Natural selection in compartmentalized environment with reshuffling

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Abstract

The emerging field of high-throughput compartmentalized in vitro evolution is a promising new approach to protein engineering. In these experiments, libraries of mutant genotypes are randomly distributed and expressed in microscopic compartments—droplets of an emulsion. The selection of desirable variants is performed according to the phenotype of each compartment. The random partitioning leads to a fraction of compartments receiving more than one genotype making the whole process a lab implementation of the group selection. From a practical point of view (where efficient selection is typically sought), it is important to know the impact of the increase in the mean occupancy of compartments on the selection efficiency. We carried out a theoretical investigation of this problem in the context of selection dynamics for an infinite non-mutating subdivided population that randomly colonizes an infinite number of patches (compartments) at each reproduction cycle. We derive here an update equation for any distribution of phenotypes and any value of the mean occupancy. Using this result, we demonstrate that, for the linear additive fitness, the best genotype is still selected regardless of the mean occupancy. Furthermore, the selection process is remarkably resilient to the presence of multiple genotypes per compartments, and slows down approximately inversely proportional to the mean occupancy at high values. We extend out results to more general expressions that cover nonadditive and non-linear fitnesses, as well non-Poissonian distribution among compartments. Our conclusions may also apply to natural genetic compartmentalized replicators, such as viruses or early trans-acting RNA replicators.

Keywords Directed evolution · Co-compartmentalization · Group selection · Frequency-dependent selection · Acellular genotype-phenotype linkage

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List of symbols

\( \mathbb{N} \)
We assume \( 0 \in \mathbb{N} \)

\( \mathbb{R}_+ \)
The nonnegative semiaxis: \( \mathbb{R}_+ = [0, +\infty) \subset \mathbb{R} \)

\( \mathcal{C}_c \)
Space of continuous functions with compact support

\( \mathcal{C}_{c+} \)
Space of nonnegative functions from \( \mathcal{C}_c \)

\( \mathcal{C}'_c \)
Space of generalized functions on \( \mathcal{C}_c \) (Radon measures)

\( \mathcal{C}'_{c+} \)
Subset of nonegative generalized functions

\( \mathbb{P} \)
Subset of probability densities: \( \mathbb{P} = \{ \rho \in \mathcal{C}_{c+} | \langle \rho, 1 \rangle = 1 \} \)

\( \mathbb{P}_p \)
Finite point-mass densities: \( \mathbb{P}_p = \{ \rho \in \mathbb{P} | \rho = \sum_{k=1}^{n} a_k \delta_{x_k} \} \)

\( \mathcal{I} \)
Some very large closed interval: \( \mathcal{I} = [0, \ell] \)

\( \mathbb{P}_\mathcal{I} \)
Densities in \( \mathcal{I} \): \( \mathbb{P}_\mathcal{I} = \{ \rho \in \mathbb{P} | \text{supp} \rho \subset \mathcal{I} \} \)

\( \mathbb{P}_p \mathcal{I} \)
Finite point-mass densities in \( \mathcal{I} \): \( \mathbb{P}_p \mathcal{I} = \mathbb{P}_p \cap \mathbb{P}_\mathcal{I} \)

\( \chi_A \)
Indicator function of the set \( A \): \( \chi_A(x) = \begin{cases} 1, & x \in A \\ 0, & x \notin A \end{cases} \)

\( C_n^k \)
Binomial coefficient \( \frac{n!}{k!(n-k)!} \)

\( \langle \rho, \varphi \rangle \)
The action of the generalized function \( \rho \) on the test function \( \varphi \)

\( \langle \rho, \varphi(x) \rangle \)
Implicitly \( \langle \rho(x), \varphi(x) \rangle \), where \( x \) is the internal variable

\( \langle \rho, \varphi(x, y) \rangle \)
Implicitly \( \langle \rho(y), \varphi(x, y) \rangle \), where \( y \) is internal and \( x \) is external

\( \langle \rho_x, \varphi(y) \rangle \)
Implicitly \( \langle \rho_x(y), \varphi(y) \rangle \), where \( y \) is the internal variable and \( x \) is a parameter of the distribution family \( \{ \rho_x \} \)

