



Astrobiotechnological perspective of lichens

A new hypothesis and its comparative assessment using the fuzzy logic model ASTRALIFE*

* $\underline{\textbf{ASTR}}$ obiotechnological $\underline{\textbf{A}}$ ssessment for $\underline{\textbf{LI}}$ fe by $\underline{\textbf{F}}$ uzzy logic $\underline{\textbf{E}}$ valuation

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This dissertation is dedicated to the memory of my late mother Celine (1 Corinthians 13:13).

The MSc. thesis of Louk Andrianos is approved by
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SHORT BIOGRAPHY

Louk Andrianos (Andriantiatsaholiniaina) was born in Madagascar in 1969. After his graduation in Agronomy (University of Antananarivo, 1993) and his Master's Degree in Hydrology (University Libre of Brussels, 1995), he received his PhD from the Technical University of Crete, specializing in Environmental Sciences and Sustainable Development, in 2001. He started his university career at Simon Fraser University in Vancouver, BC, Canada. Since 2006 he has worked at the Foundation for Research and Technology (FORTH) and at the Orthodox Academy of Crete (OAC) in Kolympari as scientific officer and Head of the Institute of Theology and Ecology, where he organizes seminars, conferences and lectures on ecological theology, fuzzy methodology, hydrology, economic anthropology, sustainable development, environmental ethics, and structural greed. He is the founder and editor of a series of environmental books on "Ecological Theology and Environmental Ethics (ECOTHEE) "Conservation and Sustainable Use of Wild Plant Diversity" (CSUWPD) and "Sustainable Alternatives for Poverty Reduction and Ecological Justice" (SAPREJ). He is an active member of various academic and environmental organizations in America, Asia, Africa and Europe. He is currently a member of Planta Europa Steering Committee, and collaborates with the World Council of Churches as sustainability consultant for the care for Creation and Climate justice. His research experience covers plant science, ecological theology, fuzzy logic, environmental management, administration, and sustainable development.

Αστροβιοτεχνολογική προοπτική των λειχήνων

Μια νέα υπόθεση και η συγκριτική αξιολόγησή της με τη χρήση του μοντέλου ασαφούς λογικής ASTRALIFE¹

ΕΚΤΕΤΑΜΕΝΗ ΠΕΡΙΛΗΨΗ

Κάθε οργανισμός, όπως και κάθε φυσικό σύστημα έχει όρια αντοχής σε ακραίες συνθήκες περιβάλλοντος. Η ανοιχτή συζήτηση περί κλιματικής αλλαγής και οι νέες τεχνολογίες στην κατάκτηση του διαστήματος έχουν οδηγήσει πολλά επιστημονικά πεδία στη μελέτη οργανισμών που αντέχουν σε ακραίες συνθήκες. Οι μελέτες αυτές συνήθως επικεντρώνονται στην αντοχή αυτών των οργανισμών σε πολύ υψηλές ή πολύ χαμηλές τιμές θερμοκρασίας, ατμοσφαιρικής πίεσης, ραδιενέργειας, υγρασίας, και οξυγόνου.

Εδώ και πολλά χρόνια είναι γνωστό πως οι λειχήνες συγκαταλέγονται στους πλέον ενδιαφέροντες οργανισμούς που αντέχουν σε ακραίες περιβαλλοντικές συνθήκες του πλανήτη μας ή πιθανόν και σε άλλους πλανήτες όπως ο Άρης. Πρόσφατα όμως, τεκμηριώθηκε πειραματικά ότι οι λειχήνες επιβιώνουν σε υπερβολικά ακραίες συνθήκες περιβάλλοντος, όπως για παράδειγμα στην απόλυτη ξηρασία και στην ακραία χαμηλή θερμοκρασία των -196 °C, διατηρώντας τον μεταβολισμό τους αναλλοίωτο, όταν επανέλθουν σε φυσιολογικές συνθήκες (Parasyri et al. 2018). Επίσης πρόσφατα ανακαλύφθηκε και τεκμηριώθηκε πειραματικά το ότι οι λειχήνες έχουν την ικανότητα σε ανοξικές συνθήκες να παράγουν μεγάλες ποσότητες υδρογόνου, στοιχείο ιδιαίτερα σημαντικό για την ενεργειακή αυτονομία (Papazi et al. 2015).

Οι δύο παραπάνω ανακαλύψεις, παρ' ότι έγιναν σε ελεγχόμενες πειραματικές συνθήκες στο γήινο περιβάλλον, ανοίγουν ένα νέο κεφάλαιο βιοτεχνολογικών εφαρμογών σε «αφιλόξενους» για την ώρα πλανήτες, στα πλαίσια μίας αστροβιοτεχνολογικής

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¹ *ASTRobiotechnological Assessment for LIfe by Fuzzy logic Evaluation

προσέγγισης. Έτσι, η παρούσα εργασία, αξιοποιώντας αυτά τα νέα πειραματικά ευρήματα και κινούμενη γύρω από το βασικό ερώτημα στην εποίκιση του διαστήματος — υπάρχουν οργανισμοί ή συστήματα οργανισμών, που θα επιβιώσουν σε άλλον πλανήτη όπως στον Άρη και πώς μπορεί κανείς να ελέγξει αυτή τη δυνατότητα; — χρησιμοποιεί την ασαφή λογική για να αξιολογήσει συγκριτικά αυτή τη νέα προοπτική που ανοίγεται για τους λειχήνες.

Η αντοχή ενός οργανισμού σε ακραίες συνθήκες και κατ' επέκταση η αστροβιοτεχνολογική προοπτική του είναι μία πολυδιάστατη και ασαφής έννοια για την οποία μέχρι στιγμής δεν υπάρχει κοινώς αποδεκτός ορισμός, με αποτέλεσμα να προσδιορίζεται με διαφορετικά κριτήρια για τα οποία δεν υπάρχει ενιαίο πλαίσιο αποδοχής αλλά ούτε και σύστημα μέτρησης.

Έτσι, στην παρούσα εργασία η ασαφής λογική χρησιμοποιείται ως εργαλείο σύνθεσης των διαφόρων κριτηρίων και παραμέτρων που ρυθμίζουν την αστροβιοτεχνολογική προοπτική και αντοχή των βιολογικών συστημάτων αλλά και ως εργαλείο συγκριτικής αξιολόγησης. Η χρήση μεθόδων τεχνητής νοημοσύνης και ασαφούς λογικής στην ποσοτικοποίηση της αστροβιοτεχνολογικής προοπτικής οργανισμών σε άλλο πλανήτη εφαρμόζεται, από όσο γνωρίζουμε, για πρώτη φορά.

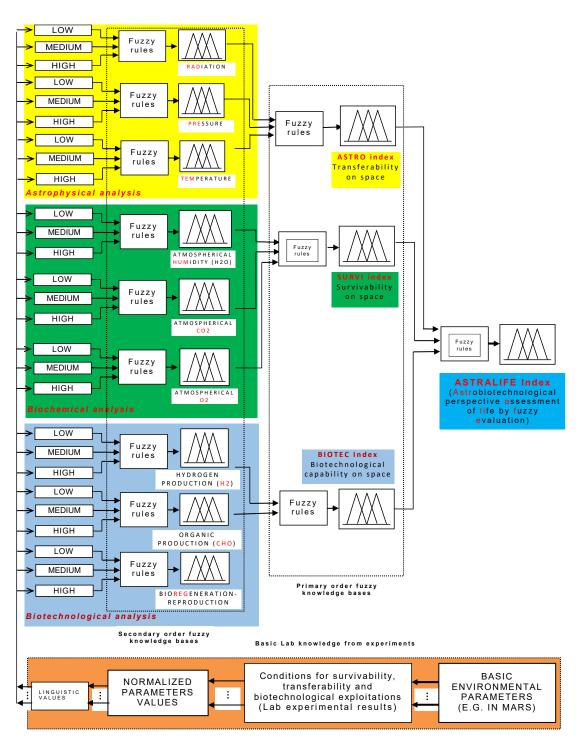
Συγκεκριμένα στην παρούσα διατριβή επιχειρείται η συγκριτική αξιολόγηση της αστροβιοτεχνολογικής προοπτικής των λειχήνων με τη χρήση του μοντέλου ασαφούς λογικής ASTRALIFE (*Astrobiotechnological Assessment of Life using Fuzzy logic Evaluation) που είναι προγραμματισμένο σε περιβάλλον MATLAB.

Συνοπτικά η λειτουργία του μοντέλου μπορεί να περιγραφεί ως εξής: Από εκτεταμένη βιβλιογραφική έρευνα συλλέγονται μετρήσεις, εκτιμήσεις και γενικώς πληροφορίες σχετικές με την εξέλιξη μεγεθών τα οποία συνδέονται με το περιβάλλον, όπως θερμοκρασία, διαθεσιμότητα νερού, πίεση, οξυγόνο, ραδιενέργεια, διοξείδιο του άνθρακα κ.α. Τα μεγέθη αυτά ονομάζονται δείκτες επιβίωσης και αποτελούν τα δεδομένα εισόδου του συστήματος ASTRALIFE. Οι διαθέσιμες πληροφορίες για τους δείκτες αυτούς αναλύονται, και με τη βοήθεια ασαφούς λογικής και κανόνων γνώσης συντίθενται σε επιμέρους συνιστώσες διαστημικής αεϊφορίας, όπως η επιβίωση, η μεταφορά, η βιοτεχνολογική ικανότητα κ.α. που χρησιμοποιούνται ως μέτρο στη συνολική εκτίμηση της αστροβιοτεχνολογικής προοπτικής του εκάστοτε οργανισμού. Το αποτέλεσμα της αξιολόγησης με τη χρήση του μοντέλου είναι ο βαθμός αστροβιοτεχνολογικής προοπτικής, των υπό εξέταση οργανισμών στο διάστημα τιμών [0, 1], μέσω του οποίου μπορεί να προκύψει η συγκριτική αξιολόγηση των οργανισμών για τις ακραίες συνθήκες που ελέγχονται. Το μοντέλο είναι ανοικτό και μπορεί να δέχεται νέους δείκτες επιβίωσης και γνώση υπό τη μορφή νέων κανόνων ανοχής/ανθεκτικότητας.

