PHOTON®

INSTANT

Knowing the disruptive technology behind Photon

BIONOVA®

Terragene®

The Bionova® Photon BT225 is a self-contained biological indicator (SCBI) for monitoring steam sterilization processes with a reading time of 7 seconds.

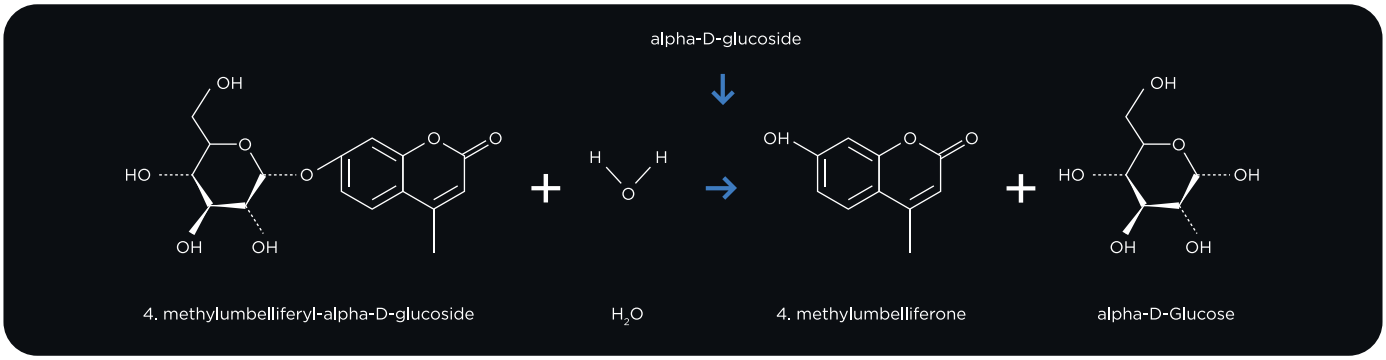
This product was designed for controlling both pre-vacuum and gravity displacement steam sterilizers between 132-135 °C and was designed following these international quality Standards: ISO 13485:2016, ISO 11138-1:2017, and ISO 11138-3:2017.

The Bionova® Photon BT225 BI makes use of *Geobacillus stearothermophilus* bacterial spores and a specific fluorophore capable of interacting in a differential manner with different spore-associated proteins. The spores are placed in a carrier at the bottom of the tube and the fluorophore (developer) in the culture medium contained in a glass tube. This culture medium allows determining both the germination and growth of the spores as well as the efficacy of the sterilization cycle instantly, which makes it a dual indicator as well as instantaneous. The culture medium predicts the structural change of the various proteins associated with the spore by fluorescence readouts. The transformation of these proteins is directly related to the conditions of the sterilization cycle.

Once exposed, the indicator must be placed in the dedicated incubator Bionova® BPH Instant™. It can maintain a stable average temperature of 60 °C, irradiating with light at a wavelength of 375 nm and recording fluorescence emission between 400 and 540 nm. If the sterilization process has not been successful, in addition to the instantaneous indication of failure given by the BPH Instant™ Auto-reader, the biological indicator Bionova® Photon BT225 culture medium will change its color to yellow after 48 hours of incubation at 60 °C, indicating the presence of living spores. If sterilization is successful, the culture medium will remain purple after this incubation process.

A radical technological shift to everything known in Biological Indicators with fluorescence technology: Traveling to the heart of Photon's biotechnological principle.

Traditional rapid biological indicators (BIs) use enzymes associated with spores capable of performing specific biochemical reactions. If the spores are alive (in the case of a BI not exposed or subjected to an inefficient sterilization cycle), these enzymes will remain active and carry out these biochemical reactions. The clear and well-known example is the specific enzymatic reaction between α-glucosidases (enzyme naturally present in spores of *Geobacillus stearothermophilus*) on the non-fluorescent compound α-MUG (4-methylumbelliferyl-alphaD-glucopyranoside) to produce the fluorescent compound 4-MU (4-methylumbelliferone) and glucose.



