms, spectral and intensity properties, detection techniques and pioneering applications of ultra-weak photon emission.

2. Definition, terminology and types of ultra-weak photon emission

2.1. Definition

Ultra-weak photon emission originates from the oxidative metabolic reaction in microbial, plant and animal cells [7,14–16]. It is generally considered that electronically excited species formed during the oxidative metabolic processes are solely responsible for the ultra-weak photon emission. Spontaneous photon emission without any special dedicated high-intensity-luminescent enzymatic systems (e.g. luciferin/luciferase) is what distinguishes ultra-weak photon emission from ordinary bioluminescence. Photon emission without external stimulation by light is a feature that distinguishes ultra-weak photon emission from fluorescence and phosphorescence. Experimental evidence for other types of luminescence than chemiluminescence (for instance mechanoluminescence [17,18] and electroluminescence [19,20]) in biological systems is very limited [17,18,21].

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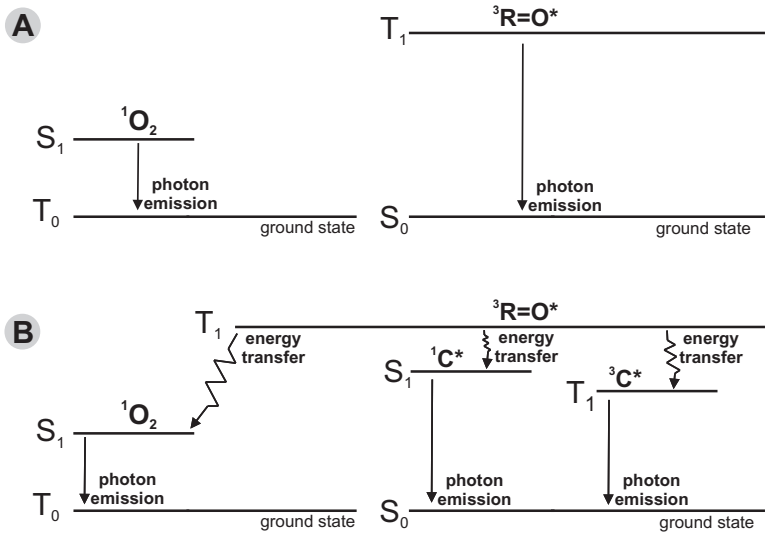

Fig. 1. Singlet and triplet excited level of electronically excited species formed during oxidative metabolic and oxidative stress processes in cells. (A) The energy level of triplet excited carbonyles (^{3}R = O*) and energy level of singlet oxygen (^{1}O$_2$). (B) Alternatively, the energy of excited triplet carbonyl can be transferred to pigment forming singlet excited or triplet excited pigment or to molecular oxygen forming singlet oxygen. Singlet or triplet excitation energy of electronically excited species is emitted in the form of near UVA, visible or infrared photons. S_0 ... singlet ground state, S_1... singlet excited state, T_1 ... triplet excited state.

The electronic transition of electronically excited species formed during the oxidative metabolic processes either from the singlet or triplet excited state to the singlet ground state is accompanied with photon emission at short and long wavelength regions of the spectrum, respectively (Fig. 1). When other molecules such as pigments or molecular oxygen are in the close proximity to electronically excited species formed during the oxidative metabolic processes, excitation energy transfer results in the formation of excited pigments or singlet oxygen (^{1}O$_2$).

2.2. Terminology

Often other terms than ultra-weak photon emission are used in the literature to describe the phenomenon (Table 1). The choice of the term expresses usually authors' attitude to the phenomenon: low level chemiluminescence [22,23] puts stress on the origin of the ultra-weak photon emission from chemical reactions; autoluminescence [24] emphasizes that the origin of the ultra-weak photon emission is without any external stimuli; biophoton emission [25,26] highlights the biological origin and/or function of the ultra-weak photon emission process.

2.3. Types

One can distinguish two types of ultra-weak photon emission: spontaneous and induced. Spontaneous ultra-weak photon emission can be defined as the one which is generated in the course of the oxidative metabolism without any influence of external stressors or stimuli. Induced ultra-weak photon emission can be initiated by various biotic and abiotic stresses and oxidative factors. The biotic factors include viral [5], bacterial [27] and fungal infestation of organism [28] or herbivorous stress [29]. The abiotic factors include the temperature [30] and gas composition of the surrounding environment [30], mechanical damage [31], stress by light [24,32,6] as well as ionizing radiation [33]. All these factors lead, among other effects, to the increase of ROS production and oxidative damage. The intensity of the induced ultra-weak photon emission can be several orders of magnitude higher than that of the spontaneous. After induction, the increased intensity of the ultra-weak photon emission undergoes either partial or full recovery to the original intensity. One has to acknowledge a difference between delayed luminescence and light-induced ultra-weak photon emission. While light-induced ultra-weak photon emission originates from oxidative stress process similar to processes that produce other types of stress induced ultra-weak photon emission and involve chemical reactions, delayed luminescence is essentially only other term for extremely long lived phosphorescence (hundreds of seconds). Time scales of the two overlap and therefore it may not be straightforward to separate these two processes.

Table 1
Alternative terms for ultra-weak photon emission used by various authors.

Alternative terms for ultra-weak photon emission	References
Autoluminescence	[24]
Weak luminescence	[47]
Biophotons/biophoton emission	[26,25]
Low-level chemiluminescence	[22]
Spontaneous chemiluminescence	[73]
Spontaneous ultra-weak light emission	[74]
Ultra-weak bioluminescence	[33]

Table 2
Electronically excited species responsible for ultra-weak photon emission.

Electronically excited specie	Wavelength	Ref.
Triplet excited carbonyls	350–550 nm	[36,75]
Singlet excited pigments	360–560 nm melanin	[76,77]
	680–740 nm chlorophyll	[78]
Triplet excited pigments	870–1000 nm chlorophyll	[+]
Dimolar singlet oxygen	634 nm, 703 nm	[22,23,37]
Monomolar singlet oxygen	1270 nm	[*]

[+] expected based on the singlet excited pigment emission but not yet directly confirmed as a contribution to ultra-weak photon emission. Emission wavelength from triplet excited chlorophyll is known from phosphorescence measurement [79].

[*] expected based on the dimolar oxygen emission but not yet directly confirmed as a contribution to ultra-weak photon emission. Emission wavelength of monomolar singlet oxygen is known [80].