Με την παραπάνω μεθοδολογία εξετάστηκαν συγκριτικά πέντε διαφορετικοί οργανισμοί, συγκεκριμένα δύο είδη λειχήνων και τρία taxa μικροβίων και επελέγησαν οκτώ δείκτες επιβίωσης που θεωρήθηκε ότι έχουν την πλέον σημαντική (θετική ή αρνητική) συμβολή. Από τη συγκριτική αξιολόγηση προέκυψε η υπεροχή των λειχήνων ως προς την αστροβιοβιοτεχνολογική προοπτική, κάτι που ανοίγει τον δρόμο για τη βιοτεχνολογική χρήση τους σε ειδικές αποστολές σε πλανήτες υψηλού ενδιαφέροντος ως προς τη ζωή που μπορεί να συντηρήσουν, όπως είναι ο πλανήτης Άρης.

Η μεθοδολογία που ακολουθήθηκε αποτελεί μια πρωτοποριακή προσέγγιση, που αναμένεται να επανεκκινήσει τη συζήτηση που αφορά τη θεωρία της επιβίωσης οργανισμών/συστημάτων οργανισμών στο διάστημα με παράλληλη χρήση τους σε βιοτεχνολογικές εφαρμογές και να αποτελέσει ένα πρακτικό εργαλείο που θα συμβάλλει στη λήψη σχετικών αποφάσεων.

Graphical Abstract



The fuzzy logic model ASTRALIFE (<u>ASTR</u>obiotechnological <u>A</u>ssessment for <u>Li</u>fe by <u>F</u>uzzy logic Evaluation)

Abstract.

Every life form has range of tolerance to extreme environmental conditions. The open debate on climate change and the new technologies have led to many scientific fields in the study of organisms that are tolerant to extreme conditions. These studies usually focus on the tolerance of these organisms to extreme high or extreme low values of temperature, atmospheric pressure, radioactivity, humidity, and oxygen.

For many years it has been known that lichens are among the most interesting organisms that withstand extreme environmental conditions on Earth or possibly other planets such as Mars. Recently, however, it has been experimentally proven that lichens survive in extremely harsh environmental conditions, such as complete dehydration and extremely low temperature (-196°C/77°K) and most importantly that they retain their metabolism unchanged when they return to normal conditions (bio-regeneration) (Parasyri et al. 2018). Recently, experimental studies reveal that lichens have the ability to produce large amounts of hydrogen in anoxic conditions (Papazi et al. 2015), which is particularly important for energy autonomy. The two above discoveries, for first time demonstrated that the unprecedented poly-extremophile characteristic of lichens could be linked to biotechnological applications, following exposure to these extreme conditions maintained unchanged their ability to produce high yield of hydrogen. That opens the way for astrobiotechnological applications for these organisms. Thus, this work, exploiting these new experimental findings and moving around the basic question of space mission — are there organisms or systems of organisms that will survive on another planet as on Mars and how one can control this possibility? — uses the fuzzy logic to evaluate comparatively this new hypothesis for lichens.

The astrobiotechnological perspective and the extremophile behaviour of an organism is a multidimensional and vague notion that has so far no commonly accepted definition, with

the result that it is defined by different criteria for which there is neither a single acceptance framework nor a measurement system.

In this work, fuzzy logic is used as a tool for synthesizing the various criteria and parameters that regulate the astrobiotechnological perspective and tolerance of biological systems in extraterrestrial environments, but also as a tool of comparative assessment. The use of artificial intelligence and fuzzy logic methods in the quantification of the astrobiotechnological perspective on another planet is being applied for the first time.

Specifically, this thesis attempts to benchmark the astrobiotechnological perspective of lichens using the ASTRALIFE (*Astrobiotechnological Assessment of Life using Fuzzy logic Evaluation) fuzzy logic model, which is programmed in a MATLAB environment.

In summary, the operation of the model started from extensive bibliographic research, measurements, estimates and generally information relating to the variability of environmental-related quantities, such as temperature, water, pressure, oxygen, radioactivity, carbon dioxide, and others. These variables are called survival indicators and are the inputs data of the ASTRALIFE model. The available information on these indicators is analysed and, with the help of fuzzy logic and knowledge rules, transformed into partial composed components of space sustainability, such as survival, transport and biotechnology, which are used for the measurement of the overall astrobiotechnological perspective of an organism. The result of the evaluation using the model is the degree of the astrobiotechnological perspective of the organisms under consideration in the interval [0, 1], through which the comparative assessment of organisms for the extreme conditions under control can be obtained. The model is open and it can incorporate new survival indicators and knowledge rules in the form of new standards of tolerance.

Five different organisms that were formerly studied in astrobiological studies, namely two lichen species and three taxa microbes, were examined with the above methodology, and eight survival indices, considered to have the most significant (positive or negative) contribution, were selected. The comparative assessment has shown the lichens' superiority in the astrobiotechnological perspective, paving the way for their biotechnological use in special assignments on planets of high interest in life they can sustain, such as the planet Mars.

The methodology followed is a pioneering approach that is expected to relaunch the debate on the theory of organism/system survival on space while being used in biotechnological applications and to be a practical tool in decision making process.

1. Introduction

Astrobiology is the study of the origin, evolution, distribution, and future of life in the universe: extra-terrestrial life and life on earth (National Aeronautics and Space Administration — NASA²). It addresses the question of whether life exists beyond earth, and how humans can detect it if it does. Astrobiology makes use of physics, chemistry, astronomy, biology, molecular biology, ecology, planetary science, geography, and geology to investigate the possibility of life on other worlds and help recognize biospheres that might be different from that on earth.

On the other side, Biotechnology is the use of living systems and organisms to develop or make products, or "any technological application that uses biological systems, living organisms, or derivatives thereof, to make or modify products or processes for specific use" (UN convention on biological diversity, art.³). In the late 20th and early 21st centuries, biotechnology has expanded to include new and diverse sciences such as genomics, recombinant gene techniques, applied immunology, pharmaceutics, diagnostic tests and renewable energy resources.

Astrobiotechnology is the natural interface between astrobiology, biotechnology, space and geological sciences (Steele and Toporski, 2002), while the Astrobiotechnological perspective of a species is related to the possibility of astrobiological and biotechnological uses of the species for the production of valuable compounds/products in order to facilitate life establishment or human setting in extra-terrestrial conditions, such as on Mars conditions.

Survivability of living organisms in other planets is nowadays a leading discussion among scientists as a solution to earth sustainability problems and climate change adaptations. Many conversations concerning what humans will do when Earth becomes less habitable,

² https://astrobiology.nasa.gov/about/

³ https://www.cbd.int/convention/articles/default.shtml?a=cbd-02

whether due to climate change or a series of manmade disasters, led to the conclusion about leaving Earth for another planet. Mars, a planet of similar size to Earth and at the outreach of humans, is often brought up as an option. Even if the idea of abandoning the earth is not plausible, the technology acquired from astrobiotechnological research would be helpful to cope with possible harsh environmental challenges on earth in the future.

Astrobiotechnological perspective and survivability are inherently vague and complex concepts. As pointed out in the literature, it is not that survivability parameters are lacking but their fragmentary and polymorphous nature hampers their direct usefulness in the quest of strategies for survival (Cockell, 2016). Despite the fact that the concept of survivability and astrobiotechnological perspective are still vague, the development of astrobiotechnological and survivability policies is a necessity if we adopt the precautionary approach for the future of our planet.

What we need is adequate information that is tailored to quantitative astrobiotechnological objectives. Such information should: (a) give a clear indication as to whether objectives of survivability are met, (b) concern the system as a whole, (c) have a quantitative character, (d) be understandable to non-scientists, and (e) contain parameters which can be used for periods of one or more decades. The need for a practical tool to assess astrobiotechnological perspective is crucial to scientists and policy-makers if they are to secure future development of human kind on space or under extreme environmental conditions. Since such a tool is not available, management by trial-and-error instead of management by acknowledge and prediction is currently the way to establish sustainable policies.

In this work, fuzzy logic is proposed as a systematic tool for the assessment of astrobiotechnological capability of organisms. Fuzzy logic based technology is one of the fastest growing areas in engineering (Bastian, 1995). The application of fuzzy logic is widespread in the realm of automation, optimization and simulation techniques. Since

1965, when the foundation of fuzzy logic theory was first announced and, especially, after the discovery of the first fuzzy chips in 1987, the literature on both theory and applications of fuzzy logic has been growing. In this subsection, we report a synopsis of the foundations of fuzzy systems (Berkan and Trubatch, 1997).

Almost all statements and propositions found in practical life are within some proximity of the absolute truth. However, in practice, most uncertainties are tolerable, manageable, or negligible. We do not cancel our flight although the risk of a crash is not zero. We live in a world of partial truth. So do the systems we build and operate.

Science has evolved around the idealism of mathematics, which sometimes falls short of dealing with the reality of life. Realism, referring to perception of partial truth, cannot be accurately expressed by true/false duality for obvious reasons. The mathematical approach has been improved recently to accommodate partial truth by the introduction of fuzzy set theory invented by Professor Lotfi A. Zadeh. Unlike classical set theory, fuzzy set theory is flexible since sets are composed of elements, each with different degree of membership. Similarly, in fuzzy logic, each proposition is assigned a truth-value ranging between 0 (false) and 1 (true).

The theory of fuzzy sets provides a more realistic mathematical representation of the perception of truth than traditional, two-valued logic and Boolean algebra. In the transition from crisp sets to fuzzy sets, the key element is possibility theory and its extended interpretations. In a collective manner, possibilities are defined by a distribution function, often called the *membership function*. Membership functions give the truth-values of expressions like "Hercules is muscular" or more complex expressions articulated in daily language. As a result there is a way to compute with words using fuzzy set theory and possibility theory.