\( g(x) \)
a shortcut for \( (1 - e^{-x})/x \)

\( \delta_a \)
\( \delta \)-function concentrated at \( a \): \( \langle \delta_a, \varphi \rangle = \varphi(a) \)

\( \text{supp} \varphi \)
Support of the function \( \varphi \): the closure of \( \{ x \in \mathbb{R} | \varphi(x) \neq 0 \} \)

\( \text{supp} \rho \)
Support of the generalized function \( \rho \): \( \text{supp} \rho = \mathbb{R} \setminus O_\rho \), where \( O_\rho \) is the largest open subset \( O \subset \mathbb{R} \) such that \( \rho |_O = 0 \)

\( \bigotimes_k \rho_k \)
Tensor product \( \rho_1 \otimes \rho_2 \otimes \ldots \)

\( \rho^{\otimes n} \)
\( n \)-th tensorial power: \( \rho \otimes \rho \otimes \ldots \otimes \rho \)

\( \bigodot_k \rho_k \)
Convolution product \( \rho_1 * \rho_2 * \ldots \)

\( \rho^{*n} \)
\( n \)-th convolution power: \( \rho * \rho * \ldots * \rho \)

\( f_* \)
Pushforward of a generalized function by the map \( f \) of the domain: \( \langle f_* \rho, \varphi \rangle = \langle \rho, \varphi \circ f \rangle \)

\( \text{Corr}(\rho_1, \rho_2) \)
Cross-correlation of densities \( \rho_1 \) and \( \rho_2 \)

\( \rho \)
Probability density of the phenotypes (in the model description and application)

\( \sigma \)
Probability density of the fitness in a compartmentalized population

\( \sigma_x \)
Probability density of the fitness conditioned on phenotype \( x \)
Mean phenotypic trait: mathematical expectation of the function $x \mapsto x$ with respect to the phenotype distribution, $\langle \rho, x \rangle$ (in the model description and application)

The $n$-th moment of the phenotypic trait: mathematical expectation of the function $x \mapsto x^n$ with respect to the phenotype distribution, $\langle \rho, x^n \rangle$ (in the model description and application)

Mean fitness of an individual in a compartmentalized population: $\langle \sigma, x \rangle$ (in the model description and application)

Mean fitness of an individual with phenotype $x$ in a compartmentalized population: $\langle \sigma_x, y \rangle$ (in the model description and application)

Hyperbolic cosine of $x$: $\cosh x = (e^x + e^{-x})/2$

Poisson parameter: the mean number of individuals per compartment

Logical conjunction, implication, and negation, respectively

Introduction

In vitro directed evolution is a laboratory technique that mimics natural evolution and can be used to obtain proteins with new or improved properties (Packer and Liu 2015). Modern approaches are applied to the search of catalytic properties (i.e. artificial enzymes), for which they can test millions or billions of variants in parallel. These approaches use microcompartments, such as water-in-oil droplets in an emulsion, in which variants are randomly distributed, to enforce the phenotype-genotype linkage between the gene and its protein products. A selection pressure is then applied, at the level of compartments, in order to drive the population of enzymes towards the desired property. Cycles of mutation-selection are usually iterated until a satisfying variant is obtained.

The technique has been successfully used to improve existing enzymes (in relation to their direct catalytic characteristics, or to thermal stability, resistance to inhibitors, etc.), to change the substrate specificity of an enzyme, to develop a completely new enzymatic activity, or even new catalytic pathways (specific examples can be found in reviews by Martínez and Schwaneberg (2013), Wójcik et al. (2015), and Zeymer and Hilvert (2018)). Compartmentalized in vitro selection has also been used to study fundamental questions of protein physics such as the local shape of the protein fitness landscape on its sequence space with respect to a particular selection pressure (Romero et al. 2015; Zeymer and Hilvert 2018). Here the gene frequency change after a selection round for millions of mutants at once is used as a readout.

