MICROBIAL VIABILITY:
THE UNCERTAINTY THAT NEEDS TO BE MEASURED

Evgeny Puchkov

All-Russian Collection of Microorganisms,

G.K. Skryabin Institute of Biochemistry and Physiology of Microorganisms RAS,

Pushchino, Russia

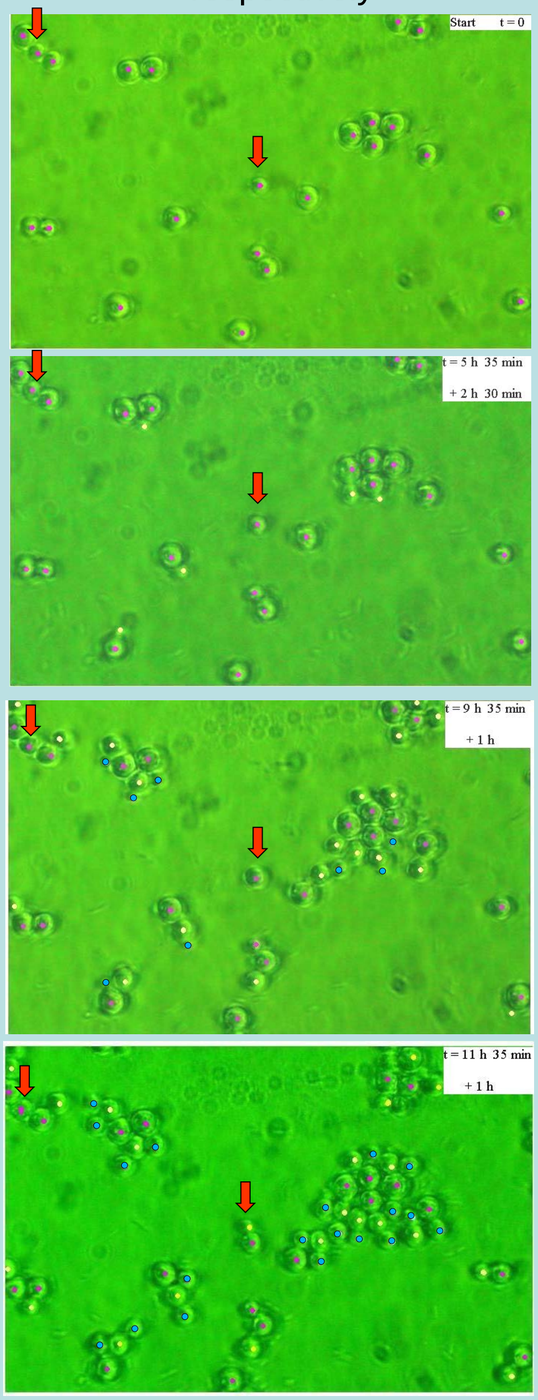
THE MAIN POINTS

1. Theoretically, the meaning of microbial viability is uncertain, there is no unified concept on this subject and the debates on it continue.
2. For practical reasons, the microbial viability is consider as an ability of the cells to multiply and to give progeny.
3. Quantitative assessment of this ability is not strait forward, and there is no any universal method that could be used in all cases.
4. Various direct and indirect methods for the multiplication ability assessment have been developed.
5. The direct methods enable to assess viability by cultivating microorganisms at appropriate conditions. So, viability is considered as culturability.
6. The indirect methods are based on the evaluation of intactness of cytoplasmic membrane, DNA replication and transcription, RNA translation and generation of energy.
7. The main challenges are associated with the lack of information on metabolic features and cryptobiotic (anabiotic, dormant, persistant) states of the vast majority of species of the divers microbial world.

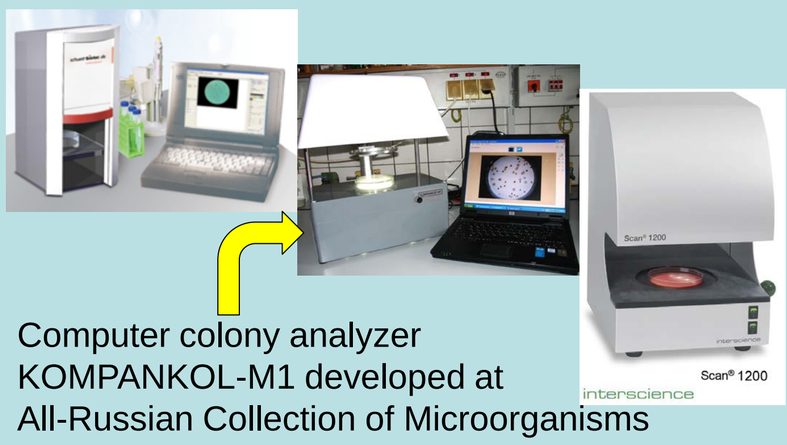
An example of “dormancy”