One way to understand the relationship between fuzzy sets and fuzzy logic is to examine natural language. Expressions in natural language such as "Hercules is muscular" or "Hercules can lift a heavy weight" are phrases describing an event or state of being. When they are put together in a sense-making order, a context is created that leads to reasoning. For example, the combination of "Hercules is muscular" and "Hercules can lift a heavy weight" creates a context in which muscular volume and strength become related. Such statements are called unconditional statements. One step further is the combination of simple expressions using some linguistic connectors (also called operators) such as "If Hercules is muscular then he can lift a heavy weight". Such propositions are called rules where the connectives "if-then" modify the context and make a conditional statement. When such conditions are imposed, reasoning gets more restricted than in a simple relationship, which leads to the subject of logical inference.

Classical logic, also referred to as Boolean logic, consists of three elements: truth-values, linguistic connectors, and reasoning types. In Boolean logic, truth-values are either 1 or 0, which correspond to true/false duality. In fuzzy logic, truth is a matter of degree, thus truth-values range between 1 and 0 in a continuous manner. This concept of continuum constitutes the most outstanding difference between classical logic and fuzzy logic. Linguistic connectors (or operators) in fuzzy logic have the same function as in Boolean logic (union, intersection, negation).

Fuzzy logic is capable of representing uncertain data, emulating skilled humans, and handling vague situations where traditional mathematics is ineffective.

The following three basic features justify the use of the fuzzy logic reasoning for our study:

 a. Fuzzy logic has the ability to deal with complex and polymorphous concepts, which are not amenable to a straightforward quantification and contain ambiguities. In addition, reasoning with such ambiguous concepts may not be clear and obvious, but rather fuzzy.

- b. Fuzzy logic provides the mathematical tools to handle ambiguous concepts and reasoning, and finally gives concrete answers ("crisp" as they are called) to problems fraught with subjectivity. Perspective is, indeed, quite subjective.
- c. Another important aspect of fuzzy logic is that it uses linguistic variables, thus performing computation with words. If a traditional mathematical approach towards perspective assessment were adopted, such as cost-benefit analysis or algebraic formulas, then certain factors, which are impossible to quantify, would be left out. There exist, however, aspects of astrobiotechnological perspective, which cannot be quantified and yet are very important as, for example, values and opinions. In this area of human thought fuzzy logic performs successfully (Zimmermann, 1991; Zadeh, 1994).

Fuzzy logic is a scientific tool that permits simulation of the dynamics of a system without a detailed mathematical description. Knowledge is represented by IF-THEN linguistic rules, which describe the logical evolution of the system according to the linguistic values of its principal characters that we call linguistic variables. Real values are transformed into linguistic values by an operation called fuzzification, and then fuzzy reasoning is applied in the form of IF-THEN rules. A final crisp value is obtained by defuzzification, which does the opposite to fuzzification. A simple example of IF-THEN fuzzy approximate reasoning is the assessment of human happiness based on the popular feeling about the importance of health. Choosing money and health as the principal factors of happiness, the fuzzy rules might be:

- IF one has "much" money AND "good" health, THEN he is "very" happy,
- IF one has "much" money AND "bad" health, THEN he is "insufficiently" happy, and

- IF one has "little" money AND "good" health, THEN he is "satisfactorily" happy.

"Much" and "little" are linguistic values of the linguistic variable money; they correspond to the fuzzification of a fixed amount of money. (Good, bad), and (very, satisfactorily, insufficiently) are, respectively, linguistic values of the state of health and happiness. Defuzzification of the linguistic values "very", "satisfactorily", and "insufficiently" provides a crisp measurement of happiness.

Accordingly, to assess astrobiotechnological perspective and survivability using fuzzy logic, the followings have to be defined:

- Linguistic variables, which best represent the survivability of the whole system,
- Linguistic rule bases and fuzzy logical operators which express qualitatively the knowledge and the key features of the overall system,
- The membership functions which determine quantitatively the corresponding value of the assessment, and
- A defuzzification method to convert fuzzy statements into a single crisp value of overall survivability.

Towards this direction, a computer-based information system has been configured, which uses common indicators of astrophysical, biochemical and biotechnological environmental integrity as inputs and employs fuzzy logic reasoning to provide astrobiotechnological perspective measures on cosmic or planet level. The new model is called ASTRALIFE (Astrobiotechnological perspective Assessment for Life using Fuzzy logic Evaluation).

The launched in 2007 space experiment with the European BIOPAN "" facility for a 10-day spaceflight on board a Russian "Foton" retrievable satellite included for the first time the

vagrant lichen species *Aspicilia fruticulosa* from Guadalajara steppic highlands (Central Spain), as well as other lichen species (Raggio et al, 2011).

Lichens are the symbiotic phenotype of nutritionally specialized fungi (the mycobiont) that acquire, in an ecologically obligate, mutualistic symbiosis, fixed carbon from a population of green algal and/or cyanobacterial cells (the photobiont) (Honegger, 1998; Grube and Hawkasworth, 2007). Most mycobionts belong to the Ascomycota, whereas only a few species of Basidiomycota form lichens. Concerning photobionts, about 85% of lichenforming fungi associate with green algae, about 10% with cyanobacteria, and about 4% simultaneously with both.

Lichens have proven to be exceptionally suitable organisms for experiments in astrobiology (Raggio et al. 2011). During space flight, the lichen samples were exposed to selected space conditions, that is, the space vacuum, cosmic radiation and different spectral ranges of solar radiation. After retrieval, the algal and fungal metabolic integrity of the samples were evaluated in terms of chlorophyll a fluorescence, ultrastructure, and CO₂ exchange rates. Whereas the space vacuum and cosmic radiation did not impair the metabolic activity of the lichens, solar electromagnetic radiation, especially in the wavelength range between 100 and 200 nm, caused reduced chlorophyll a yield fluorescence; however, there was a complete recovery after 72 h of reactivation (Raggio et al. 2011).

Nevertheless, new findings, such as the ability of lichens to produce hydrogen under anoxic conditions activating appropriate bioenergetics pathways (Papazi et al. 2015), as well as their ability to survive in extreme environments (extremophile behavior), similar to those encountered in extraterrestrial environments (Parasyri et al. 2018), pave the way for future astrobiotechnological applications. Of course, it is widely accepted that a reliable measure of astrobiotechnological capacity should be the result of integrating astrobiological survivability

and biotechnological accounts. However, this is not readily achievable due to lack of data and yet unsolved methodological problems.

Thus, taking into consideration all the above, a comparative assessment of astrobiotechnological perspective of lichens has been attempted using the fuzzy model ASTRALIFE, which configured for the needs of the current study, where two lichens species and three other microorganisms were included among the selected organisms, which have been tested and analyzed for Martian conditions.

2. Configuration of the ASTRALIFE model

For the needs of the current study the configuration of the ASTRALIFE model has been achieved and the followed procedure is described below.

2.1 Linguistic variables for the ASTRALIFE model

Briefly, a linguistic variable is defined by four items: (a) the name of the variable (e.g. money), (b) its linguistic values (e.g. "much" and "little"), (c) the membership functions of the linguistic values, and (d) the physical domain over which the variable takes its quantitative values. The membership function of a linguistic value gives the degree to which any quantitative value belongs to the linguistic value. For example, the membership functions of "much" and "little" could be exponential functions of the amount of income per month in dollars, and the range of income is the physical domain of the variable.

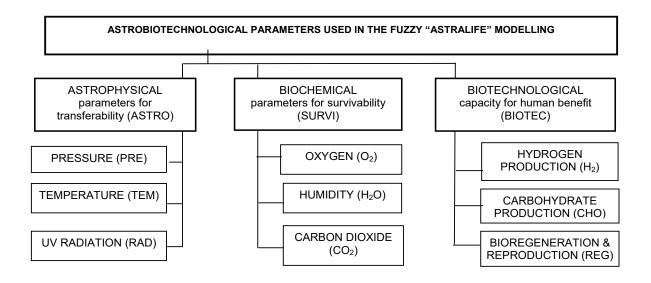
In the ASTRALIFE model for the assessment of astrobiotechnological perspective on Mars, we adopted the following linguistic variables:

a) Nine linguistic variables: PRESSURE (PRE), TEMPERATURE (TEM), UV RADIATION (RAD), HUMIDITY (H₂O), OXYGEN (O₂), CARBON DIOXIDE (CO₂), HYDROGEN

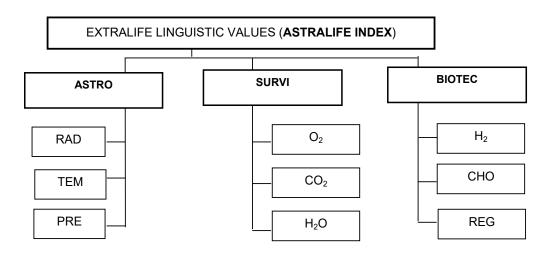
- production (H_2) , BIOREGENRATION or reproduction (REG) and CARBOHYDRATE/OXYGEN production (CHO),
- b) Three primary linguistic variables: ASTROPHYSICAL parameters for transferability (ASTRO), BIOCHEMICAL parameters for survivability (SURVI), and the BIOTECHNOLGICAL capacity for human benefit (BIOTEC),
- c) One final output linguistic variable: ASTRALIFE index, which stands for the astrobiotechnological perspective for life index.

The following Figures show the relations between the twelve linguistic variables used in the ASTRALIFE model (Fig 1 a and b), while the overall configuration of the ASTRALIFE model is presented in Figure 2.

The ASTRALIFE model may be viewed as a tree-like network of knowledge bases. The inputs of each knowledge base are basic indicators provided by the user or composite indicators collected from other knowledge bases. By using fuzzy logic and IF-THEN rules, these inputs are combined to yield a composite indicator as output, which is then passed on to subsequent knowledge bases.



a.



b.