A typical high throughput directed evolution experiment starts with a large library of mutated genes of interest. Because enzymatic activity is carried by the encoded proteins, these genes require transcription and translation for phenotypic expression. This expression step is performed either by microbiological means (via recombinant expression of a gene-carrying plasmid in bacteria) or with direct in vitro approaches. In all cases, the mutant library is distributed among a large number of small compartments (e.g. microdroplets within an emulsion). The selection is carried out on each compartment, by evaluating its global -or apparent- phenotype and converting this
information into an artificial “fitness”. This critical step can be done in two different ways. The first one uses an external observation and subsequent physical separation of “good” and “bad” compartments, depending on the detected phenotype. In many cases, the phenotypic properties are converted into some fluorescent readout and then the compartments are sorted based on their spectral properties. An experiment of this kind, pioneering high-throughput emulsion sorting and in vitro compartmentalization, was performed by Tawfik and Griffiths (1998).

The second approach is based on an internal biochemical reaction that autonomously replicates the genotype in relation to some metrics of the phenotype (this situation is usually referred to as “self-selection”). This approach is well suited to the selection of DNA- and RNA-polymerases and was pioneered by Ghadessy et al. (2001) with the evolution of Taq polymerase towards higher resistance to PCR inhibitors. In that work, bacteria containing a plasmid with Taq gene and the corresponding Taq polymerase were randomly encapsulated in water-in-oil emulsion, with the addition of PCR primers targeting the Taq gene variant. After bacterial lysis released both plasmid and polymerase, droplets were submitted to thermal cycles and those with better polymerases produced more copies of the genetic variants they contained. At the end of the selection cycle the emulsion is broken and the genetic material is collected. If multiple selections rounds are used, the recovered genes serve as the initial library for the subsequent cycle. An idealized scheme of the self-selection process is depicted on Fig. 1.

Note that while the two selection processes described above correspond to very different experimental setups, from a modelling point of view, one can consider the former approach as a special case of the latter, where the fitness and the phenotype in a droplet are related via a step-function.

An essential feature of all such experiments is that selection does not act on individuals, but rather on groups of individuals which are randomly formed at each generation, during the compartmentalization process. When multiple individuals happen to share the same compartment, the selection outcome for a given genotype depends not only on its own identity, but also on identities of the others. This is because, inside each compartment, all genetic molecules are copied (or sorted) without distinction, but as a function of the combined phenotypic composition in the compartment. Therefore, as the statistics of the genotype distribution in compartments depends on the distribution of genotypes in the population, the selection effectively becomes frequency-dependent. In fact, these selection systems represent an extreme case of group selection model, where the reproductive success of an individual depends entirely on the combined phenotype of the group it belongs to Wilson (1973).

In the context of the search of new enzymes by high-throughput in vitro evolution the problem of random co-encapsulation is of great practical significance. The most efficient selection, in the sense of the selection pressure, is achieved in the situation when any compartment contains no more than one genotype. Such situation brings no dependence on frequency. However, unless sophisticated methods are used to enforce single occupancy (Edd et al. 2008; He et al. 2005), this implies a very high fraction of empty droplets, and thus a loss in throughput. A question naturally arises about the effect of allowing multiple genotypes in one droplet. Obviously, the selection pressure will drop, because if a stronger genotype $A$ and a weaker genotype $B$ meet in the same
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Fig. 1 The process of compartmentalized selection considered in the article. The initial population of size $N$ (here only two genotypes are shown, $\circ$ and $\bullet$) is randomly encapsulated inside $M$ compartments. Some compartments contain multiple individuals with possibly different genotypes. All individuals in a given compartment collectively contribute to the overall reproductive/replicative activity inside the compartment by pooling together their reproductive/replicative phenotypes. The content of the compartment then reproduces such that the total number of new individuals in the compartment depends on the collective phenotype via the selection function $f$ (not shown, see the model description for details). All individuals of the compartment equally contribute to this total progeny regardless of their genotype. Then the compartments are broken and a new generation is formed by sampling from the pool of the offspring. Note that a genotype of a weaker phenotype ($\circ$) has an opportunity to reproduce more efficiently if co-compartmentalized with a genotype of a stronger phenotype ($\bullet$). The article deals with the deterministic infinite population model, when both $N \to \infty$ and $M \to \infty$ with the conservation of the average number of individuals per compartment $\lambda = N/M$.

droplet, the shared replicative phenotype is weaker than that of two $A$-s and stronger than that of two $B$-s, while the number of copies is evenly shared. The precise response in terms of selection dynamics, and the relevant parameters, are however less clear. Lacking a general understanding of the process dynamics, researches are bound to empirical approaches to perform in vitro evolution experiments (Collins et al. 2015; Dodevski et al. 2015).

