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Bachelor Thesis

Utilization of population viability analysis in amphibian conservation

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Thesis title

Utilization of population viability analyses in amphibian conservation

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Objectives of thesis

Population viability analyses (PVAs) represent a complex of quantitative modeling methods to predict the likely future status of a population of conservation concern. The tool is used worldwide in nature conservation across all taxa including amphibians. However, in the Czech Republic, this useful tool still has been used rarely.

The goal of the bachelor thesis is to create comprehensive literature review focused on: (i) the principles of PVAs – their potential uses, data and sources required including their quality, software tools which are used for analyses, restrictions and weaknesses of PVAs; (ii) examples of PVA uses in nature conservation focusing mainly on amphibians and on the status and extent of PVA use in the Czech Republic; (iii) establish concise instructions for the usage of a selected software (probably VORTEX).

Methodology

To write literature review, all accessible and relevant publications will be used, mainly thematic scientific papers, but also books and web pages. Selected freeware will be downloaded and thoroughly studied. To find out and describe the current status and extent of using PVAs in the Czech Republic, the potential users of PVA at universities and conservation agencies will be contacted.

The proposed extent of the thesis

20–30 pages and appendices

Keywords

PVA, conservation biology, species conservation, VORTEX

Recommended information sources

Akçakaya H.R. and P. Sjögren-Gulve. Population viability analysis in conservation planning: an overview. Ecolog. Bulletins 2000. 48:9-21.

Brook, B. W. et al. Critiques of PVA ask the wrong questions: throwing the heuristic baby out with the numerical bath water. Conserv. Biol. 2002. 16: 262-263.

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Declaration

I declare that I have worked on my bachelor thesis titled "Utilization of population viability analyses in amphibian conservation" by myself and I have used only the sources mentioned at the end of the thesis. As the author of the bachelor thesis, I declare that the thesis does not break copyrights of any their person.

In Prague on 1.4.2019 ______________________________

Acknowledgement

I would like to thank my supervisor doc. Ing. Jiří Vojar, Ph.D. for helping me in writing of this bachelor thesis, for his advice, support and giving me appropriate data that were used in my work and VORTEX software.

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Abstract. This bachelor thesis provides an introduction for understanding the principle of the population viability analysis (PVA) and how it works, using *Rana dalmatina* as an example. After many years of discovering about amphibian declines and trying to pinpoint the exact cause of the problem, we can use software which can help us to predict population extinction. About half of amphibian species are in decline, and one third is already in danger of extinction. Population viability analyses represent a complex of quantitative modelling methods to predict the future status of a population of conservation concern. Population viability analysis is a very useful tool in conservation biology of threatened or endangered species. The tool is worldwide used in nature conservation across all taxa including amphibians. However, in the Czech Republic, this useful tool still has been neglected.

Keywords: amphibians, amphibian conservation, the agile frog, *Rana dalmatina*, spoil banks, VORTEX.

Abstrakt. V této bakalářské práci je uveden úvod který popisuje analýzy životaschopnosti populace (Population Viability Analysis) na příkladu druhu *Rana dalmatina*. Po mnoha letech objevování o poklesu obojživelníku a snaze přesně určit příčinu problému uhynuti jejich populace, můžeme použít software, který nám může pomoci předpovědět vymírání populace. Přibližně polovina druhů obojživelníků je na ústupu a třetina je již ohrožena vyhynutím. Analýzy životaschopnosti obyvatelstva představují komplex metod kvantitativního modelování, které předpovídají pravděpodobný budoucí stav populace, která se týká ochrany přírody. Analýz životaschopnosti obyvatelstva se stal běžně používaným nástrojem v biologii ochrany ohrožených nebo ohrožených druhů. Nástroj je celosvětově používán v ochraně přírody napříč všemi taxony včetně obojživelníků. V České republice je však tento užitečný nástroj stále opomíjen.

Klíčová slova: obojživelníci, ochrana obojživelníků, skokan štíhlý, *Rana dalmatina*, výsypky, VORTEX.

Table of content

1. Introduction

Many scientists approve that we are either entering or in the midst of the sixth great mass extinction. Intense human pressure, both direct and indirect, is having profound effects on natural environments. The amphibians: frogs, salamanders, and caecilians may be the only major group currently at risk globally (Wake and Vredenburg, 2019). Amphibian population declines were initially recognized as a global phenomenon in the early 1990s. Nowadays current extinction rates for amphibians may be as much as 200 times higher than background (Hayes et al., 2010). Amphibians are usually encountered during the breeding season when they often produce loud breeding choruses or form aggregations at ponds, and because of non-breeding behaviour or movements, a very little amount of them is known. Moreover, they are difficult organisms to study at the population level (Bishop et al., 2019).

In this case, effective protectionist measures need to be implemented. These include the protection of species and their environment, international cooperation and practical protection. Any protection should be based on evidence-based conservation that is a relatively new tool being used to streamline relevant knowledge, improve effectiveness and support decision making in conservation management. Several Conservation Evidence synopses have been compiled for various species groups and habitats, aimed at providing conservation practitioners, scientists and policymakers with a relevant list, and accompanying methods, of realistic interventions to conserve biodiversity (Tarrant and Weldon, 2014).

One of the most useful sophisticated protection tools is population viability analysis, that represents a collection of methods for evaluating the threats faced by populations of species, their risks of extinction or decline, and their chances for recovery, based on species-specific data and models (Akcakaya and Sjögren-Gulve, 2000). Compared with other alternative environmental solutions, PVA is one of the most popular methods that is widely used abroad. It includes the ability to be used in simple and complex models, including spatial and temporal variations. These models can assess the effect of habitat quality, migration rates between population groups, and genetic effects (for example, inbreeding depression) on the viability of a population (Keedwell, 2014).

Users of statistical software are expected to be sufficiently familiar with the methods of statistical analyses to be able to choose appropriate models to apply to their problem, to be able to provide the proper input and to be able to interpret the results (Lacy, 2000). PVA has been used for a range of species from large mammals to birds, reptiles, invertebrates, fish, and plants. The tool is also worldwide used in nature conservation, for example (Keedwell, 2014):

1. accounting for uncertainty in risk assessment. Slooten et al. (2000), used population variability analysis to discover how Hector's dolphins' survival is affected by levels of uncertainty;
2. effects of harvesting and predator control. Hamilton and Moller (1995) using population variability analysis, tried to find out whether the mainland population is declining and understand whether harvesting on the mainland could be sustainable;
3. Survival prospects of populations under threat of predation. Elliott (1996), used a population variability analysis computer model to control the effects of frequency of predation episodes, predation of adults, the number of broods, and the carrying capability on the extinction probabilities of mohua populations;
4. effects of aerial poisoning of rodents on forest bird populations. Armstrong and Ewen (2001), used population variability analysis to analyse the effects of administration on the robin population.

In the Czech Republic, PVAs are prepared for species that are at a very high risk of extinction, unless corrective action is taken without delay. In case of a lack of data required to carry out the PVA, work on the rescue program should be directed to obtain data so that PVA can be implemented as it is one of the basic planning tools management measures to support the species. When assessing the risk of extinction, it should be taken into account time trends (population trends) and spatial arrangement of populations based on current knowledge spatial ecology (Ministerstvo životního prostředí). For example, in the Czech Republic, population variability analyses were used for some plants and mammals. So far, these tools have not been used for amphibians. One of the reasons (how it was already mentioned above) is the complexity of input data quality, where it is necessary to obtain a lot of relevant information about the population.

