# Estimating Bacterial Surface Contamination by Means of ATP Determinations: 20 p short of a pound.

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Mulvey and co-workers[1] have recently added their voices to calls for the imposition within healthcare facilities of quantitative standards for monitoring surface cleanliness. These are generally expressed as surface concentrations of heterotrophic aerobic bacteria and quoted as colony forming units (CFU) per $cm^2$. However, these authors have gone further and suggest that such standards might be formulated in terms of 'relative light units' (RLU) - the units in which the concentrations of adenosine tri-phosphate (ATP) are normally quoted. ATP has been referred to as the universal energy 'currency' and its detection is taken as indicative of the presence of living cells. The direct, culture-based, method of obtaining heterotrophic counts is widely seen as inherently impractical because of the delays – which may be as long as 48 hours – before colonies have reached a size that permits counting. By contrast, measurement of surface ATP levels based on detection of luminescence during the reaction of luciferin with luciferase may be conducted in minutes using hand-held luminometers and combined swab and reagent kits. In fact it is not difficult to see how one might become beguiled by the apparent ease that the technology of ATP testing offers.

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It is widely acknowledged that surfaces can also be contaminated with traces of body fluids and food residues all of which can contribute to ATP readings. There is also awareness that chlorine-containing surface disinfectants are able to attenuate the light signal generated and so confound measurement of the true ATP concentration. These factors aside, the notion that a direct equivalence exists between ATP concentration and microbial surface concentrations is one that needs to be critically examined.

The first consideration is that only a small proportion of micro-organisms present in any given environment is actually culturable using synthetic growth media. For exotic environments such as lake sediments and termite guts the culturable proportion has been estimated as less than 1 %. Although the culturability of the human microbiota is somewhat higher, it can be as low as 10 % for that of saliva and the gastrointestinal tract[2] and only marginally higher for human skin[3]. Traces of faeces, saliva and desquamated skin cells would be common contaminants of objects in the near-patient environment. Viable unculturable micro-organisms from such sources would contain ATP and therefore contribute to surface ATP levels but would remain undetected by conventional culture-based assays.

An additional consideration is that intracellular ATP levels are not identical for all micro-organisms. In fact there appears to be a correlation with cell volume, and eukaryia such as yeast with their characteristically larger cell size will contain higher levels of ATP than bacteria. There is also evidence to suggest that Gram +ve bacteria contain higher levels than Gram –ve bacteria, and that bacterial spores contain only relatively low levels of ATP[4]. The media used for hetertrophic counts tend to be selective for bacteria and may therefore fail to permit certain yeast to grow

whilst their ATP contribution will nonetheless be detectable. In contrast, bacterial spores will yield only low ATP levels but may well germinate and go on to form colonies on the media used to obtain surface counts.

Finally, the ATP content of a cell should not be taken as a fixed entity as organisms are apparently able to regulate ATP levels in response to environmental stresses. It has been reported that when starved of nutrients *Mycobacterium tuberculosis* was able to downregulate its ATP levels to one fifth that of normal[5].

With the discrediting of visual methods of assessing the cleanliness of surfaces, the formulation of hygiene standards in terms of ATP measurements would appear to be a positive step forward. Moreover, it would not be inconceivable that conditions might arise when it might be safely assumed that a direct relationship existed between surface ATP levels and microbial counts. However, for the reasons presented above such occurrences are likely to be very rare indeed.

References


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