Figure 1. a. Parameters for the astrobiological perspective assessment with fuzzy logic **b.** Linguistic variables for the evaluation of astrobiotechnological perspective with fuzzy logic

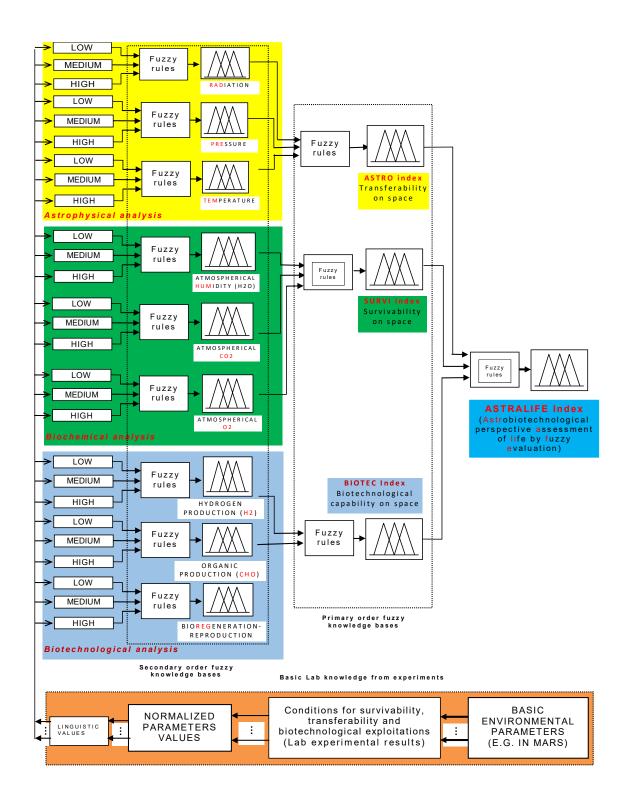


Figure 2. Configuration of the ASTRALIFE model

The model is flexible in the sense that users can choose the set of indicators and adjust the rules of any knowledge base according to their needs and the characteristics of the environmental system to be assessed.

The model we develop herein serves a dual purpose: it provides a flexible framework defining astrobiotechnological perspective as a function of a number of variables (any but given) and at the same time it gives the mathematical machinery to compute numerical values of the concept. The ASTRALIFE model provides also linguistic values of the overall astrobiotechnological perspective, as well as its components.

- The single final output (ASTRALIFE index) linguistic variable takes the linguistic values
 "poor" (P), "acceptable" (A), and "excellent" (E).
- For the three primary (ASTRO, SURVI, BIOTEC) linguistic variables, we use the linguistic values *Very low (VL), low (L), intermediate (I), high (H), and very high (VH).*
- For the nine basic secondary variables (TEM, RAD, PRE, H₂O, O₂, CO₂, H₂, CHO and REG), the linguistic values "weak" (W), "medium" (M) and "strong" (S).

In order to obtain a common scale and allow aggregation the data for each parameter indicator have to be normalized before fuzzy computations. Measurements or data of each basic indicator are normalized on a scale between zero and one to allow aggregation and to facilitate fuzzy computations. This is done as follows. To each basic parameter indicator c we assign a target, a minimum \underline{c} (min) and a maximum value \overline{c} (max). The target can be a single value or, in general, any interval on the real line of the form $[t_c, T_c]$ representing a range of desirable values for the indicator. The maximum and minimum values are taken over the set of available measurements of the indicator from various experimentations. Let x_c be the indicator value for the variable we want to assess. The normalized value y_c is calculated as follows

$$y_c(x_c) = \begin{cases} \frac{x_c - \underline{c}}{t_c - \underline{c}} & \epsilon \le x_c < t_c \\ 1 & t_c \le x_c \le T_c \\ \frac{\overline{c} - x_c}{\overline{c} - T_c} & T_c < x_c \le \epsilon \end{cases}$$
(1.1)

The normalization of indicator values is illustrated in Figure 3, while different curves of normalization can be used according to needs and context.

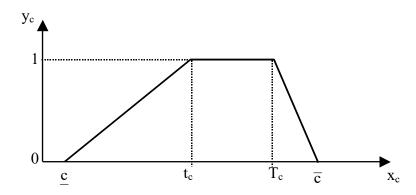


Figure 3. Normalized value of variable x_c .

Triangular functions are used for the basic and primary variables, while trapezoidal functions are chosen for the secondary variables to represent an increased uncertainty involved in the computation (see Driankov et al., 1996; Zimmermann, 1991) for a rather detailed exposition on membership functions). The horizontal axis of each membership function expresses the normalized values of each astrobiotechnological capability variable and ranges over [0, 1], whereas the vertical axis expresses membership grades ranging again over [0, 1]. Triangular membership functions are selected because they are simple and agree with widely held opinions. Most importantly, triangular membership functions can approximate most non-triangular ones (Pedrycz, 1994). Trapezoidal membership functions are straightforward extensions of triangular ones.

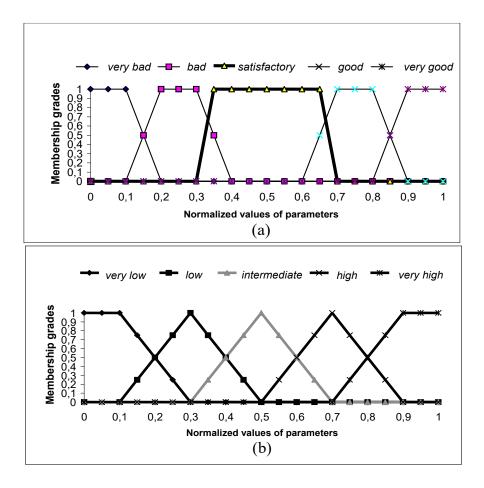


Figure 4. Graphical presentation of membership functions of the linguistic values

The fuzzification module transforms the crisp, normalized value y_c of indicator c into a linguistic variable in order to make it compatible with the rule base. Loosely speaking, a linguistic variable is a variable whose values are words or phrases. In the model, the linguistic values of each variable are recapitulated in Table 1.

A linguistic value LV is represented by a fuzzy set using a membership function $\mu_{LV}(y)$. The membership function associates with each normalized indicator value y_c a number $\mu_{LV}(y_c)$ in [0, 1] which represents the grade of membership of y_c in LV or, equivalently, the truth-value of proposition "indicator c is LV". The ASTRALIFE model uses trapezoidal and triangular membership functions.

Table 1. Linguistic values used in the ASTRALIFE model

Variables	Linguistic values
1 (one) Final ASTRALIFE index	(P) poor, (A) acceptable, (E) excellent
3 (three) Primary components (ASTRO, SURVI, BIOTEC)	(VL) very low, (L, low, (I) intermediate, (H) high and (VH) very high
9 (nine) Secondary variables (TEM, RAD, PRE, H ₂ O, O ₂ , CO ₂ , H ₂ , CHO, REG)	(W) weak, (M) medium, (S) strong

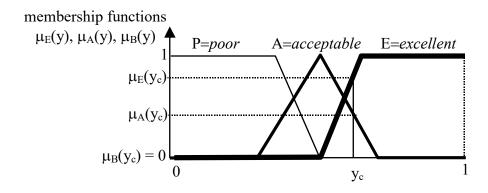


Figure 5. Examples of linguistic values of indicator c

An example of a linguistic variable corresponding to indicator c, which takes on three linguistic values, P (poor), A (acceptable), and E (excellent) is presented in Figure 5. The horizontal axis of each membership function expresses the normalized values of the indicator.

2.2 Linguistic inference rules and fuzzy operators

Knowledge concerning the astrobiotechnological perspective of any organism is represented by fuzzy rules whose general form is

"IF (PREMISE) THEN (CONCLUSION)"

The rules are expressions of the role of interdependencies among factors or variables of the assessment. They are the result of multidisciplinary studies about astrobiotechnological capacity of organisms. We consider a typical knowledge base that computes indicators from a number of input indicators, say, 1, 2, ..., c, ... Suppose that s is represented by the linguistic values LV_{α_i} , LV_{β_i} , ..., LV_{ν_i} , ... with membership functions μ_{α_i} , μ_{β_i} , ..., μ_{ν_i} , ..., μ_{ν_i} , ..., μ_{ν_i} , ... with membership functions μ_1 , μ_2 , ..., μ_k , Finally, for each input indicator c the following are available:

 y_c normalized value of c (computed from the data or by some other inference engine), $c=1,\,2,\,...$

 $\mu_k(y_c)$ grade of membership of y_c in each linguistic value LV_k, where k=1,2,... and c=1,2,...

A rule r of the rule base has the form

"indicator 1 is LV_i " AND "indicator 2 is LV_j "...AND "indicator c is LV_k "...THEN

"indicator s is LV_v "

To determine the overall ASTRALIFE index, the rule base needs 5^3 = 125 rules because we have five linguistic values and three variables, ASTRO, SURVI and BIOTEC. For each of these variables, the rule base has 3^3 = 27 rules because we have three basic variables for their

determination. The linguistic rule base for the determination of the overall biotechnological capability and the secondary variables are summarized in Tables 2, 3, 4 and 5.

The rule base was built in such a way that the minimal value of one variable dominates all others. Thus, in each rule, the final output value (consequent part) depends essentially on the minimal value of the inputs (premise or antecedent part).