As part of a long-term project (since 2004), *Rana dalmatina* (the agile frog) is monitored on spoil banks in Northwest Bohemia. During this time, sufficient evidence was obtained of the biology and ecology of the local populations necessary to carry out the analysis. At the same time, they are very endangered populations, because in some areas reclamation or other mining is planned. The realization of PVA analyses will thus enable to predict possible impacts of these activities and to take necessary precautions to protect these endangered animals.

2. Objectives

The goal of the bachelor thesis is to elaborate a comprehensive literature review focused on:

a) The principles of PVAs – their potential uses, data, and sources required including their quality, software tools which are used for analyses, restrictions, and weaknesses of PVAs;
b) Examples of PVA uses in nature conservation focusing mainly on amphibians and on the status and extent of PVA used in the Czech Republic;
c) Establish concise instructions for practical using of a selected software (probably VORTEX).

In this work, you will find a short description of the advantages and disadvantages of population viability analysis and discussion about the most useful PVA programmes which are used in conservation planning. Also, you get an example of using PVA analysis for a model population of the agile frog (*Rana dalmatina*) in Hornojiřetínská spoil ponds in North-west Bohemia in the Czech Republic.

3. Literature Review

To write a literature review, all accessible and relevant publications will be used, mainly thematic scientific papers, but also books and web pages. Selected freeware will be downloaded and thoroughly studied. To find out and describe the current status and extent of using PVA in the Czech Republic, the potential users of PVA at universities and conservation agencies will be contacted. In the first part of bachelor work you can find the description of the basic principles of PVA, how it can be used in nature conservation, its advantages and disadvantages, data we need and a short description of some PVA's programmes. In the second part will be an example of utilization one of PVA's programmes – VORTEX which was applied to *Rana dalmatina* in the the Czech Republic for better understanding of using population variability analysis in nature conservation.

3.1 The basic principles of PVA

Population viability analysis (PVA) is a modelling tool that estimates the future size and risk of extinction for populations of organisms. PVA works by using life-history or population growth-rate data to parameterize a population model that is then used to project dynamics and estimate future population size and structure. (Coulson et al., 2001). PVA software packages allow predicting future population sizes and risks of extinction for any population that has been chosen. It remains an important form of risk analysis for the management of threatened species. This is the quantitative evaluation of factors that influence population growth and persistence for single species (Heard et al., 2013). It is a process of identifying the viability requirements of, and threats faced by a species and evaluating the likelihood that the population(s) under study will persist for a given time into the future (Akcakaya and Sjogren-Gulve, 2000), also it works like as a central tool for conservation assessments. PVA is an important tool for the management of endangered species and information generated by these models must be considered for conservation policies. These models can be used to predict the fate of a species in short and middle term, but can also help to implement conservation actions on a particular species (Zambrano L. et al., 2007).

3.2 Using of PVA in nature conservation

Population viability analysis (PVA) is used to quantify the risks faced by species under alternative management regimes. For example, Bayesian PVAs allow uncertainty in the parameters of the underlying population model to be easily propagated through to the predictions (Heard et al., 2013). Models of population variability analysis are tools for organizing relevant information and assumptions about a species or population, also they can predict viability indicators based on demographic data: surveys and observations of reproduction and dispersion, presence / absence data, census, recapture studies (Akcakaya and Sjogren-Gulve, 2000). PVAs can be used to (Coulson et al., 2001.):

1. predict the future size of a population;
2. estimate the probability of a population going extinct over a given time;
3. assess which of a suite of management or conservation strategies is likely to maximize the probability of a population persisting;
4. explore the consequences of different assumptions on population dynamics for small populations.

In reality, only the predictive accuracy of the first two cases is estimable, as there are rarely sufficient replicate populations from which to collect data to determine whether the comparative predictions of the third use are accurate, and the fourth use has not generated testable predictions. The applications of PVA are also now less concerned with the outright risk of extinction and instead focus on comparative risks. Given a set of options for future management, analysis seek to quantify and compare the relative risk of extinction under each. Population viability analysis is often oriented towards the management of rare and threatened species, with two broad objectives. The short-term objective is to minimize the risk of extinction. The longer-term objective is to promote conditions in which species retain their potential for evolutionary change without intensive management. Population viability analysis (PVA) may be used to address the following aspects of management for threatened species (Akcakaya and Sjogren-Gulve, 2000):

Planning research and data collection. PVA may show that population viability is indifferent to particular parameters. Research may be based on targeting factors that may have an important effect on the probabilities of extinction or recovery.

Assessing vulnerability. PVA may be used to assess the vulnerability of a population to extinction. The results will be used to determine priorities for their preservation.

Impact assessment. PVA may be used to compare model results with the consequences of human activity at the population level or without it.

Ranking management options. PVA may be used to predict the reaction of species on reintroduction, captive breeding, prescribed burning, weed control, habitat rehabilitation, or different designs for nature reserves or corridor networks.

3.3 Restrictions and weaknesses of PVA

Population variability analysis is one of the central tools for conservation planning and evaluation of management options. Compared to other methods reviewed above, PVA has several advantages (Akcakaya and Sjogren-Gulve, 2000). Brook et al. have tested the predictive accuracy of PVA using data from many populations and conclude that population variability analysis is not a useless tool and that it should not be dispensed with in favour of alternative untested methods (Coulson et al., 2001). There have been few observational attempts to verify the precision of Population viability analyses. For example, population variability analysis is good at predicting the future dynamics of populations. In the article, Coulson et al., 2001 present arguments that "PVAs can only be accurate at predicting extinction probabilities if data are extensive and reliable, and if the distribution of vital rates between individuals and years can be assumed stationary in the future, or if any changes can be accurately predicted."

The disadvantages of population variability analysis include its homogeneity of data requirements, which can be not appropriate for many species. Although PVA is a useful technique, it can be used incorrectly, potentially to the detriment of the species being modelled. A quality PVA cannot be performed without sufficient data on the target species (Keedwell, 2014). PVAs are most useful when they address a specific question involving the main species, and when they focus on comparative rather than absolute results, and risks of decline rather than extinction (Akcakaya and Sjögren-Gulve, 2000).

PVAs could be useful for comparing the consequences of different management or conservation strategies, and for exploring theoretically the implication of model assumptions on extinction probabilities and population dynamics. However, exist a doubt about the general claim that they can be accurate in their ability to predict the future status of wild populations (Possingham et al., 1993).

3.4 Examples of PVA using in amphibian conservation

Amphibians are the most endangered group of vertebrates, these declines stem from a variety of causes including habitat destruction, climate change, disease, ozone depletion and introduced predators. For example, habitat fragmentation can cause the isolation of populations, so we can face the problem of the genetic exchange, which contributes to reducing the overall genetic diversity.

The decline in some amphibian species can also be the result of high population fluctuations associated with their population characteristics. In these cases, the analyses of these attributes are imperative in order to elucidate the fate of an endangered population in an intensively managed system, by knowing which age classes are more vulnerable to modifications, and consequently, to identify major threats for a particular population (Zambrano et al., 2007). The key question in preserving a population is whether populations and species can survive in isolated locations, or need regional connectivity for long-term survival (Kirchhoff et al., 2017).

Viability analyses (PVA) is a common tool used to evaluate sustainability by calculating population persistence in response to changes in demographic parameters. Such analyses can also determine the minimum viable population size necessary for population persistence (Green and Bailey, 2015), but has been little used on amphibians. Population variability analysis is particularly useful in small population sizes. By estimating the individual fate of each member of the population, PVA simulates temporal population changes and estimates extinction risk over a time period (Zambrano et al., 2007).