The goal of our work was to establish a general model of group selection suitable for studying in vitro evolution with random co-encapsulation. To be useful for interpreting experiments, the model must have a form of a dynamical equation that governs the temporal evolution of genotypic and phenotypic distributions in the library of mutants. The initial data and parameters must be: the initial genotypic or phenotypic distribution, the microscopic (on the level of a single group) rules of genotype interaction and its effect on the group survival and/or reproduction, and the statistics of the co-encapsulation. The model must eventually allow to study the effect of random co-encapsulation on the selection efficiency. This article takes a first step towards that goal, where we assume no selection inside the groups.

The existing general theoretical works on group selection are typically focused on two different aspects of this phenomenon. The first group of works studies the conditions on selection of individually disadvantageous but collectively advantageous traits
(like altruism). This direction of research has produced a very large body of literature. We refer to the works by Gardner and Grafen (2009) and Queller (1992) and the links therein. The main instrument here is Price’s covariation formalism (Price 1970, 1972). However, as Price’s identity does not provide a dynamically complete equation, this approach is not appropriate for the question studied in this article. Furthermore, the construction of this identity is based on the knowledge of the individuals’ fitness, which itself requires the development of a theory for compartmentalized selection (van Veelen 2005). The second traditional direction is related to the selection of an altruistic trait, too, but the main goal is to find its dynamics (Smith 1964; Wilson 1975). These works assume groups of the same size, additive fitness effect with altruism cost and benefits. This approach does produce a dynamical equation for the selection process starting from the model of interactions. The key point of such models is the interplay between the inter- and intragroup selection. Although such models with no intragroup selection do approach closely to the problem of compartmentalized selection, they carry a number of limitations. The group size is fixed, the number of possible phenotypic values is finite, and the emphasis on elementary mathematics complicates the treatment of nonlinear genotype/phenotype relations. Finally, there is a recent attempt to build a very general dynamic equation of group selection in form of PDE by Simon et al. (2013). The assumptions of the model, however, departure from the context of the compartmentalized experiments, too. They include a well defined finite set of phenotypes and a continuous-time dynamics of group restructure, while in the experiments, the phenotypic distributions are often continuous and the groups are completely reformed at discrete times.

There is also an overlap of the problem of co-encapsulated group selection with the problem of evolution in patched/structured populations, first conceived by Wright (1931). More specifically, the problem in question is isomorphic to a certain limit of the metapopulation model with migrant-pool gene flow (Bürger 2014; Hamilton 2011). Here the role of abstract groups is played by ecological demes. This direction is currently experiencing a renewed theoretical interest with the development of strict mathematical models of exact or approximate stochastic evolution dynamics with mutations based on Markov process formalism (for a modern treatment see, for example, Bitbol and Schwab 2014). However, typical simplifications of the models include few phenotypes (in most cases only two), weak migration, and an essential competition inside demes. Compartmentalized directed evolution experiments are, instead, characterized by a very large number of phenotypes, panmixing, and no intracompartment selection. Therefore, a new approach is needed to tackle the problem.