Multiplication of the *Cryptococcus terreus* cells on agarose with nutrients

The first, second and third generations, respectively

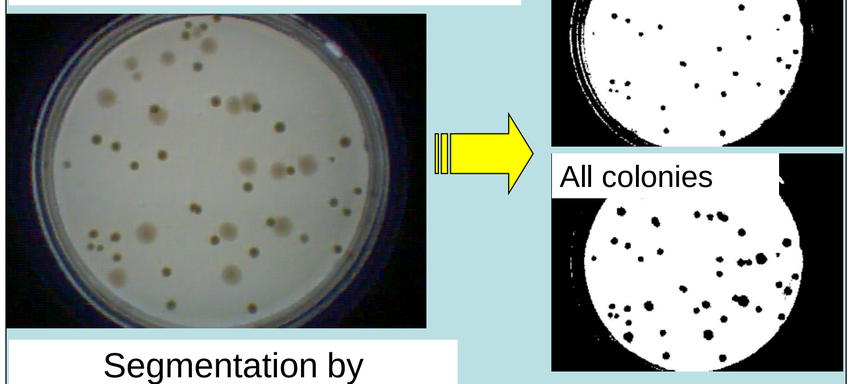


Automated colony counting

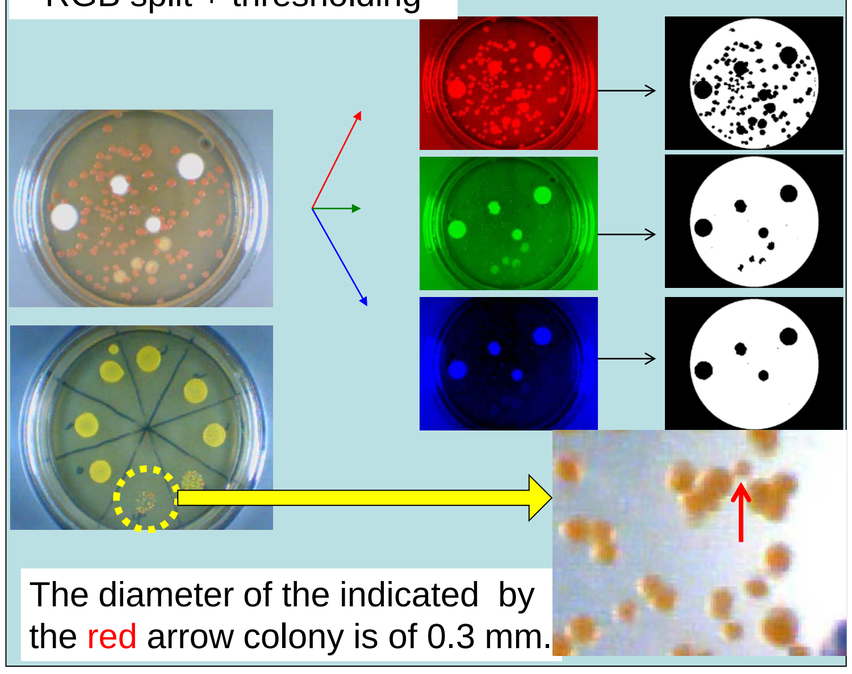


Computer colony analyzer KOMPANKOL-M1 developed at All-Russian Collection of Microorganisms

Segmentation by thresholding



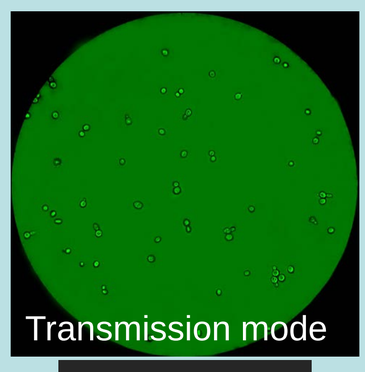
Segmentation by RGB split + thresholding