3.5 Tools used for PVA

3.5.1 Data and their quality

The amount of data needed to build a PVA model depends mostly on the question addressed and on the ecology of the species. The assumptions of a PVA can be (and should be) explicitly stated and enumerated; they can also be validated given sufficient data (Akcakaya and Sjogren-Gulve, 2000).

In order to get the most reliable information, we need to know which data we require and which criterion it must belong to. Criterion one: data quality. The first of these criteria will only be met in a handful of cases where a large amount of information is known about the biology of the target species and population. Criterion two: the predictive accuracy of a PVA will depend on the purpose to which it is being applied. So, the question is if good estimates of the distribution of vital rates can be made, are the shapes, means, temporal variances and autocorrelation of these distributions likely to apply into the future? There are biotic and abiotic phenomena that can lead to changes in the shape, mean, temporal variance and autocorrelation of these distributions over time. Such processes can be classified into two categories (Coulson et al., 2001.):

1. those that are the result of a catastrophe;
2. those that result in a longer-term change in the processes and vital rates that limit the population growth rate.

Data derived from the computer simulation analysis using VORTEX were (Brito and Fernandez, 1999):

a) population growth rate (± 1 SD);
b) probability of population and metapopulation extinction;
c) median time to extinction;
d) mean population size (± 1 SD);
e) the decline in genetic variability (expressed as the expected heterozygosity or gene diversity).

The types of data that can be used in PVA include the habitat of the population, their vital rates (percentage of fertility and survival), as well as spatial fluctuations and changes in these parameters. They can also use presence-absence data, habitat relationships, GIS data on landscape characteristics, mark-recapture data, surveys and censuses (Akcakaya, and Sjogren-Gulve, 2000). The more data, the more detailed models you can build. As more details we include, as realistic, it becomes and allows for more specific issues to be resolved. However, in most practical cases, the available data allows only the simplest models.

3.5.2 Software used for PVA

Population viability analyses are widely used in conservation biology to compare management strategies and predict probabilities of extinction for endangered species. (Brook et al., 1999). PVA introduce a small variety of software, that we can use for our objectives. Applying different modelling systems allows us not only to indicate problems with data, model structure or specific parameters but also allowed us to take advantage of a wider range of outputs. In this paragraph, you can familiarize with some of them.

Widely available PVA software can serve the same role as do statistical analyses packages. The ease of use, flexible application to diverse needs, and extensive prior testing facilitate many applications that would not otherwise be attempted. Individual population processes can be modelled in various ways, requiring different sets of driving variables, using different output to describe the population dynamics (Lacy, 2000). As a result, a potential problem for many conservation biologists may be the selection of the program that is best suited to answer specific management questions and the attributes of the target species (Lindenmayer et al., 1995). PVA comparisons should ideally be carried out on a varied range of taxa (i.e. mammals, birds, amphibians, reptiles, fish and insects), to determine whether PVA packages differ in the consistency and sensitivity of their predictions depending on life-history strategy (Brook et al., 1999). Computer simulation programs are being used increasingly in the assessment of the viability of populations of threatened or vulnerable organisms and in the development of applied conservation strategies. Computer modelling programs are increasingly used in developing strategies for the preservation viability of populations of threatened or vulnerable organisms, as well as in assessing the viability of the population. The most used for this purposes are three computer packages: ALEX (Analysis of the Likelihood of EXtinction), RAMAS/space (Risk Assessment and Management Alternatives

System) and VORTEX (from the "extinction vortex"), which were used to analyse the viability of metapopulation.

The main goal is to choose the most suitable program for your purposes, since not all their functions can be used. The function used in one study may not be suitable for another.
The selection of the most appropriate program should be based on a range of key criteria including: (1) the key question(s) and objectives of the study, and, (2) the strengths, limitations and assumptions that underpin the program and how these match the attributes, life history parameters and available data for the target species (Lindenmayer et al., 1995). Data collection processes give the user a more complete picture of the behaviour and dynamics of the population. A wide variety of threatened taxa and a variety of conservation problems underscore the value of computer packages for modelling the viability of metapopulations. In this paragraph, we review three of the most popular computer simulation programs that are used for the analyses of metapopulation dynamics: ALEX, VORTEX, and RAMAS/space. All three programs are widely available and have been applied in many studies of threatened species. This investigation aims to review the strengths, limitations and assumptions of each model, and more generally to evaluate the usefulness of programs of this kind in metapopulation viability analyses (Lacy, 2000).

VORTEX

Users of PVA models should understand the basic structure of models they use, and it is important that models used for scientific studies and conservation efforts can be examined and replicated. Yet often the details of PVA computer programs are not available to the users, because the code is proprietary information or otherwise not provided to users, or simply because the task of reading and understanding the source code for large and complex programs is formidable. (Lacy, 2000). The Conservation Breeding Specialist Group (CBSG) of the World Conservation Union (IUCN) has conducted more than 80 PVAs using VORTEX (Brook et al., 1999). VORTEX is an individual-based simulation model that follows the fates of each animal in the population and simulates the events of its life history such as birth, death and catastrophe as discrete events that happen according to defined probabilities (Zeoli et al., 2008). This package is one of the most often used programs for PVAs that focus on endangered populations, including in workshops with officers from conservation and land management agencies (Lindenmayer et al., 1995).

ALEX

ALEX is a Monte Carlo simulation model. Pseudorandom numbers are used to simulate the stochastic processes in the model. Every scenario must be run many times to gather statistics on the likelihood of extinction. The user specifies the number of runs for each scenario and the length of the simulation in years. ALEX allows for complex habitat spatial structure. Each patch has a unique location and the patches may differ in quality. Patches can be connected by corridors that facilitate movement. The annual cycle of events modelled in ALEX is shown in Fig. 1 (Possingham and Davies,1995).

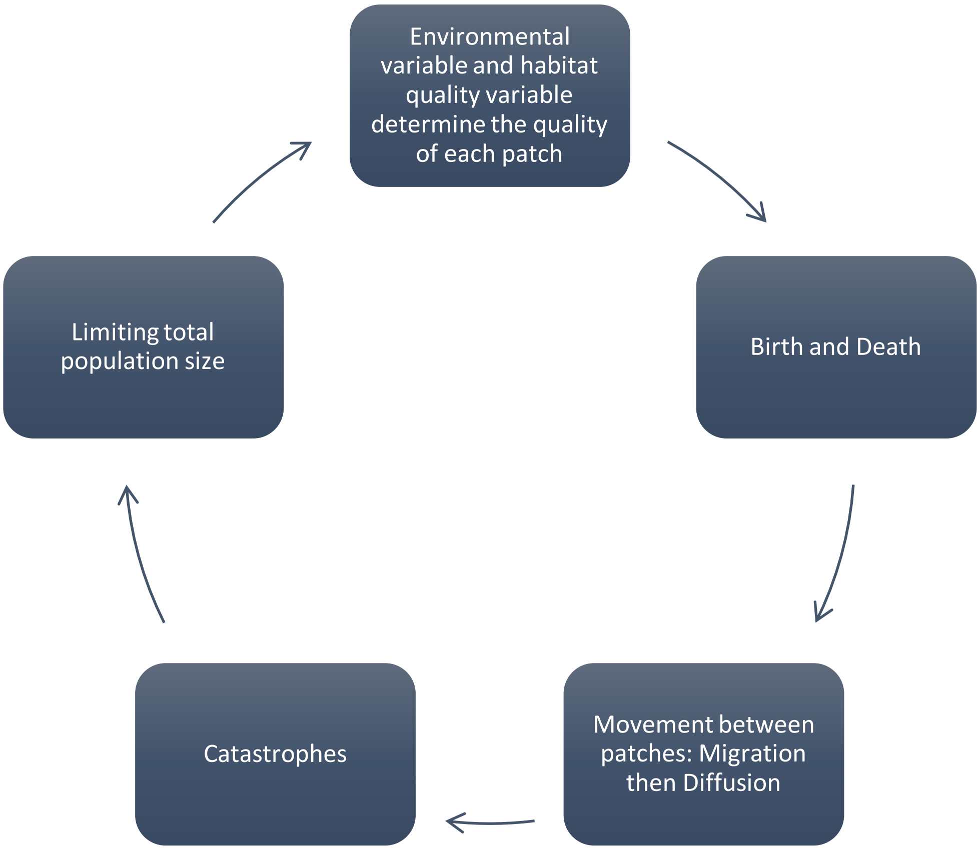


Fig. 1 Flow diagram showing the annual cycle of events simulated in ALEX. This structure simulates the dynamics of animal populations which experience an annual cycle of events.