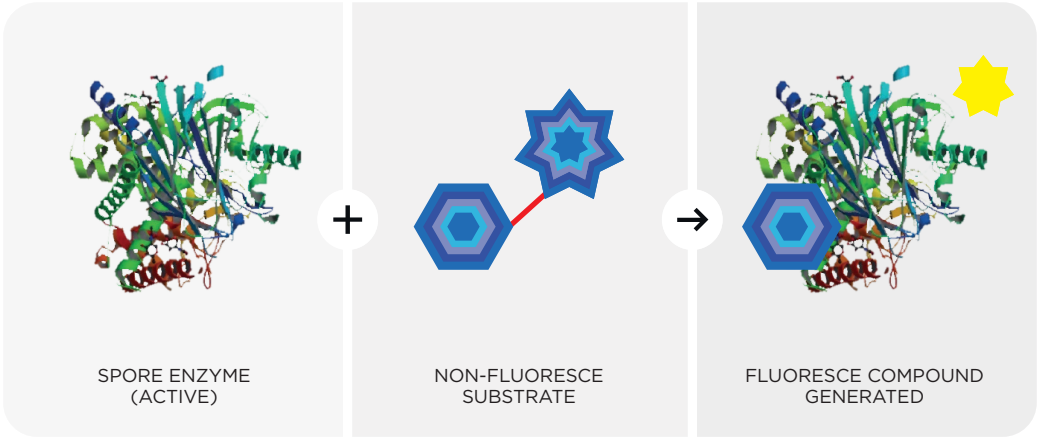
In this way, only these SCBI with viable spores can give a detectable fluorescence signal. Those with dead or inactivated spores will not catalyze that reaction and therefore generate that fluorescent signal.

Bionova® Photon BT225 technological principle is not based on an enzymatic reaction catalyzed by spore-associated enzymes (as described above), but on the non-covalent interaction between different spore-associated proteins and a sensor compound present in the culture medium. Physical sterilization processes such as steam generate drastic changes in the structure of all these proteins that result in their denaturation. The magnitude of the structural changes is directly proportional to the sterilization times and conditions, but the most important thing for this technology is that it can be measured as soon as the process is completed. The specific sensor compound developed in the biological indicator Bionova® Photon BT225 can interact with different regions of these proteins depending on the polarity of their environment, changing its fluorescent properties. We describe here a hydrophobic sensor that emits a higher fluorescent signal (or significant) when is in a nonpolar environment like when it associates with these spore proteins in their "natural shape" or their native state. In a simplified way, in living spores, these proteins are in their native state and can interact with this compound, giving a high fluorescent signal. When the spores are subjected to the sterilization process, these proteins suffer an "irreversible deformation" of their three-dimensional structure (denaturation) that prevents them from interacting with this novel compound, thus generating an almost zero (or minimal) fluorescent signal.

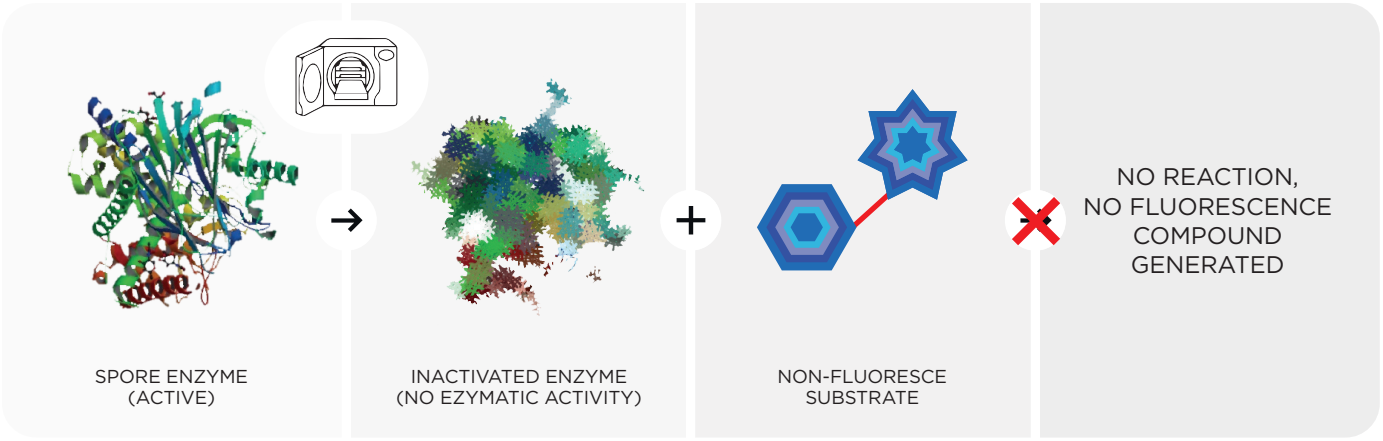
The following figure shows, in a simplified way, a comparison between the traditional fluorescence system and the one corresponding to Photon in order to graphically understand the differences between both systems.