 Table 2. PRIMARY order rule base for the computation of ASTRALIFE INDEX

Rule	if	and	and	then
Nr.	ASTRO is	SURVI is	BIOTEC is	ASTRALIFE is
1	very low	very low	very low	weak
2	very low	low	very low	weak
3	very low	intermediate	very low	weak
4	very low	high	very low	weak
5	very low	very high	very low	weak
6	very low	very low	low	weak
7	very low	low	low	weak
8	very low	intermediate	low	weak
9	very low	high	low	weak
10	very low	very high	low	weak
11	very low	very low	intermediate	weak
12	very low	low	intermediate	weak
13	very low	intermediate	intermediate	weak
14	very low	high	intermediate	weak
15	very low	very high	intermediate	weak
16	very low	very low	high	weak
17	very low	low	high	weak
18	very low	intermediate	high	weak
19	very low	high	high	weak
20	very low	very high	high	weak
21	very low	very low	very high	weak

22	very low	low	very high	weak
23	very low	intermediate	very high	weak
24	very low	high	very high	weak
25	very low	very high	very high	weak
26	low	very low	very low	weak
27	low	low	very low	weak
28	low	intermediate	very low	weak
29	low	high	very low	weak
30	low	very high	very low	weak
31	low	very low	low	weak
32	low	low	low	weak
33	low	intermediate	low	weak
34	low	high	low	weak
35	low	very high	low	weak
36	low	very low	intermediate	weak
37	low	low	intermediate	weak
38	low	intermediate	intermediate	weak
39	low	high	intermediate	weak
40	low	very high	intermediate	weak
41	low	very low	high	weak
42	low	low	high	weak
43	low	intermediate	high	weak
44	low	high	high	weak
45	low	very high	high	weak
46	low	very low	very high	weak
47	low	low	very high	weak
48	low	intermediate	very high	weak
49	low	high	very high	weak
50	low	very high	very high	weak
51	intermediate	very low	very low	weak
52	intermediate	low	very low	weak

53	intermediate	intermediate	very low	weak
54	intermediate	high	very low	weak
55	intermediate	very high	very low	weak
56	intermediate	very low	low	weak
57	intermediate	low	low	weak
58	intermediate	intermediate	low	weak
59	intermediate	high	low	weak
60	intermediate	very high	low	weak
61	intermediate	very low	intermediate	weak
62	intermediate	low	intermediate	weak
63	intermediate	intermediate	intermediate	medium
64	intermediate	high	intermediate	medium
65	intermediate	very high	intermediate	medium
66	intermediate	very low	high	weak
67	intermediate	low	high	weak
68	intermediate	intermediate	high	medium
69	intermediate	high	high	medium
70	intermediate	very high	high	medium
71	intermediate	very low	very high	weak
72	intermediate	low	very high	weak
73	intermediate	intermediate	very high	medium
74	intermediate	high	very high	medium
75	intermediate	very high	very high	medium
76	high	very low	very low	weak
77	high	low	very low	weak
78	high	intermediate	very low	weak
79	high	high	very low	weak
80	high	very high	very low	weak
81	high	very low	low	weak
82	high	low	low	weak
83	high	intermediate	low	weak

84	high	high	low	weak
85	high	very high	low	weak
86	high	very low	intermediate	weak
87	high	low	intermediate	weak
88	high	intermediate	intermediate	medium
89	high	high	intermediate	medium
90	high	very high	intermediate	medium
91	high	very low	high	weak
92	high	low	high	weak
93	high	intermediate	high	medium
94	high	high	high	strong
95	high	very high	high	strong
96	high	very low	very high	weak
97	high	low	very high	weak
98	high	intermediate	very high	medium
99	high	high	very high	strong
100	very high	very high	very high	strong
101	very high	very low	very low	weak
102	very high	low	very low	weak
103	very high	intermediate	very low	weak
104	very high	high	very low	weak
105	very high	very high	very low	weak
106	very high	very low	low	weak
107	very high	low	low	weak
108	very high	intermediate	low	weak
109	very high	high	low	weak
110	very high	very high	low	weak
111	very high	very low	intermediate	weak.
112	very high	low	intermediate	weak
113	very high	intermediate	intermediate	medium
114	very high	high	intermediate	medium

115	very high	very high	intermediate	medium
116	very high	very low	high	weak
117	very high	low	high	weak
118	very high	intermediate	high	medium
119	very high	high	high	strong
120	very high	very high	high	strong
121	very high	very low	very high	weak
122	very high	low	very high	weak
123	very high	intermediate	very high	medium
124	very high	high	very high	strong
125	very high	very high	very high	strong

ASTRO= Transferability under astrological conditions; SURVI= Survivability under biochemical space conditions; BIOTEC= Biotechnological conditions ASTRALIFE= Astrobiotechnological perspective assessment index;

Table 3. Second order rule base for the computation of ASTRO

Rule	if	and	and	then
Nr.	PRE is	RAD is	TEM is	ASTRO is
1	weak	strong	strong	very high
2	weak	medium	strong	very high
3	weak	weak	strong	high
4	weak	strong	medium	very high
5	weak	medium	medium	high
6	weak	weak	medium	intermediate
7	weak	strong	weak	high
8	weak	medium	weak	intermediate
9	weak	weak	weak	very low
10	medium	strong	strong	very high
11	medium	medium	strong	high
12	medium	weak	strong	intermediate

13	medium	strong	medium	high
14	medium	medium	medium	intermediate
15	medium	weak	medium	low
16	medium	strong	weak	intermediate
17	medium	medium	weak	low
18	medium	weak	weak	very low
19	strong	strong	strong	high
20	strong	medium	strong	intermediate
21	strong	weak	strong	low
22	strong	strong	medium	intermediate
23	strong	medium	medium	low
24	strong	weak	medium	very low
25	strong	strong	weak	low
26	strong	medium	weak	very low
27	strong	weak	weak	very low

Table 4. Second order rule base for the computation of SURVI

Rule	if	and	and	then
Nr.	O ₂ is	CO ₂ is	H₂O is	SURVI is
1	weak	strong	strong	very high
2	weak	medium	strong	very high
3	weak	weak	strong	high
4	weak	strong	medium	very high
5	weak	medium	medium	high
6	weak	weak	medium	intermediate
7	weak	strong	weak	high
8	weak	medium	weak	intermediate
9	weak	weak	weak	very low
10	medium	strong	strong	very high
11	medium	medium	strong	high

12	medium	weak	strong	intermediate
13	medium	strong	medium	high
14	medium	medium	medium	intermediate
15	medium	weak	medium	low
16	medium	strong	weak	intermediate
17	medium	medium	weak	low
18	medium	weak	weak	very low
19	strong	strong	strong	high
20	strong	medium	strong	intermediate
21	strong	weak	strong	low
22	strong	strong	medium	intermediate
23	strong	medium	medium	low
24	strong	weak	medium	very low
25	strong	strong	weak	low
26	strong	medium	weak	very low
27	strong	weak	weak	very low

Table 5. Second order rule base for the computation of BIOTEC

Rule	if	and	and	then
Nr.	H ₂ is	CHO is	REG is	BIOTEC is
1	weak	strong	strong	very high
2	weak	medium	strong	very high
3	weak	weak	strong	high
4	weak	strong	medium	very high
5	weak	medium	medium	high
6	weak	weak	medium	intermediate
7	weak	strong	weak	high
8	weak	medium	weak	intermediate
9	weak	weak	weak	very low
10	medium	strong	strong	very high

11	medium	medium	strong	high
12	medium	weak	strong	intermediate
13	medium	strong	medium	high
14	medium	medium	medium	intermediate
15	medium	weak	medium	low
16	medium	strong	weak	intermediate
17	medium	medium	weak	low
18	medium	weak	weak	very low
19	strong	strong	strong	high
20	strong	medium	strong	intermediate
21	strong	weak	strong	low
22	strong	strong	medium	intermediate
23	strong	medium	medium	low
24	strong	weak	medium	very low
25	strong	strong	weak	low
26	strong	medium	weak	very low
27	strong	weak	weak	very low

There are many ways to quantitatively express fuzzy rules by choosing a specific mathematical representation of the AND, OR, and IF-THEN connectives (Tsourveloudis and Phillis, 1998). In our model, the connective AND is given by the min-operator.

Consider, for example, the linguistic fuzzy rule of Table 2 with number 43, the premise is as followed:

"IF ASTRO is "Low" and SURVI is "Intermediate" and BIOTEC is "High"

THEN ASTRALIFE is "Weak"

If the values of ASTRO, SURVI and BIOTEC are y_{ASTRO} , y_{SURVI} , and y_{BIOTEC} respectively, then the degree to which the above rule is applicable is given by

 $\mu_{PREMISE} = \min \{ \mu_L(y_{ASTRO}), \mu_I(y_{SURVI}), \mu_H(y_{BIOTEC}) \}$

where μ_L , μ_I and μ_H are the membership functions of the linguistic values "Low" (L), "Intermediate" (I) and "High" (H), respectively.

It is easy to understand that the final resistance of the organism is depending on the limiting factor in the existing environment.

The min operator is a natural choice for the logical AND. Bellman and Giertz (1973) have devised a set of axioms which the membership functions of the AND operator should satisfy and then prove that the min operator satisfies them. Intuitively, the AND operator corresponds to the intersection of sets, which is the largest common set (Zimmermann, 1991). The choice of fuzzy logic operators can be deduced from the structure of the linguistic rule base but the use of real data could also help in validating, modifying and improving the mathematical interpretations of the fuzzy operators or the linguistic rule base itself.

In general, a rule base may contain several rules assigning subsets of the same linguistic value LV_{ν} of the conclusion indicator s. For example, the rule base of the secondary component ASTRO contains the following five rules which gives the final output of "Low" as premises:

Rule nr. 15:

IF PRE is Medium AND RAD is Weak AND TEM is Medium THEN ASTRO is Low

Rule nr. 17:

IF PRE is Medium AND RAD is Medium AND TEM is Weak THEN ASTRO is Low

Rule nr. 21:

PRE is Strong AND RAD is Weak AND TEM is Strong THEN ASTRO is Low

Rule nr. 23:

IF PRE is Strong AND RAD is Medium AND TEM is Medium THEN ASTRO is Low

Rule nr. 25:

IF PRE is Strong AND RAD is strong AND TEM is Weak THEN ASTRO is Low

To combine the results of these rules into a single truth-value, we use the union of the individual-rule meanings via the max-operator. In general, if $R_{s,v}$ is the collection of all rules assigning the linguistic value LV_v to indicator s, the truth value of the conclusion "indicator s is LV_v " is expressed by

$$\max_{\mathsf{f}_{\mathsf{s},\mathsf{v}}} \mu_{\mathsf{PREMISF}}$$

$$\mathsf{f}_{\mathsf{s},\mathsf{v}} = {}^{\mathsf{r} \in \mathsf{R}_{\mathsf{S}},\mathsf{v}}$$

$$\tag{1.1}$$

Finally, the inference engine produces a single fuzzy subset $LV_{s,v}$ for each linguistic value LV_v . The membership function of $LV_{s,v}$ assigns a degree of fulfillment $\mu_{s,v}(z)$ of any numerical value $z \in [0, 1]$ of indicator s to the linguistic value and it is computed from

$$\mu_{s,v}(z) = \min\{\mu_v(z), f_{s,v}\}$$
 (1.2)

The maximum value of $\mu_{s,v}(z)$ is $f_{s,v}$ and it is called the height of the fuzzy set $LV_{s,v}$ and the function $\mu_{s,v}(z)$ is a clipped version of $\mu_v(z)$.