RAMAS

RAMAS/GIS is designed to cope with such spatial complexity by interfacing population structure with maps imported from a geographic information system (GIS) so that spatial structure can be identified. RAMAS/GIS is exceptional “user-friendly”, with a nice loose-leafed manual and help screens to go with each function. It provides options for a variety of useful graphical illustrations of results (Boyce and Mark, 1996).

RAMAS/metapop is a new software modelling tool intended to be used by conservation biologists and wildlife managers in estimating a species extinction risk, designing reserves and assessing the potential impact of changes in habitat size and distribution. It extends the scope of the previous software package by allowing the user to include a species age structure as well as variations in vital rates and carrying capacity into their model. RAMAS/metapop is a potentially valuable tool for environmental decision makers who must project trends of populations over fragmented landscapes (Witteman et al., 1995).

RAMAS/landscape, predictions of the landscape model LANDIS are used as input maps for the metapopulation model RAMAS/GIS. The program combines landscape predictions, information about the habitat requirements of the species, and demographic data on its population dynamics into a metapopulation model, which has dynamic spatial structure simulating the changes in the landscape. This metapopulation model is then run to simulate future changes in the abundance of the species and its distribution in the landscape, to estimate the risk of extinction or decline, time to extinction and other measures of threat and viability (online http://www.ramas.com/landscape).

4. Materials and methods

We had no need to write instructions for using VORTEX since there is already exists a manual, which contains all information about input/output data, functions description etc., some of this information you will find in the following text below. VORTEX manual can be found on the official website (http://www.vortex10.org/Vortex10.aspx).

4.1 Study species

Taxonomic Notes

Rana dalmatina (the agile frog) belongs to the group of brown frogs. It is very similar in morphology to another long-legged frog from the former Soviet Union, the Caucasian *Rana macrocnemis*. In the past, they were considered as the same species. Beyond the breeding period, *R. dalmatina* morphologically resembles long-legged individuals of *R. arvalis*, especially *R. arvalis wolterstorffi*, with which it is syntonic in Transcarpathia (Kuzmin, 1999).

It is listed on Appendix II the Bern Convention and on Appendix IV of the EU Habitats Directive. This species is protected by national legislation in many countries and is recorded in several national and sub-national Red Data books and lists (The IUCN Red List of Threatened Species, 2009). In the Czech Republic, according to the legislation № 395/1992 Sb., the agile frog is highly endangered species (Nature Conservation Agency of the Czech Republic). According to the Red List (since 2018) it is vulnerable species (Jeřábková et al., 2017).

Morphology

Agile frog is medium to large, slender brown with long hind legs. Snout relatively long and sharp. Eardrum about the size of the eye is small. Hind feet partially webbed. When the hind leg is stretched forwards along the body, the heel reaches beyond the snout. Generally uniform light to dark brown above, sometimes reddish. Hindlimbs barred. Underside creamy white (Beukema, 2016).

Adults	Tadpole
- SVL-36-74 mm; - body slender; - snout sharp; - no male resonators; - legs very long; - shin shorter than body by 1.46-1.86 times.	The newly metamorphosed tadpole: - SVL measures 15-20 mm; - just after hatching it measures TL 12 mm; - prior to metamorphosis it measures TL 52-60 mm; - tadpole tooth formula is 1:2+2/1+1:3; - the clutch contains 600-1400 eggs deposited in small clumps of 15-27, rarely 50 or more, eggs;

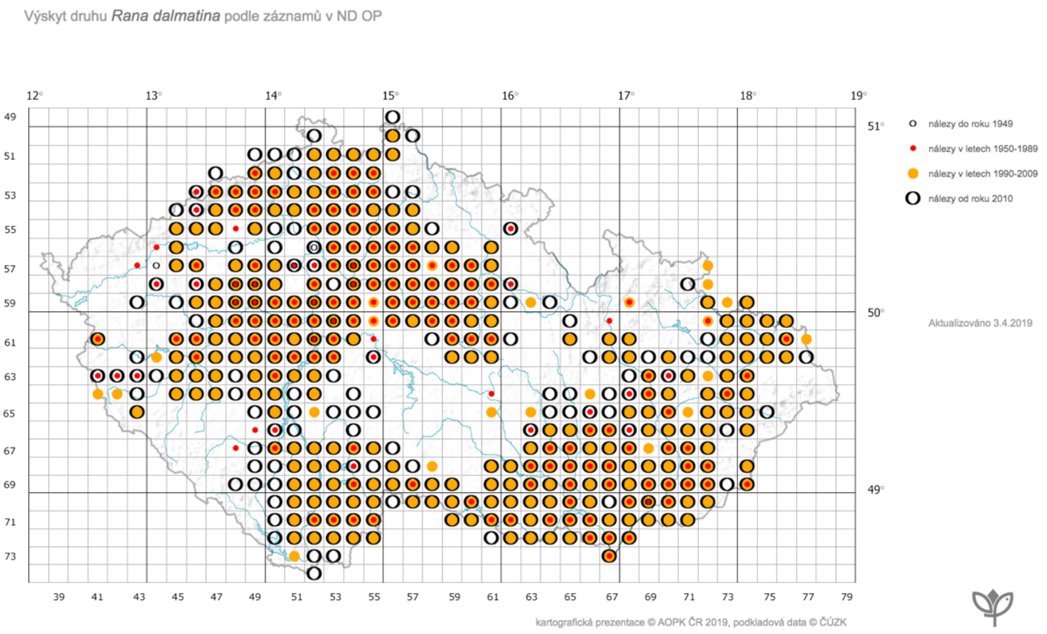
Table 1: Morphological parameters of *R. dalmatina* (Kuzmin, 1999)

Distribution

R. dalmatina inhabits Western and Central Europe including Drenthe, the Netherlands, Italian Peninsula and Turkey (Beukema, 2016). In the former Soviet Union, it is known with certainty only in Ukraine on the Transcarpathian Plain and from adjacent foothills and mountain slopes. *R. dalmatina* resembles morphologically the long-legged individuals of *R. arvalis*. This has led to erroneous indications of the presence of this species far away from its actual range, in Byelorussia, Ukraine, Moldavia (Kodry area) and Orel Province of Russia. In particular, this caused reports of the wider distribution of *R. dalmatina* in the Carpathians (Kuzmin, 1999).

In the Czech Republic, the agile frog is situated on the northern border.

This species occurs here mainly in warmer areas at lower altitudes (Bio monitoring ČR).



Pic.1 map of *Rana dalmatina* expansion in the Czech Republic (Obojživelníci České republiky).