Traditional System

Positive fluorescence readout: Unexposed or Living spores

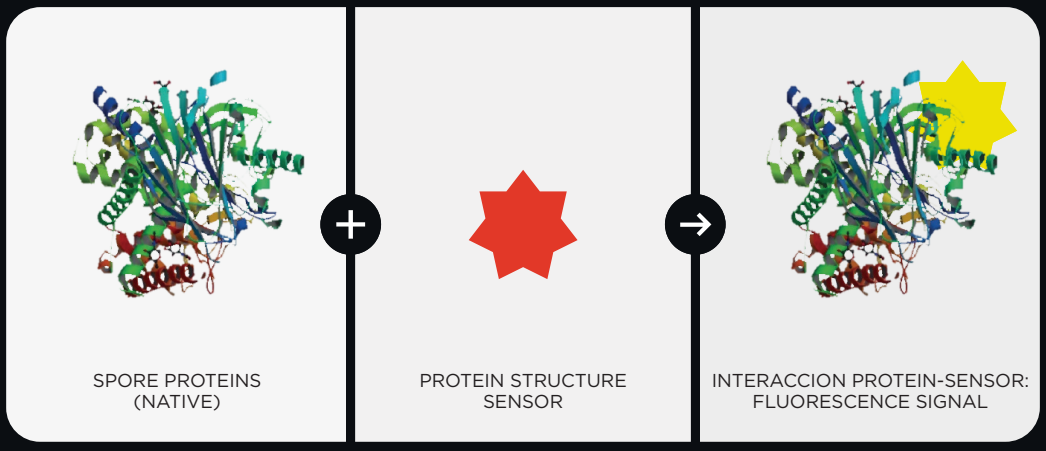


Negative fluorescence readout: Sterilized

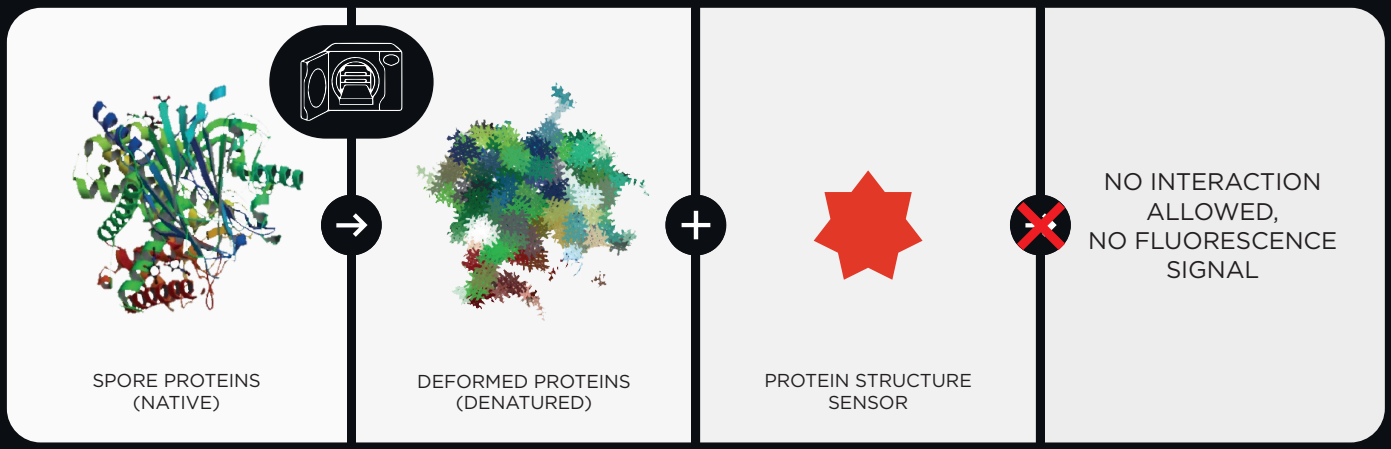


Photon System

Positive fluorescence readout: Unexposed or Living spores

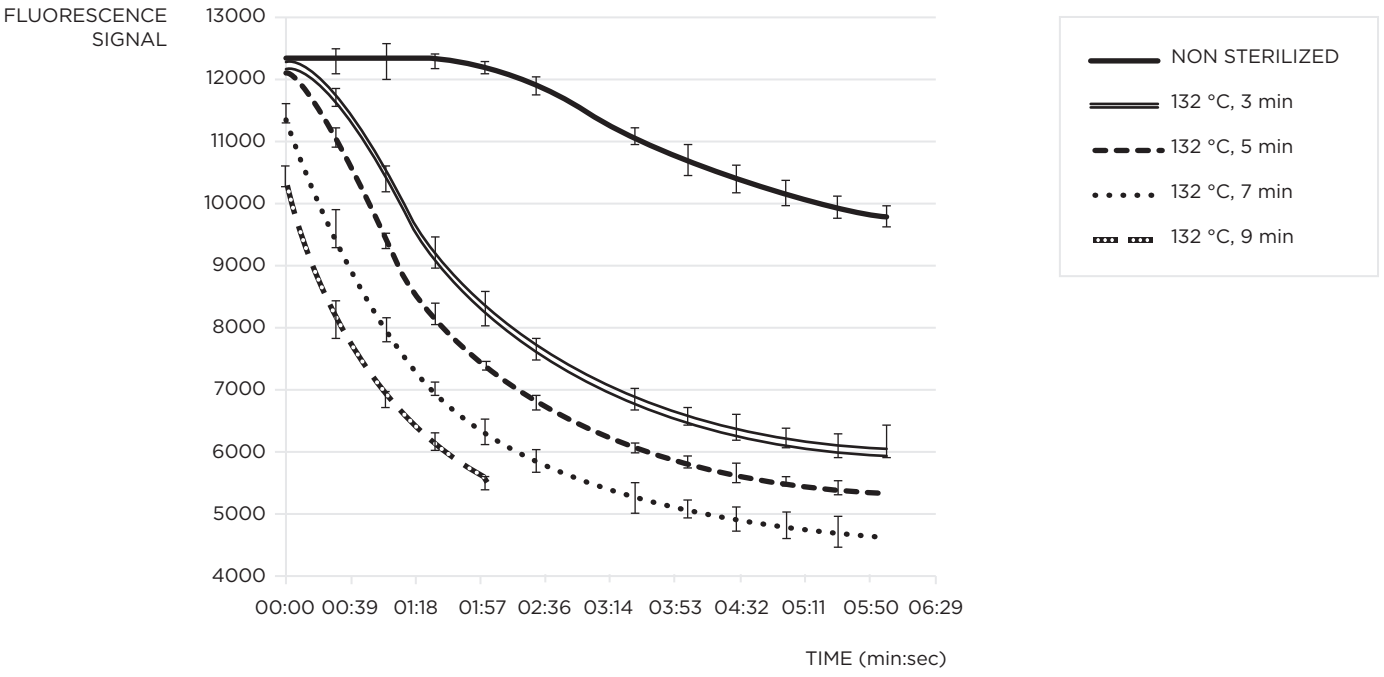


Negative fluorescence readout: Sterilized



As explained above, the effectiveness of the sterilization process is directly related to the denaturation of these spore-associated proteins. Hence, the fluorescence change generated by the interaction of these proteins with the sensor guarantees the prediction of the *G. stearothermophilus* spore population death.

The following figure shows the fluorescent signal measured at several incubation/reading times of different Photon BIs not exposed (not sterilized) or exposed to sterilization cycles at 132 °C for 3, 5, 7 and 9 minutes. The fluorescence response of indicators exposed to these sterilization cycles can thus be observed. By comparing the different responses, it is evident that the more lethal the sterilization cycle (longer exposure time), the less the fluorescent signal emitted.



In the graph, we can observe a marked difference between the fluorescent signal of unexposed indicators and the signal of those exposed to different sterilization cycles. The 7-second reading time with Photon allows maximizing monitoring safety with unprecedented speed.

Let’s work together to create a better future

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