As the co-encapsulation of multiple genotypes under selection also arises in a number of biologically relevant scenario (e.g. multiple infection for viruses, parasites), it has also been the subject of a number of more specific previous work. Some recent examples include the works by Bianconi et al. (2013), Fontanari et al. (2006), Fontanari and Serva (2013), Lampert and Tlusty (2011), Matsumura et al. (2016), Zintzaras et al. (2010), as well as by Higgs and Lehman (2015) and links therein. Most of them, however, focus on aspects of kin/group selection, altruistic trait fixation, coexistence of selfish and cooperative genes, and the error catastrophe. Typically, these works treat more complicated cases of primordial evolution in presence of parasitic
sequences with different reproduction rates. These complex problems are explored primarily with numerical simulations, and are not directly applicable to the present problem of in vitro evolution. A model, very similar to the one that we use in the current work, was considered by Geritz et al. (1988, 1999) in the context of the seed size selection under the condition of a tradeoff with the seed number in annual plants existing as a subdivided population with the Poisson distribution in infinitely many growth sites. The main focus of these and related works was, however, the bivariate case (either two different species, or a resident species and a newly appeared mutant). The main question was either the species coexistence or the sympatric speciation (in the metapopulation). The influence of the mean occupancy on the selection process was not investigated. A preliminary analysis of the selection dynamics directly related to the context of in vitro co-compartmentalization was done by Zheng and Roberts (2007) for the case of two alleles, one of which is completely inactive, where any droplet with at least one active genotype inside is selected and all its content propagates to the next generation. A similar study of the effect of co-infection on selection from two viral phenotypes that share, when located in the same cell, both the replicative activity and the total offspring number was also carried out by Novella et al. (2004).

Here we present a more general description of the compartmentalized selection problem, that we initially motivate on a model related to in vitro evolution experiments in droplets, but which may apply also to biological situations. We consider an infinite population distributed in an infinite number of identical compartments, where the reproduction happens at discrete moments of time, the generations do not overlap, and the population is randomly redistributed at each cycle. We take into account only selection and ignore mutations and stochasticity. We derive, and solve for special cases, the update equation that defines the temporal evolution of the phenotypic distribution. Such approach explicitly incorporates the dependence of the selection pressure on the mean occupancy of droplets in the emulsion.

The structure of the article is the following. Section 1 gives the detailed description of the model. Section 2 deals with the general theory for an additive phenotype and a linear phenotype-fitness dependence. There we derive the probability distribution densities of a fitness experienced by a given phenotype in the emulsion for an arbitrary initial library. This allows to write the general update equation for the evolution of the library in course of selection. In Sect. 3 we study some general properties of solutions to Cauchy problems for the derived update equation. We also obtain exact solutions for some special cases. Sect. 4 deals with generalizations to nonlinear phenotype-fitness dependencies, including polynomial functions and sums of exponentials. We also demonstrate how, at least in principle, to deal with an arbitrary continuous phenotype-fitness dependence using already established results. In Sect. 5 we outline the framework to capture more general situations like a non-Poissonian distribution of individuals in the compartments, nonadditive phenotype, and multiple traits. In Sect. 6 we provide the results of numerical simulations to test some predictions from previous sections. Finally, we briefly discuss the result and their relevance to biological situations. Technical introduction and detailed mathematical proofs are given in appendices.
1 Model

We consider a population of haploid individuals that is subject to a group selection without any selection inside the groups. The population reproduces at discrete moments of time, different generations do not overlap, and the phenotype is strictly inherited. The groups model the compartments of the in vitro evolution experiments. We will refer to groups as compartments in the following.\(^1\) The number of compartments \(M\) and the population size \(N\) are assumed to be very large, so we consider the infinite population limit and all the stochasticity at the population level is ignored. Both numbers are the same in all generations. At each generation, the compartments are entirely reformed de novo and randomly repopulated from the pool of descendants produced by the previous generation. No intercompartment migration is assumed. Following the infinite population limit, we assume that the number of individuals in each compartment is given by the Poisson statistics with the Poisson parameter \(\lambda\), the average number of individuals per compartment (understood as a limit of \(N/M\)). Each individual produces as many descendants as any other member of the same compartment. The number of descendants of an individual can be called the \((local)\) fitness of the individual. It is defined by the total phenotype \(x_{\text{tot}}\) of the compartment in the following way. Firstly, we assume the total phenotype of the compartment to be additive. More specifically, if there are \(n\) individuals in the compartment with phenotypes \(x_1, x_2, \ldots, x_n\), then \(x_{\text{tot}} = x_1 + \cdots + x_n\). Secondly, each of the individuals produces \(f(x_{\text{tot}})/n\) descendants. Function \(f\) (we call it the selection function) bears all the specific information on the selection process by defining the overall replication/reproduction activity in compartments. The division by \(n\) manifests the sharing of that replication activity by all the members of the current compartment. In the simplest case of linear selection we have \(f(x_{\text{tot}}) = x_{\text{tot}}\) and the fitness of an individual is equal to the average phenotype in the compartment it belongs to. The phenotypic distribution of the descendants is obtained by averaging the result of the reproduction process over all compartments. We explicitly assume that the phenotypic distribution of the compartmentalized population at the next generation is equal to the phenotypic distribution of the descendants of the current generation. The corresponding lifecycle is schematically depicted on Fig. 1.