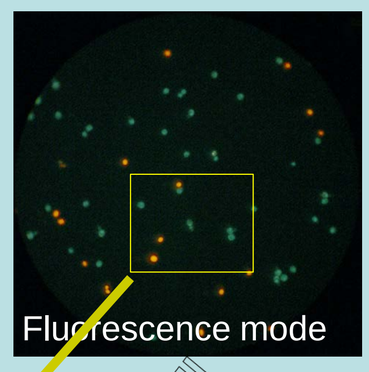


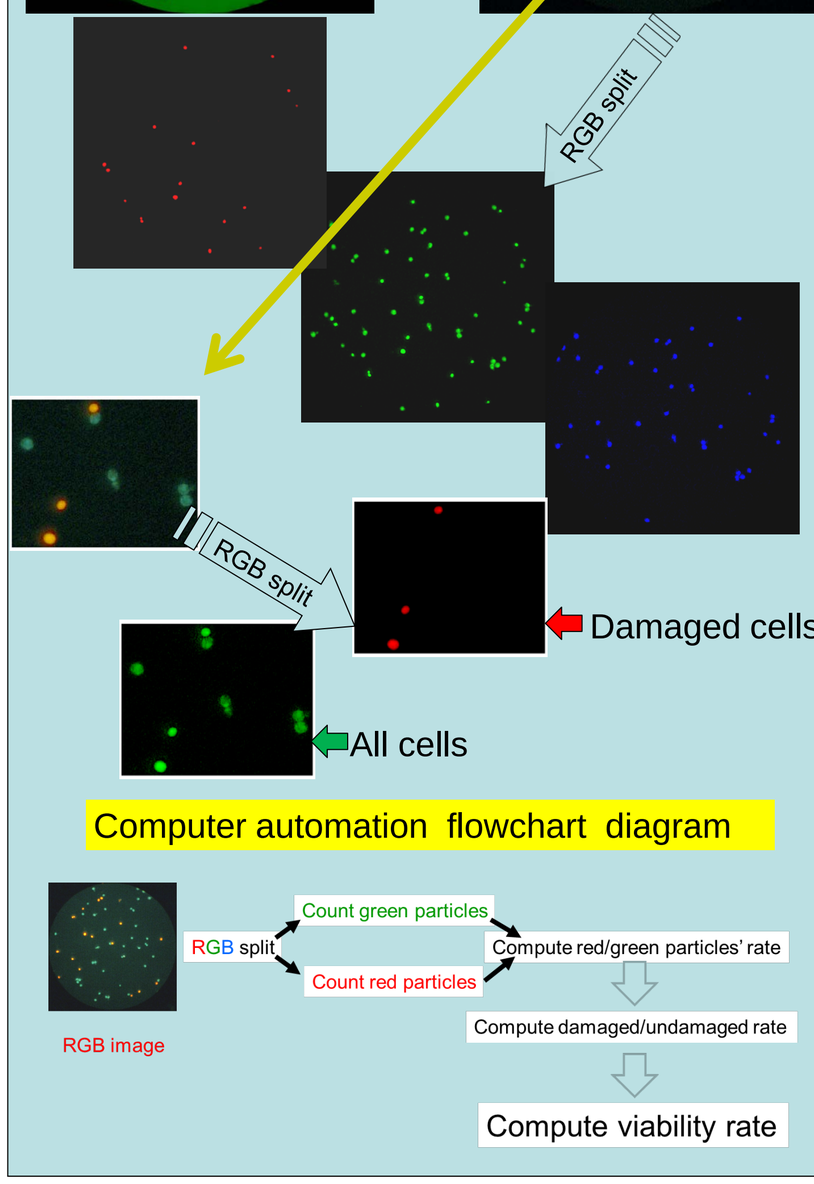
The diameter of the indicated by the red arrow colony is of 0.3 mm.

Rapid viability assessment

Fluorescence microscopy of the *Saccharomyces cerevisiae* cells stained by DAPI + ethidium bromide







Computer automation flowchart diagram

References

1. Postgate, J. R. (1969) Viable counts and viability. Methods Microbiol. 1, 611-628.
2. Lloyd, D. and Hayes, A. J. (1995) Vigour, vitality and viability of microorganisms. FEMS Microbiol. Lett. 133, 1-7.
3. Barer, M.R. and Harwood, C.R. (1999) Bacterial viability and culturability. Adv. Microb. Physiol. 41, 93-137.
4. Davey, H.M. (2011) Life, death, and in between: meanings and methods in microbiology. Appl. Environm. Microbiol.77, 5571-5576.
5. Puchkov, E.O. and Lasukov, Yu.I. (2008) Computer analyzer KOMPANKOL-M1 to monitor microorganism colonies. Water: chemistry and ecology. 5, 29-33. (In Russian)
6. Puchkov, E.O. (2014) Computer image analysis as a tool for microbial viability assessment: examples of use and prospects. Journal of Biosciences and Medicines. 2, No. 3, 1-6.
7. Puchkov, E. (2016) Image analysis in microbiology: a review. Journal of Computer and Communications. 4, 8-32.
8. Emerson, J.B. et al. (2017) Schrödinger's microbes: Tools for distinguishing the living from the dead in microbial ecosystems. Microbiome. 5, No. 1, 86.