The collection of the heights $f_{s,v}$ and membership functions $\mu_{s,v}(z)$ of the fuzzy sets $LV_{s,v}$, $v=\alpha$, β , ..., constitutes the output of the inference engine.

2.3 Defuzzification

Defuzzification is the final operation assigning a numerical value in [0, 1] to the composite indicator *s*. The ASTRALIFE model can use either height method defuzzification or center-of-gravity formula for defuzzification (Driankov et al., 1996).

Height defuzzification is done in two steps:

1. We determine the peak value $p_{s,v}$ of each fuzzy set $LV_{s,v}$, $v=\alpha$, β , The peak value of $LV_{s,v}$ is the element of the maximizing set of $\mu_{s,v}(z)$, that is, $\{z: \mu_{s,v}(z) = f_{s,v}\}$ such that half of the values of the set are above $p_{s,v}$ and half are below it (see Fig. 6). Since $\mu_{s,v}(z)$ is a clipped version of $\mu_s(z)$, the maximizing set of $\mu_{s,v}(z)$ is the closed interval $[I_{s,v}, u_{s,v}]$ such that $\mu_s(I_{s,v}) = \mu_s(u_{s,v}) = f_{s,v}$. Therefore,

$$p_{s,v} = \frac{l_{s,v} + u_{s,v}}{2}$$
 (1.3)

2. The crisp value of indicator *s* is computed from the height formula of defuzzification:

$$y_s(x_1, x_2, ...) = \frac{\displaystyle\sum_{\nu=\alpha,\beta,...} p_{s,\nu} f_{s,\nu}}{\displaystyle\sum_{\nu=\alpha,\beta,...} f_{s,\nu}}$$
 (1.4)

The above procedure is illustrated in Figure 6 for a hypothetical indicator with two linguistic values LV_{α} and LV_{β} and heights $f_{s,\alpha}=0.5$ and $f_{s,\beta}=0.7$. By applying Eq. (1.4) we obtain $y_s=(0.2\times0.5+0.8\times0.7)/(0.5+0.7)=0.66/1.2=0.55$.

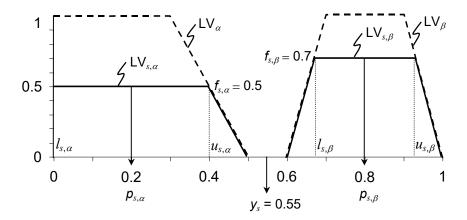


Figure 6. Illustration of height defuzzification

The center-of-gravity defuzzification is done also in two steps:

1. The outputs of the inference engine are the membership grades of s in the fuzzy subsets $LV_{s,\alpha}$, $LV_{s,\beta}$, ..., $LV_{s,\nu}$, The meaning of the whole set of membership functions is the union of all individual-function meanings, forming the composite conclusion "indicator s is $LV_{s,\alpha}$ OR $LV_{s,\beta}$ OR...OR $LV_{s,\nu}$ OR...". The fuzzy relation representing this proposition is expressed by the max-operator:

$$\max_{\mu_s(z) = v = \alpha, \beta, \dots} \mu_{s,v}(z) \tag{1.5}$$

for every $z \in [0, 1]$. It should be noted that the above membership functions are also functions of the observations $x_1, x_2, ..., x_c, ...$

2. The final crisp value of indicator s is given by the center-of-gravity formula

$$\int_{0}^{1} z \, \mu_{s}(z) \, dz$$

$$\int_{1}^{0} \mu_{s}(z) \, dz$$

$$y_{s}(x_{1}, x_{2}, ..., x_{c_{s}}, ...) = \int_{0}^{0}$$
(1.6)

We now apply the above procedure to the hypothetical indicator s whose linguistic values are shown in Figure 6. Since the membership functions of LV_{α} and LV_{β} do not overlap, Eq. (1.5) reduces to

$$\max_{\mu_s(z) \,=\, \begin{subarray}{l} \max_{\nu = \alpha,\beta} \mu_{s,\nu}(z) \\ \mu_{\beta}(z) \,=\, \begin{subarray}{l} \mu_{\alpha,\nu}(z), & z \,\in\, (0,0.5] \\ 0, & z \,\in\, (0.5,0.6) \\ \mu_{\beta,\nu}(z), z \,\in\, [0.6,1] \end{subarray}$$

Next we approximate the integrals of Eq. (1.6) by sums whose terms are obtained by sampling at the points $z_i = 0.0, 0.1, ..., 1.0$. This yields

$$\begin{split} \sum_{\substack{j \\ \sum j \\ \sum j \\ \mu_s(z_j)}} & \sum_{\substack{j \\ \sum j \\ \mu_s(z_j)}} \mu_s(z_j) \\ & \underbrace{\begin{bmatrix} 0 \times 0.5 + 0.1 \times 0.5 + 0.2 \times 0.5 + 0.3 \times 0.5 + 0.4 \times 0.5 + 0.5 \times 0 \\ + 0.6 \times 0 + 0.7 \times 0.7 + 0.8 \times 0.7 + 0.9 \times 0.7 + 1 \times 0 \end{bmatrix}}_{\begin{bmatrix} 0.5 + 0.5 + 0.5 + 0.5 + 0.5 + 0 \\ 0 + 0.7 + 0.7 + 0.7 + 0 \end{bmatrix}} & \underbrace{\frac{2.18}{4.6}}_{= 0.474} \end{split}$$

The ASTRALIFE model has been built using MATLAB's fuzzy toolbox and all computations involving Eqs. (1.1)-(1.6) are done automatically.

2.4 Candidate organisms for Mars conditions

Bibliographical studies of existing organisms that have been suggested by previous astrobiological researches for spatial experimentations on Mars have led to our selection of five candidate organisms for a comparative assessment of their astrobiotechnological perspective for Martian conditions. Specifically the bacterium *Carnobacterium* spp. (Rothschild and Mancinelli, 2001), the Black Antarctic fungi *Cryomyces antarcticus* (Onofri et al., 2008), the methanogen *Methanosarcina barkeri* (Mickol and Kral, 2017), the

lichen *Xanthoria elegans* (Sancho et al, 2007), and the lichen *Pleurosticta acetabulum* (Papazi et al. 2015).

Mars is much colder than earth (the average temperature is about -80 F (-60 °C), although it can vary from -195 F (- 125 °C) near the poles during the winter to as much as a comfortable 70 F (20 °C). The air on Mars consists of 95,97% carbon dioxide, 1,89% nitrogen, 1.93% argon, oxygen (0,146%), CO (0,0557%), water vapor, and other gases. The atmospheric pressure on the Martian surface averages 600 Pascals (0.087 psi; 6.0 mbar), about 0.6% of Earth's mean sea level pressure of 101.3 Kilopascals (14.69 psi; 1.013 bar).

On Mars, the total integrated UV radiation flux between 200-400 nm is comparable to the Earth's. However, on Mars the shorter UV wavelengths contribute a much greater proportion of this UV flux. These wavelength ranges, such as UVC (200-280 nm) and UVB (280-315nm) are particularly biologically damaging. The atmosphere is quite dusty, giving the Martian sky a light brown or orange-red color (particles of about 1.5 micrometers in diameter). The light intensity on the planet Mars is between 715 to 492 W/m² whereas the intensity of sunlight at the Earth's distance from the Sun is about 1361- 1366 W/m² [Kopp et al., 2011].

The main characteristics of these five organisms, which have been taken into consideration for their comparative assessment through ASTRALIFE model, are summarized in Tables 6 and 7.

 Table 6. Astrophysical, Biochemical and Biotechnological characteristics of the studied organisms. CRI: critical, RES: Resistant

ORGANISMS	ASTROPHYSICAL RESISTANCE		BIOCHEMICAL RESISTANCE			BIOTECHNOLOGICAL			
	>UV	<temp< th=""><th><pressure< th=""><th>>CO₂</th><th><02</th><th><hum< th=""><th>H₂</th><th>СНО</th><th>REG</th></hum<></th></pressure<></th></temp<>	<pressure< th=""><th>>CO₂</th><th><02</th><th><hum< th=""><th>H₂</th><th>СНО</th><th>REG</th></hum<></th></pressure<>	>CO ₂	<02	<hum< th=""><th>H₂</th><th>СНО</th><th>REG</th></hum<>	H₂	СНО	REG
Carnobacterium spp.	CRI	RES	CRI	CRI	CRI	CRI	CRI	CRI	CRI
Cryomyces antarcticus	RES	RES	CRI	CRI	RES	RES	CRI	CRI	RES
Methanosarcina barkeri	CRI	CRI	RES	RES	RES	RES	RES	CRI	CRI
Xanthoria elegans	RES	CRI	RES	RES	CRI	RES	RES	RES	RES
Pleurosticta acetabulum	RES	RES	RES	RES	RES	RES	RES	RES	RES