Habitats and Abundance

The *R. dalmatina* is comparatively stenotopic, significantly more thermophilic, as evidenced by its geographical distribution, capable of living even on drier from the waters of more distant habitats (Oliva and Barush, 1992). Deciduous forests, generally humid and herbaceous. Found either within the forests or in meadows adjacent to them. Various waterbodies are used in early spring for breeding, such as ditches, ponds, submerged meadows or woodlands, occasionally also in slow-flowing rivers (Beukema, 2016). It is found in glades and open sites within light deciduous woodland (oak, beech, hornbeam etc.), and less frequent in meadows and thickets. It generally it does not occur in the pasture, arable areas or coniferous forests (The IUCN Red List of Threatened Species, 2009).

Reproduction

Males have a first toe mating callus, small mating calluses are also developed between the second and third, possibly the third and fourth fingers. At the time of breeding, when the secondary sex traits are most developed, the mating calluses have a grey-yellow or whitish colour and are lighter than the highly pigmented to black mating calluses of the other two species of our brown jumpers. Males are smaller in size than females. The largest of the males is 6-7 cm. (Oliva and Barush, 1992). Breeding starts at the end of March-beginning of April. Breeding individuals do not form large aggregations. The male call is weak, resembling a hen's cackle. Eggs are deposited at night on underwater vegetation in the most open parts of the pool. Frogs stay in the water for a short time (Kuzmin, 1999).

Population dynamics in the study area

The total number of *R. dalmatina* clumps varied more than 10 times during the monitored period, however, it showed no trend (annual average clutches = 765, min =
155, max = 1603). A slight decrease prevailed overgrowth (60% vs. 40% population change between years) on a larger scale, which was reflected in the comparison of clutches in individual years (1.07 – vs. 6.40 1.69 – 2.40 times) and appeared in a longer continuous period (max. 5 years vs. 2 years). The number of ponds occupied by clutches varied from 38 (28.4%) to 85 (63.4%) ponds over the years, but without any trend (Ščudlová Zuzana
et al.).

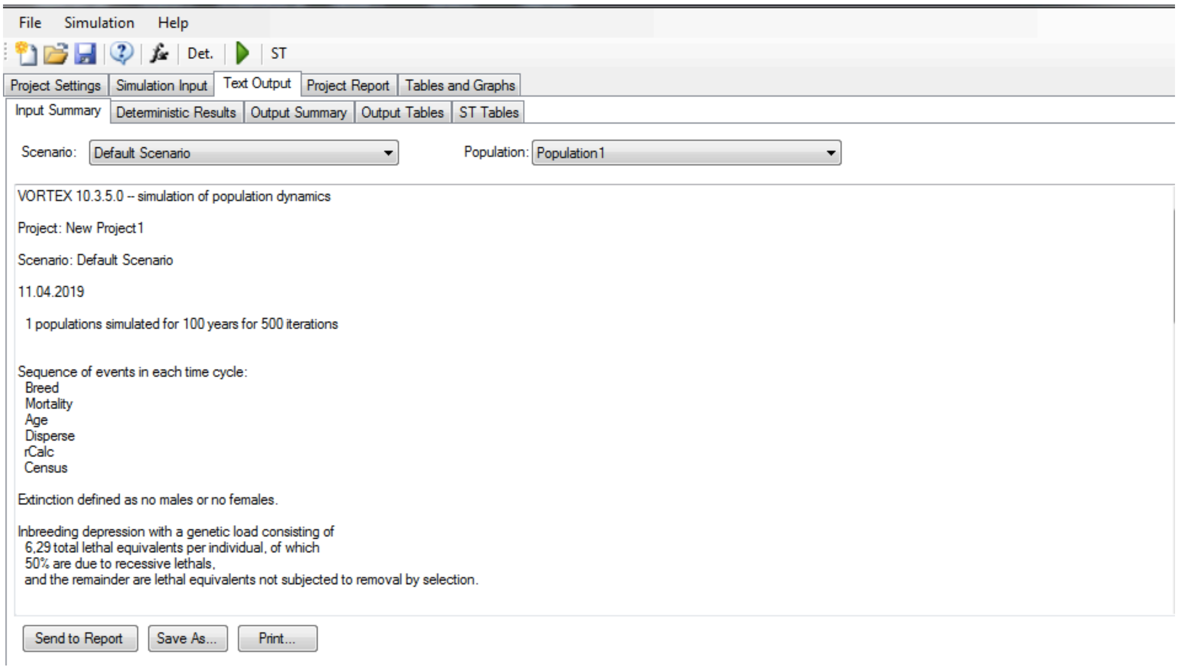
4.2 Statistical Analyses

VORTEX software (more inf. in chapter № 3.5.2) was used for running simulation of our data (you can find it in Appendix), which consists of *Scenario Settings, Species Description, Reproductive System, Reproductive Rates, Mortallity Rates, Catastrophes, Mate Monopolization, Initial Population Size* and *Carrying Capacity.* The description of our data you can find in tables (see Appendix). The detailed description is necessary for following comparison or character of entry data (their accuracy or completeness), which influences the currency of scenarios in VORTEX.

In our case we have used one type of local catastrophe – deep frost during the winter period: drying of about 10% of water surface, and affecting 10% of the population once every 10 years (see App. 1e). In the following chapter is shown the impact of this catastrophe on *Rana dalmatina* and the risk of its extinction.

Input Summary

The first tab shows a text file (saved with extension.inp in the project folder) that has all of the input values for each scenario.



Pic.2: Input Summary (VORTEX software)

We can scroll through the Input Summary to be sure that we entered all the input values correctly for our scenario, and that VORTEX interpreted the inputs as we intended.

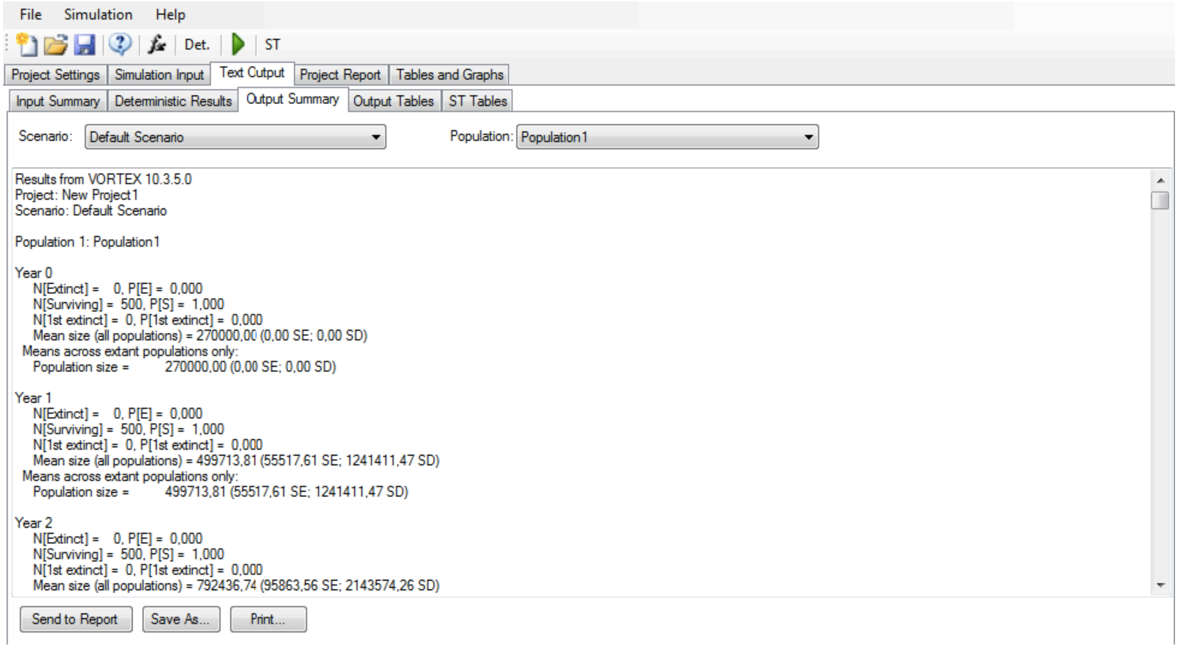
Output Summary

The third section of Text Output lists the basic status of each population at each year of the simulations. The statistics reported in this file (saved with extension .out), for Scenario and Population, are:

- The cumulative number of iterations in which the population is extinct or remains extant;
- The probability of population extinction (PE) or survival (equivalent to the proportion of iterations that the population is extinct or remains extant);
- The mean population size reported separately for all populations (N-all) and only for those remaining extant (N-extant), with standard error (SE) and standard deviation (SD) across iterations;
- The mean "expected heterozygosity" (or "gene diversity") remaining in the extant populations, with standard error and standard deviation across iterations;
- The mean "observed heterozygosity" (equal to 1 – [mean inbreeding coefficient]) remaining in the extant populations, with standard error and standard deviation across iterations;

- The mean number of alleles remaining within extant populations (from an original number equal to twice the number of founder individuals), with standard error and standard deviation;
- The mean number of mitochondrial haplotypes remaining within extant populations (from an original number equal to the number of founder individuals), with standard error and standard deviation;
- The final probability of population extinction and, the converse, the probability of population persistence;
- If at least 50% of the iterations went extinct, the median time to extinction;
- Of those iterations that suffer extinctions, the mean time to first population extinction, with SE and SD across iterations;
- The mean times to re-colonization and re-extinction of those simulations that went extinct;
- The mean final population size, with SE and SD across iterations, for all populations, including those that went extinct (e.g., had a final size of 0);
- The mean final population size for those iterations that do not become extinct, with SE and SD across iterations;
- The final age-sex composition of the extant populations;
- The mean population growth rate, r, with SE and SD across iterations.

The mean effective population size in extant populations, calculated from the loss of gene diversity from year 1 to the last year. Note that this Ne is the size of a randomly breeding population each generation (i.e., tallying only the adults, not the juveniles that constitute the next generation) across the generations. This will be approximately the harmonic mean of Ne at each generation and will be less than the arithmetic mean Ne across generations if the populations are growing, declining, or fluctuating in size over time.



Pic.3: Output Summary (VORTEX software)

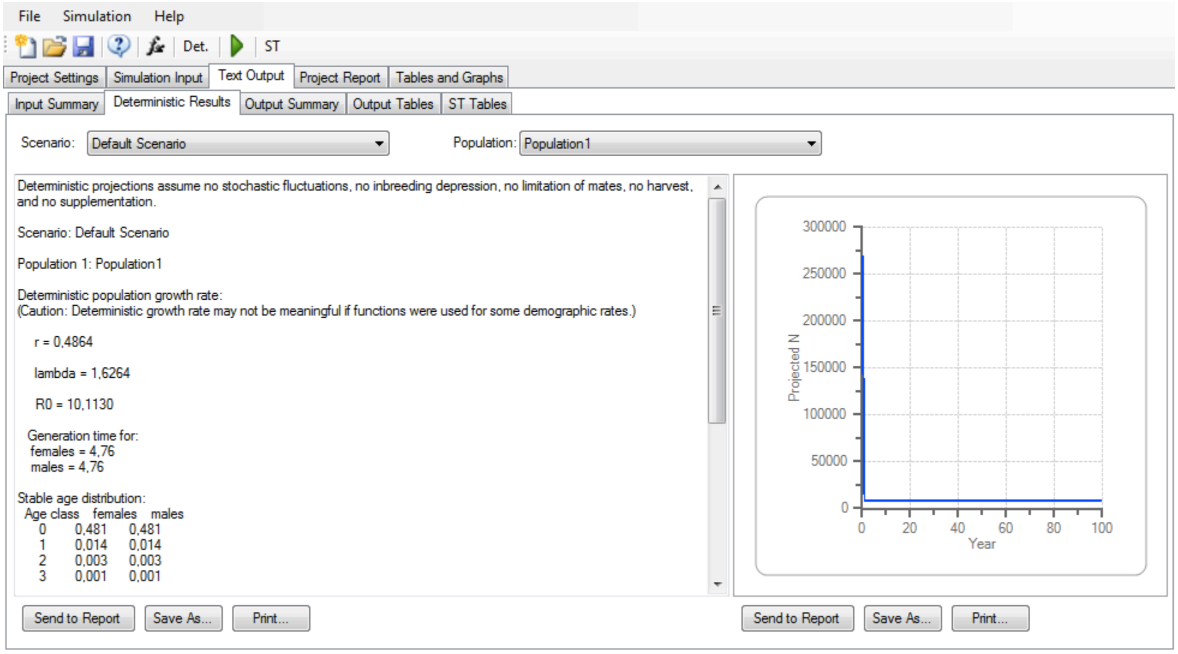
5. Results and Discussion

VORTEX was mainly used for mammals and birds, for amphibians much information has not been found. That is means, that we have some difficulties to find the appropriate answer on our “extinction” question of *Rana dalmatina* on a study area. Despite this, using of Vortex simulation on amphibians (*Hyla arborea*), was written in the article of Krug and Pröhl (2013), where simulation was used to predict the viability of a *Hyla arborea* population of about 70 adults inhabiting an isolated pond in the region of Hannover (Germany), by combining life history data with genotypic information derived from eight polymorphic microsatellite markers.

Scenario modelling can help us to understand the relative impacts of catastrophe and to develop management recommendations (Keedwell, 2014). The catastrophe (deep frost during the winter period – local catastrophe), which we have used for our research, hasn't a great effect on population persistence, so it doesn’t decline over a 100-year period. That result is described in the following description, which was taken from the VORTEX manual from its original web page (Lacy et al., 2017).

Deterministic Results
The second section of Text Output provides both text and a simple graph to display the deterministic projections of population size. The text window shows the exponential rate of:
- rate of increase (r): 0,4864;
- the annual rate of change (lambda): 1,6264;
- and the per generation rate of change or “net replacement rate” (R_0): 10,1130;
- the mean rates of reproduction and survival in your model (generation time): females = 4,76; males = 4,76;
- stable age distribution (calculated from age-specific birth and death rates): see on the picture below.

It is important to look at the deterministic projections of population growth for any analysis. If *r* is negative (we have a positive), the population is in deterministic decline (the number of deaths outpace the number of births) and will become extinct even in the absence of any stochastic fluctuations. The difference between the deterministic population growth rate and the growth rate resulting from the simulation can give an indication of the importance of stochastic factors as threats to population persistence.



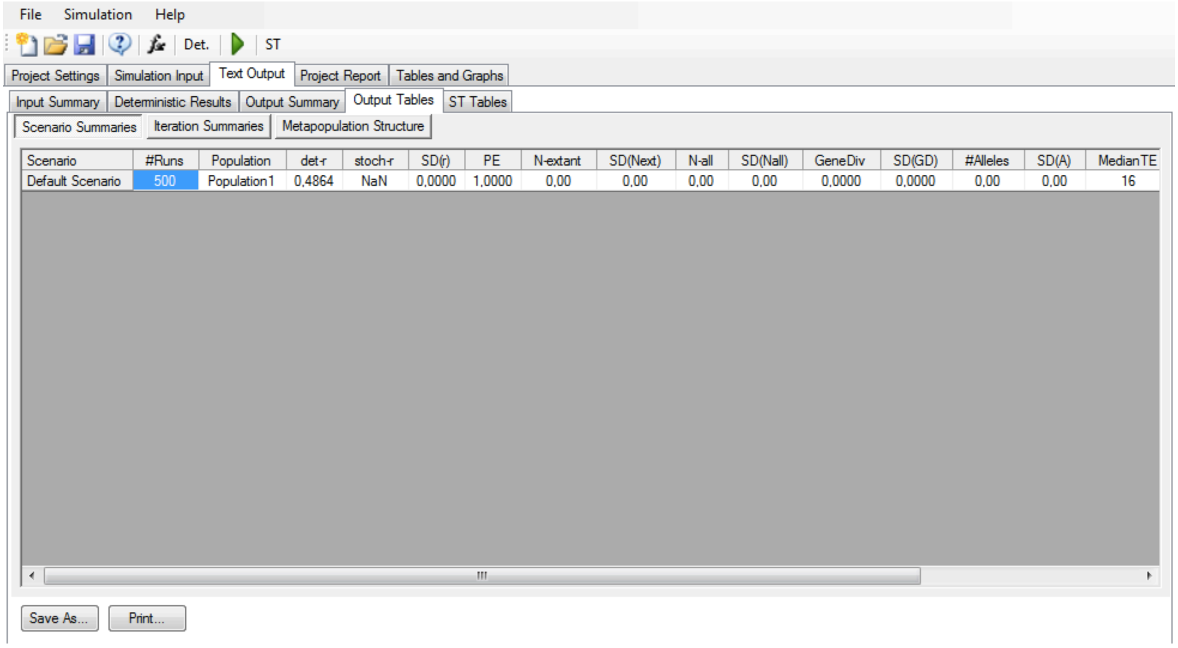
Pic.4: Deterministic Results (VORTEX software)

The graph given with the Deterministic Results is fairly simple and crude, but it shows the exponential growth (or decline) projected from the life table calculations (up to the limit set by the carrying capacity).

Output Tables

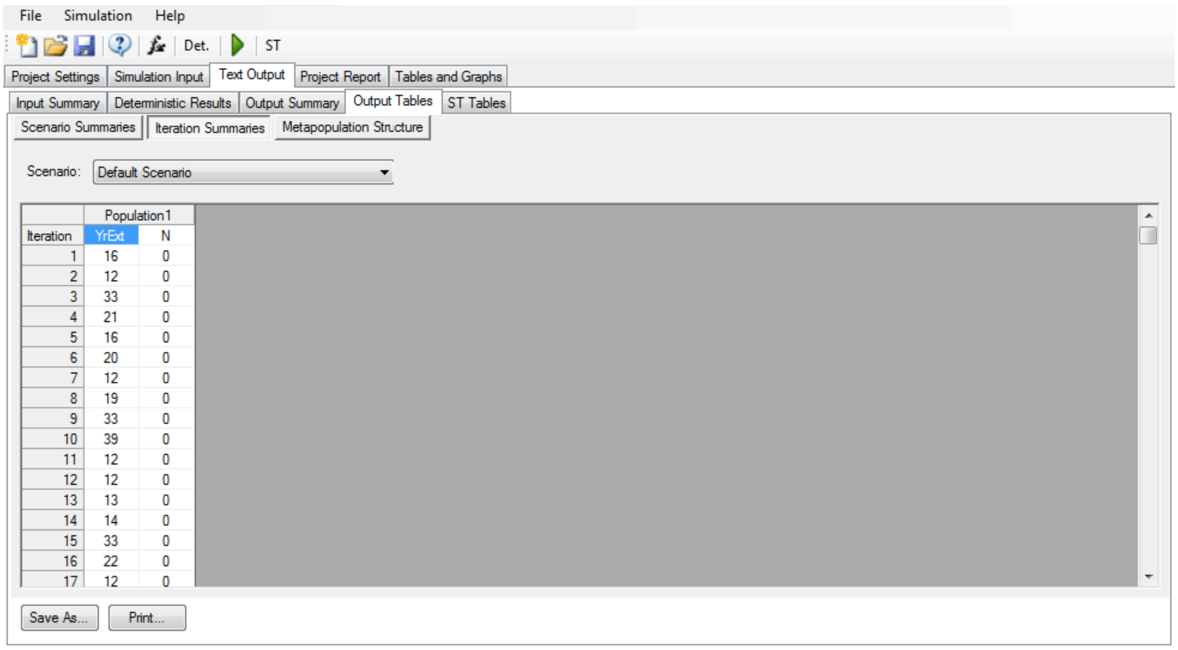
The fourth section of Text Output provides three kinds of summary tables:

the Scenario Summaries - provides a line of basic summary statistics for each Population of each Scenario that has been run. The summary statistics tabulated are the number of iterations (#Runs), the deterministic growth rate (det-r), the mean stochastic growth rate (stoch-r) experienced in the simulations, the SD of the stochastic population growth [SD(r)], and final values (at the end of the simulation) for many of the descriptive statistics listed above in the Output Summary. The Scenario Summaries tables also list the mean and SD at the end of the simulation for any Global State variables and Population State variables.



Pic.5: Output Tables (VORTEX software)

The Iteration Summaries in Output Tables provides a tabulation for each iteration of the year of extinction (if extinction occurs), final population size, and, if extinction does not occur, the final gene diversity, mean inbreeding coefficient, and a number of alleles remaining.



Pic.6: Output Tables (VORTEX software)

If more than one population is modelled, then the Metapopulation Structure in Output Tables provides in tabular format the various measures of population divergence that are included at the bottom of the Output Summary text. We have only one population.

6. Conclusions

Over the past decades, the abundance of amphibians has declined sharply. The natural fluctuations in population abundance are typical for amphibians, moreover, there are not many studies about populations due to the time-consuming. For understanding these fluctuations, we need long-term monitoring on a sufficiently large scale. PVAs give us an opportunity to carry out this monitoring and save the population. It has become commonly used tool in conservation biology and in the management of threatened or endangered species (Keedwell, 2014).

The first goal of this bachelor thesis was to give more information about software and show how we can use it in the future researches, also to show possibilities of PVAs and how to predict the extinction of the population. The second goal was to show an example of how VORTEX simulation can be used. The main problem of using VORTEX (and other PVAs) is a lack of information on studies species and how it has been used before. Software is mostly used abroad on some mammals and birds and only some works about using it on amphibians. Moreover, in the Czech Republic, on amphibians, it is not used at all. That is why, for monitoring, we chose a representative of this endangered population.

VORTEX is individual-based computer simulation program that incorporates environmental and demographic stochasticity, inbreeding depression, metapopulations, catastrophes, etc. In this bachelor paper, VORTEX was used for the monitoring of *Rana dalmatina* in Hornojiřetínská spoil ponds in North-west Bohemia in the Czech Republic. Our analyses showed that the future population size of *Rana dalmatina* on these ponds is in deterministic and doesn't decline over a 100-year period. This means, that catastrophe (deep frost during the winter period – local catastrophe) hasn't a great effect on population persistence. Besides that, no more catastrophes were tested, so we can't presume, that other catastrophes will not have an impact on the population of Agile frog. We also can't affirm, that our results are absolutely accurate. This paper only shows you how PVAs programmes may be used in scientific activities and the main functions of VORTEX simulation.

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8. Appendix

Appendix 1. Description of input data in scenario settings within software VORTEX and explanation of these settings, which were taken from the VORTEX manual (Lacy et al., 2017).