 Table 7. ASTRALIFE normalized values for the studied organisms

ORGANISMS	ASTROPHYSICAL RESISTANCE			BIOCHEMICAL RESISTANCE			BIOTECHNOLOGICAL CAPACITY		
	UV	PRESSURE	TEMP	H ₂ O	O ₂	CO ₂	СНО	H ₂	REG
Carnobacterium spp.	[0;200] > 250 0	[0-1013] < 6 0.994	[-100;50] < -60 0.78	[0-100] < 0.1 0.99	[16-21] < 0.1	[0;100] > 95,97 0	[0;136] < 580 0	[0;100] < 0,1	[0;100] > 0 0
Cryomyces antarcticus	[0;200] > 250 0	[0-1013] < 6 0.994	[0-50] < -60 0	[0-100] < 0.1 0.99	[16-21] < 0.1	[0;100] > 95,97 0	[0;1361] < 580 0	[0;100] < 0,1	[0;100] > 50 0.50
Methanosarcina barkeri	[0;200] > 250 0	[0-1013] < 6 0.994	[0-50] < -60 0	[0-100] < 0.1 0.99	[0;21] < 0.1 0.99	[0;100] > 95,97 0.95	[0;1361] < 580 0	[0;100] < 0,1 0.99	[0;100] > 0 0
Xanthoria elegans	[0;300] > 250 0.83	[0-1013] < 6 0.994	[-129; 50] <- 60 0.614	[0-100] < 0.1 0.99	[0;100] < 0.1 0,99	[0;100] > 95,97 0.95	[0;1361] < 580 0,57	[0;100] < 0,1 0.99	[0;100] > 100 1
Pleurosticta Acetabulum	[0;300] > 250 0.83	[0-1013] < 6 0.994	[-129; 50] <- 60 0.614	[0-100] < 0.1 0.99	[0;100] < 0.1 0,99	[0;100] > 95,97 0.95	[0;1361] < 580 0,57	[0;100] < 0,1 0.99	[0;100] > 100 1

3. Results

The ASTRobiotechnological Assessment for LIfe by Fuzzy Evaluation (ASTRALIFE) methodology has been implemented for the comparative assessment of the astrobiotechnological perspective of the studied organisms with respect to the Martian conditions. In Table 8, the results of the analysis are presented including the ASTRALIFE index, which can be used for the final evaluation.

Given the targets, natural limits, relevant principles and criteria of astrobiotechnological capability and survivability as well as statistical observations, normalized values are derived for each indicator (see Table 8).

An indicator may qualitatively utilize direct and precise information regarding defined objectives of survivability, but it may also use indirect or imprecise information leading to imprecise objectives of survivability. The choice of indicators is limited by the available data, but their use demonstrates the procedure of the ASTRALIFE methodology. The selected indicators possess the following attributes:

- Spatial attributes. Data should be available for all organisms and different measurement conventions should permit comparisons.
- 2. Time attributes. Time series of data for all organisms should be available.
- Goal driven attributes. Indicators should be linked to specific survivability goals or give a concrete indication of trends towards survivability goals.

Overall survivability measurements for five selected organisms show that lichens are more resistant than the others.

 Table 8. ASTRALIFE Index for the selected organisms

LIFEFORMS VARIABLES	Carnobacterium spp.	Cryomyces antarcticus	Methanosarcina barkeri	Xanthoria elegans	Pleurosticta acetabulum
TEMP	0.78	0	0	0.614	0.78
RAD	0	0	0	0.83	0.83
PRE	0.99	0.99	0.99	0.99	0.99
ASTRO	0.18	0.14	0.14	0.58	0.58
ним	0.99	0.99	0.99	0.99	0.99
O ₂	0	0	0.99	0.99	0.99
CO ₂	0	0	0.95	0.95	0.95
SURVI	0.14	0.14	0.82	0.75	0.75
H ₂	0	0	0	0.99	0.99
СНО	0	0	0	0.57	0.57
REG	0	0.5	0	1	1
BIOTEC	0.14	0.27	0.17	0.51	0.51
ASTRALIFE Index	weak	weak	weak	medium	medium
(%)	29%	29%	28%	51%	51%

Fuzzy computations are done with the aid of MATLAB's fuzzy logic toolbox (The MathWorks Inc., 1995-96) and the center-of-gravity method of defuzzification is chosen because of its higher precision. Levels of astrobiotechnological perspective are given in percentages, which can easily be understood. A completely viable and productive organism gives a value of 100% whereas a dead organism gives a value of 0%.

4. Discussions

The resulted higher advantages of fungi and lichens are in line with the finding of the recent astrobiological research through the BIOMEX (Biology and Mars Experiment) experimentation on space (Pacelli et al, 2017). BIOMEX is an experiment where microbes were exposed to space and Mars-like conditions in the EXPOSE-R2 setup, a platform outside the International Space Station (ISS). It is led by Jean-Pierre de Vera, from the German Aerospace Center (DLR), together with a team from 27 institutes in 12 countries on three continents (Rabbow et al, 2017). After leaving Earth in June 2014, samples were exposed outside the ISS for 480 days and came back on June 18th 2016. The BIOMEX consortium investigated how the journey affected them. They also assessed the habitability of Mars and the so-called 'lithopanspermia" theory, according to which life could have travelled from one planet to another (Pacelli et al, 2017).

As we enter the new era of climate change, one of the most challenging questions, if not the most challenging one to be addressed, is how to assure the survivability of living organisms on Earth or how to transfer life in other planet close to the Earth. It is fair to say that some clear measures or, at least, indicators of survivability exist, but the effectiveness of scientific proof towards a goal of survivability cannot be assessed. Attempts have been made to measure survivability using temperature, pH, or salinity approaches, but the results still lack universal acceptance.

Not only are there no common units of measurement for the indicators of organisms' survivability in extreme conditions, but quantitative criteria for certain values are still lacking. A systemic method based on a reliable scientific methodology is needed to combine multidimensional components and assess uncertainty. Such a method should be flexible in the sense that one can add or remove indicators to achieve a better assessment of the system according to the context. In reality the border between survivability and mortality is not sharp but rather fuzzy. This means that it is not possible to determine exact reference values for survivability and a scientific evaluation of uncertainty must always be considered in the procedure of survivability assessment. For this reason, the use of natural language and linguistic values based on the fuzzy logic methodology, which followed above, seems more suitable to assess survivability.

Moreover, the new dimension, which has only recently been investigated, the astrobiotechnological ability of organisms, strengthens the above followed methodology. Recently, scientists have found that lichens can produce high amount of molecular hydrogen (H₂) when incubated in a closed system without oxygen (Papazi et al., 2015). The ability of lichen to produce molecular hydrogen after dehydration and exposure to extremely low temperature (-196 °C/ 77 K) was also studied and the conclusion is that the H₂ production remains unchanged after environmental stresses. In all cases, O₂ is completely consumed in a closed system, and anoxic conditions are subsequently established, and high yield of H₂ is then being achieved. Similar results were observed when the experiment was repeated under dark conditions. These results indicate that intense dehydration and extremely low temperature do not affect H₂ production by lichens.

Given that the Martian environment is deprived of oxygen and humidity, lichens can survive and after hydration may achieve high yield of hydrogen production under dark and light conditions. Hydrogen then can be used as renewable energy resource for human

exploitation. This implies an important astrobiotechnological perspective for lichens giving to this category of organisms a great advantage compared to others.

There are three different biochemical pathways which may explain the hydrogen production by lichens under different environmental conditions (Papazi et al., 2015.): Under light incubation if the lichens are placed in closed system, anoxic conditions are established, because of the over activation of oxygen consumption through the respirational electron transport chains (from mycobiont and photobiont). The oxygen depleted conditions induce the activation of the hydrogenase and create the optimal conditions for hydrogen generation. Specifically, Electrons from PSII transferred through PSI to ferredoxin and then to hydrogenase (PSII-dependent pathway). Additionally, the hydrogen production was further induced by the reduction of organic substrates through the PSII-independent pathway. These electrons are led to the plastoquinone pool and through PSI and ferredoxin, are transferred to hydrogenase for hydrogen production (PSII-independent pathway). In parallel, electrons are alternatively allocated to pyruvate and through the PFOR protein result in ferredoxin and hydrogenase in order to produce H₂ (dark fermentation pathway). This light independent fermentative pathway is the dominant route for H₂ production by lichens (Papazi et al. 2015).

Molecular hydrogen is considered to be the cleanest renewable energy of the future as it has high efficiency and it gives only water after combustion without any risk of toxic byproducts (Antal et al., 2011). The limiting factors for the production of hydrogen are the inhibition of the enzyme hydrogenase by oxygen, the low level of hydrogenase activation and the possible reversible function of hydrogenase to consume hydrogen (Ghirardi and Togasaki, 1997; Dubini, 2014).

The innovative technique for the production of hydrogen by lichens is based on the fact that, in a closed system, mainly the mycobiont part of the lichen consumes immediately the

remaining oxygen through respiration activities and, simultaneously, the photobiont part produces hydrogen in a completely anaerobic environment that is favorable for the enzyme hydrogenase

In general, the followed methodology seems to be a valuable tool for measuring survival capacity and a tool for simulating survivability scenarios in a manner that could help scientist to design a rational path to design policies.

Suggestions regarding the values of indicators are restricted to subjective and extrapolation terms. Assigning quantitative values through laboratory experimentation is another bigger issue, which is not dealt with in this work, but would require the formulation of a constrained optimization problem. The use of fuzzy logic optimization and survival capacity experiments is the basis of the best strategy for survivability optimization on extraterrestrial environment. This could be the subject of possible future research resulting from ASTRALIFE project.

5. Conclusions

Most existing methods for the study of organisms' survivability perspective on extreme conditions or on extraterrestrial environment use only pure laboratory experiment. Moreover laboratory experimental data are sparse and do not cover the effect of combined limiting factors that are rarely encountered in nature. Given that real spatial experiments are still exceedingly expensive, we need an alternative tool to interpret experimental data for the study of combined extreme environmental factors.

Because of its capacity to handle polymorphic dimensions and complex subjects, fuzzy logic evaluation is used to combine astrobiological data and biotechnological findings results in

order to validate methodological theories for organisms' resistance and usability on space missions.