App. 1a. Scenario Settings

	Description	Data	Notes
Scenario Settings	*Number of Iterations*	500	How many times do we wish to repeat the simulation, given the data that we provide in the subsequent steps?
	Number of years	100	How far into the future do we wish to project our population?
	Duration of each year in days	365	*VORTEX* does not necessarily require "years" to be defined as calendar years. Rather, the program operates more broadly in terms of "time cycles".
	Population-based modelling	Population-based model	The simulation can be run as a population-based model, rather than as an individual-based model. In a population-based simulation, all genetic options and modelling (e.g., of inbreeding depression) are disabled, as is individual variation

			(demographic stochasticity).
	Extinction Definition	Only 1 sex remains	*VORTEX* gives us two methods to define “extinction” of our population. For most sexually reproducing species, ultimate biological extinction is assured whenever the population has declined to the point that it no longer has individuals of both sexes. In the first (and most common) choice, extinction is simply defined as the absence of at least one sex.
	Number of Populations	One population	*VORTEX* can model a single population or a complex *metapopulation* composed of any number of populations.
	Annual sequence	Breeding, mortality, ageing, dispersal, calc grow, census	The sequence of events in the annual cycle can be specified to be something other than the default (EV [setting annual rates]; Breeding; Mortality; Aging; Dispersal; Harvest; Supplementation; Calculate Growth; Carrying Capacity; Update GSVars,

			PSVars, and ISVars; Census).

App. 1b. Species Description

	Description	Data	Notes
Species Description	*Inbreeding depression*	No	We need to check this box if we want to include inbreeding depression in our model, as a reduction in first-year survival among inbred individuals.
	Lethal Equivalents	No	This box asks to specify the severity of inbreeding depression in our simulated population, if the population becomes inbred.
	Percent Due to Recessive Lethals	50	The percent of the total genetic load (quantified by the lethal equivalents we entered into the previous box) that is due to recessive lethal alleles.
	EV Correlation between reproduction and survival	0,8	Environmental variation (EV) is the annual variation in the probabilities of reproduction and survival that arise from random variation in

			environmental conditions.
	EV correlation among populations	1,0	We specify here the correlation of EV among populations (applicable, of course, only when more than one population is modelled).

App. 1c. Reproductive System

	Description	Data	Notes
Reproductive System	*Breeding system*	Polygamous	*VORTEX* models breeding systems as monogamous vs. polygamous vs. hermaphroditic, and short- term vs. long-term.
	Age of First Reproduction for Females (and Males)	Females – 3 years; Males – 3 years	*VORTEX* defines breeding as the time when the first offspring are born, not the age of onset of sexual maturity or the age of the first conception.
	Maximum Age of Reproduction	10	Maximum ages of breeding for males and for females can be set to be less than the maximum age. This allows for modelling of post-reproductive life spans.

	Maximum lifespan	12	*VORTEX* will kill any individual that reaches this age.
	Maximum Number of Broods per Year	1	*VORTEX* allows us to model more than one brood being produced by each female within each year.
	Maximum Number of Progeny per Brood	2500	Enter the most individuals born to a given female within a brood.
	Sex Ratio at Birth	50:50%	Enter here a number between 0.0 and 100.0 to represent the average percentage of newborn offspring that are male.
	Make offspring dependent on their dam for x years	No	With this option checked, a newborn is specified to be dependent on the dam until it becomes x years old.
	Density Dependent Reproduction	No	Does the reproductive rate of your species change with changing population size?

App. 1d. Reproductive Rates

	Description	Data	Notes
Reproductive Rates	*% Adult Females Breeding*	80%	Here we specify the mean percentage of adult females that breed in a given year (or, stated another way, the

			probability that a given adult female will successfully produce offspring in a given year).
	EV in % Breeding	10	Environmental variation (EV) in reproduction is modelled by the user entering a standard deviation (SD) for the percent females producing litters of offspring. *VORTEX* then determines the percent breeding for a given year by sampling from a binomial distribution with the specified mean and standard deviation.
	Distribution of broods per year	0 broods = 20 % 1 broods = 80 %	We can specify that each breeding female may have more than one brood (or clutch or litter) in each year.
	Use Normal distribution approximation/Specify exact distribution	Normal distribution Mean 1000 Sd 220	We must specify the percentage of litters/clutches/broods produced by the breeding adult females that are of a given size.

App. 1e. Catastrophes

	Description	Data	Notes
Catastrophes	*Number of Types of Catastrophes*	1	Catastrophes are extremes of environmental variation that strongly impact reproduction and/or survival.
	Catastrophe Label	Deep frost during the winter period – local catastrophe.	You first enter a label for each catastrophe, as a way to remind yourself and others what events you were modelling.
	Frequency and extent of occurrence	10%	Each catastrophe is specified to be local or global in scope (this is applicable only when more than one population is modelled).
	Frequency %	10%	Once the scope of the catastrophe is identified, we need to define the probability that a given catastrophe will occur in a particular year.
	Severity (proportion of normal values)	0, 5	For each catastrophe, we need to define the severity with respect to reproduction (percentage of adult females breeding) and survival.

App. 1f. Mortality Rates

	Description	Data	Notes
Mortality Rates	*Mortality of Females (Males) as %*	Mortality Age 0 to 1 – 95; Sd – 15; Mortality Age 1 to 2 – 65; Sd 1 to 2 – 10; Annual mortality after 2 – 55; Sd after 2 – 10.	In these tables, added the mean mortality rates for each age class, and enter also a standard deviation (SD) for each mean to describe the environmental variation (EV) in each rate.
	Delay 1st year mortality until all annual mortality is done (rather than in Breed)	No	Normally, *VORTEX* imposes the 1st year mortality immediately after a brood is produced. This avoids having the population grow to very large size during the Breed step when a species has high fecundity and high 1st year mortality.

App. 1g. Mate Monopolization, Initial Population Size

	Description	Data	Notes
Mate Monopolization	Males in the breeding pool from either the % siring offspring	80%	The Mate Monopolization input page directly prompts for the one measure of the degree of polygyny that is used directly by Vortex. Buttons provide access to simple pop-up utilities to calculate the % of males in the breeding pool from either the % siring offspring or the #mates / breeding male.

Initial Population Size	Use stable age distribution	Yes	If we choose "Use stable age distribution", then we will enter the initial N and *VORTEX* will allocate them to the age-sex classes according to the expected age distribution calculated from the birth and death rates.
	Use specified age distribution	No	If we choose, "Use specified age distribution", then you enter the number in each age-sex class and *VORTEX* totals these to fill in the Initial Population Size table.
	The initial age structure of the population	Size is 270000 at age of 3 years	The initial the age structure of the population can instead be specified by a proportional distribution with a total initial N. With this option, we need to enter both the total N and numbers for the age-sex classes.

App. 1h. Carrying Capacity

	Description	Data	Notes
Carrying Capacity	*Carrying Capacity (K)*	800	The carrying capacity describes the upper limit for the size of our simulated population within a given habitat.
	SD in K Due to EV	0	If you think that the habitat carrying capacity varies over

			time due to environmental variation (EV), we can enter a standard deviation (SD) here to account for this variability.
	Trend in K	No	*VORTEX* allows us to simulate changes in the carrying capacity. Such changes may be positive or negative and result from human activities such as resource utilization or corrective management strategies, or from intrinsic ecological processes such as forest succession.
	Implement K based	No	As a new option in *VORTEX* 10, K can be specified to be a criterion other than a limiting N. E.g., K can be a limiting number of females, or of adults, or of individuals with IS1=1, or of some function of variables.
	During K truncation, remove only individuals meeting criteria	No	Another option allows us to specify that only certain classes of individuals will be removed to bring the population back down to K.
	Prioritize K truncation based on ISvar	No	This option requires the user to create an ISvar that determines which

			individuals will be removed first.