Recent finding at the University of Crete on the extremophile behavior of lichens as well as on their capacity to produce hydrogen under Mars anoxic conditions inspired this study offering new parameters for the comparable assessment of the astrobiotechnological perspective of organisms.

For the needs of the current study a new fuzzy model entitled ASTRALIFE (Astrobiotechnological perspective Assessment for Life by Fuzzy Logic Evaluation) was created and calibrated. It consists in the simulation of the effect of spatial environmental factors and encompasses astrophysical, biochemical, and biotechnological parameters of extraterrestrial environment.

Using commonly available parameters of space transferability, survivability and biotechnological capacity of each organism, a crisp measure of overall astrobiotechnological perspective, ASTRALIFE index, computed from nine basic indicators of astrobiotechnological conditions and three primary linguistic variables. All the linguistic variables of the ASTRALIFE model are considered to be indispensable in the computation of the astrobiotechnological perspective of organism candidate for space mission on Mars.

The results of the ASTRALIFE assessments of studied organisms, three resistant microorganism and two lichen species, demonstrated the higher capacity of lichen for astrobiotechnological perspectives on Mars conditions, providing also new insights on the capacity of organisms in panspermia scenarios.

Fuzzy modeling can simulate and support astrobiotechnological experimentations, theory construction and decision-making. Future hypotheses and experimentations on organisms'

resistance and usability on space missions can improve the calibration of ASTRALIFE for the comparative assessment of organisms' astrobiotechnological perspective.

Last but not the least, the fuzzy Logic model ASTRALIFE can support the emerging needs of the two newly introduced concepts that are (1) the expansion of the panspermia theory from species to micro-ecosystemic approach and (2) the shift from astrobiology to astrobiotechnology.

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APPENDIX

Illustration of fuzzy computations

We present a numerical example illustrating how the ASTRALIFE model assesses biotechnological perspective and performs sensitivity analysis.

For simplicity, we only consider the secondary variable ASTRO and its components PRESSURE (PRE), UV RADIATION (RAD), and TEMPERATURE (TEMP). We use three fuzzy sets, weak (W), medium (M), and strong (S) to represent the basic variables' linguistic values (Fig. A1) and five fuzzy linguistic values for ASTRO, very low (VL), low (L), intermediate (I), high (H), and very high (VH). Table A.1 gives the corresponding rule base, which consists of $3^3 = 27$ rules (see also Table 3).

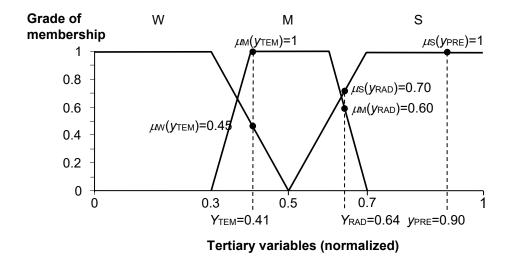


Figure A1. Linguistic values and fuzzification of crisp inputs.

Table A1. Third order rule base for the computation of ASTRO

Rule	if	and	and	then
r	PRESSURE is	UV RADIATION is	TEMPERATURE is	ASTRO is
1	weak	strong	strong	very high
2	weak	medium	strong	very high
3	weak	weak	strong	high
4	weak	strong	medium	very high
5	weak	medium	medium	high
6	weak	weak	medium	intermediate
7	weak	strong	weak	high
8	weak	medium	weak	intermediate
9	weak	weak	weak	very low
10	medium	strong	strong	very high
11	medium	medium	strong	high
12	medium	weak	strong	intermediate
13	medium	strong	medium	high
14	medium	medium	medium	intermediate
15	medium	weak	medium	low
16	medium	strong	weak	intermediate
17	medium	medium	weak	low
18	medium	weak	weak	very low
19	strong	strong	strong	high
20	strong	medium	strong	intermediate
21	strong	weak	strong	low
22	strong	strong	medium	intermediate
23	strong	medium	medium	low
24	strong	weak	medium	very low
25	strong	strong	weak	low
26	strong	medium	weak	very low
27	strong	weak	weak	very low

Suppose that information concerning the tertiary variables is expressed numerically as follows: PRESSURE has the value $y_{PRE} = 0.90$, UV RADIATION $y_{RAD} = 0.64$, and TEMPERATURE $y_{TEM} = 0.41$. Fuzzification (see Fig. A1) yields the following inputs of the inference engine:

<u>Input 1</u>: PRESSURE is *strong* with membership grade $\mu_S(y_{PRE}) = 1$;

Input 2: UV RADIATION is *medium* with membership grade $\mu_M(y_{RAD}) = 0.60$

and *strong* with membership grade $\mu_s(y_{RAD}) = 0.70$;

Input 3: TEMPERATURE is *medium* with membership grade $\mu_M(y_{TEM}) = 1$

and weak with membership grade $\mu_W(y_{TEM}) = 0.45$.

We now compute the degree to which each rule is applicable to the input. Using the minoperator to represent the AND connectives of rule r, r = 1, ..., 27,

$$\mu_{PREMISEr} = min\{\mu_i(y_{PRE}), \mu_i(y_{RAD}), \mu_k(y_{TEM})\}$$

where $\mu_{PREMISEr}$ is the degree to which rule r is applicable and i, j, k \in {W, M, S}. The only consistent rules are those in which PRESSURE is *strong*, UV RADIATION is either *strong* or *medium*, and TEMPERATURE is either *weak* or *medium*. These are rules 22, 23, 25, and 26 of Table A.1. The conclusions of these rules are expressed as follows:

Rule 22: If PRESSURE is *strong* with membership grade 1 and UV RADIATION is *strong* with membership grade 0.70 and TEMPERATURE is *medium* with membership grade 1, then ASTRO is *intermediate* with membership grade $\mu_{PREMISE22} = 0.70$ (= min {1, 0.70, 1}).

Rule 23: If PRESSURE is *strong* with membership grade 1 and UV RADIATION is *medium* with membership grade 0.60 and TEMPERATURE is *medium* with membership grade 1, then ASTRO is *low* with membership grade $\mu_{PREMISE23} = 0.60$ (= min {1, 0.60, 1}).

Rule 25: If PRESSURE is *strong* with membership grade 1 and UV RADIATION is *strong* with membership grade 0.70 and TEMPERATURE is *weak* with membership grade 0.45, then ASTRO is *low* with membership grade $\mu_{PREMISE25} = 0.45$ (= min {1, 0.70, 0.45}).

Rule 26: If PRESSURE is *strong* with membership grade 1 and UV RADIATION is *medium* with membership grade 0.60 and TEMPERATURE is *weak* with membership grade 0.45, then ASTRO is *very low* with membership grade $\mu_{PREMISE26} = 0.45$ (= min {1, 0.60, 0.45}).

For the remaining rules of the rule base, we have $\mu_{PREMISEr} = 0$. We observe that rules 23 and 25 assign the same linguistic value *low* to ASTRO. Applying Eq. (1.1) we combine the conclusions of these rules into a single conclusion whose truth-value is given by

$$f_{ASTRO,L} = max \{ \mu_{PREMISE23}, \mu_{PREMISE25} \} = 0.60$$

where subscript L stands for low. From the other two rules we obtain

$$f_{ASTRO,I} = \mu_{PREMISE22} = 0.70$$

$$f_{ASTRO,VL} = \mu_{PREMISE26} = 0.45$$

where I and VL signify *intermediate* and *very low*, respectively. The above membership grades constitute the output of the inference engine. The inference process for ASTRO is illustrated in Fig. A2. This figure shows also the membership functions of the linguistic values assigned to ASTRO. Since the membership functions of *low* and *intermediate* are symmetric about the normalized indicator values 0.3 and 0.5, respectively, the peak values used in height defuzzification are invariant and equal to these values. The peak value of the fuzzy subset of *very low* corresponding to f_{ASTRO} , $v_L = 0.45$ is obtained from Eq. (1.3): $p_{ASTRO,VL} = (0+0.21)/2 = 0.105$. Applying Eq. (1.4) for defuzzification we obtain a crisp value for ASTRO.

$$\gamma_{\text{ASTRO}} = \frac{0.45 \times 0.105 + 0.60 \times 0.3 + 0.70 \times 0.5}{0.45 + 0.60 + 0.70} = \frac{0.57725}{1.75} = 0.329857$$

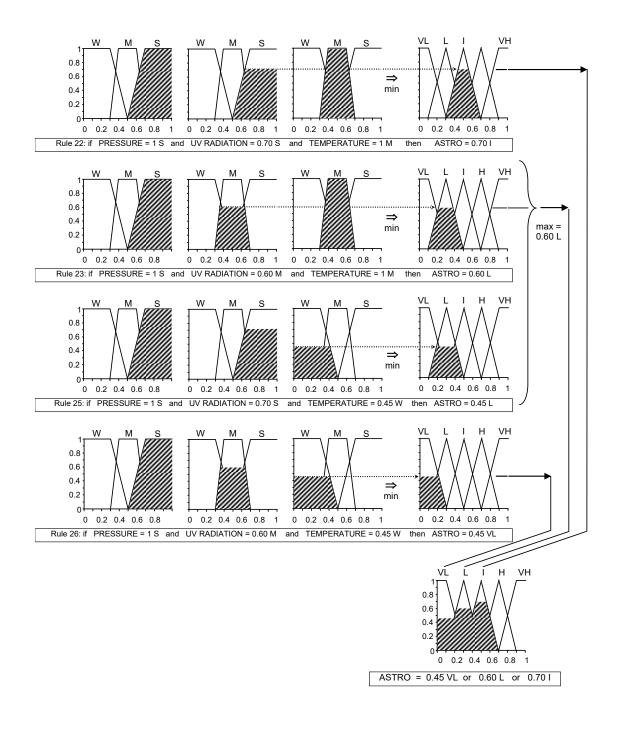


Figure A2. Inference using rules 22, 23, 25, and 26.