One essential assumption that we will always imply is that the phenotypic distribution is compactly supported and the phenotypic values are non-negative. This assumption is technical. However, it is well justified by the application domain. Indeed, we are interested in the phenotype (related to the replication activity) as a random variable. The replication activity is non-negative and has some physical upper bound (the reproduction rate cannot be infinite nor can be the rate of an enzymatic reaction).

\(^1\) The term compartment is more appropriate than the term group because we can freely talk about empty compartments. From the other hand, the related common terms patch or site from the terminology of models for geographically structured population have this advantage, too. These terms, however, usually imply (semantically but not necessarily as a property of the model) stable existence of patches (sites) in time. The model that we consider here can be seen as an extreme case of a structured population model, in which compartments play role of spatial patches/sites. As in our model, both essentially and by the implied interpretation (cyclic in vitro selection with compartmentalization), the patches are transient, we find the term compartment to be more suitable than the term patch or site.
It should be noted that, unlike the classical haploid selection, a nonlinear compartmentalized selection in general cannot be reduced to the linear selection by a simple nonlinear reparametrization of the phenotypic variable. The reason is that the phenotype additivity breaks with the reparametrization.

In the context of directed in vitro evolution of enzymes in emulsions, the individuals would correspond to gene carrying constructs or other genetic vectors, the compartments would correspond to the droplets of the emulsion, while the phenotype would correspond, for example, to the enzymatic activity. In the context of structured population in a patched environment, one may think of the pool of descendants as of the dispersion phase that, on a new cycle, randomly recolonizes the same set of patches. In this case the compartments correspond to the ecological patches.

The philosophy of the article is the following. We first use a heuristic approach to derive the mathematical formulation of the problem. Then we study implications of this formulation in the full mathematical rigor.

2 General theory of linear compartmentalized selection

In this section, we will consider only the linear selection function \( f(x) = x \). As it was written above, the linear selection corresponds to the case when the fitness of an individual is equal to the average phenotype in the compartment it belongs to. We will first develop a needed mathematical formalism. With this formalization, we will derive a general update formula that governs the dynamics of the phenotypic distribution. The update equation will be valid for any compactly supported phenotypic distribution, be it discrete, continuous, or more general than that. The equation will explicitly include \( \lambda \) as a parameter and thus will allow to directly investigate the dependence of the selection dynamics on the degree of the co-compartmentalization. The approach and the notions used in this section will be generalized and reused in the following sections to treat more complicated cases of nonlinear selection functions and non-Poissonian compartmentalizations.

We will treat random variables using the formalism of generalized functions. We will use Sobolev’s term generalized function for a linear continuous functional on an appropriate space of test functions instead of Schwartz’s term distribution to avoid confusion with the somewhat ambiguous term probability distributions. A random variable will be represented by its probability density, which in turn will be interpreted as a generalized function. This approach is equivalent to the standard treatment of random variables with the assumption of the weak convergence, where probability densities are interpreted as Radon measures. The emphasis is, however, put on the functional-theoretic aspect of the problem rather than on the measure-theoretic one. This point of view is more appropriate for the current analysis, as we are interested by computing average values of various functions rather than by finding probabilities of various events. Another advantage of the functional-theoretic approach is its intrinsic algebraicity, which strongly simplifies practical computations. See “Appendix A.1” for a brief introduction to the subject.
Any physically realistic phenotypic distribution in a real population has only finite number of different values $x$ of the phenotype and, therefore, is represented by a point-mass probability density with finite number of $\delta$-functions. General probability densities enter the picture as approximation of these point-mass generalized functions when the number of points (phenotype classes) becomes unmanageably large. Therefore, it is natural to derive the update equation first on the subset of point-mass probability densities with finite number of points and then to extend it to all probability densities by continuity. We will denote $P_p \subset P$ the subspace of such point-mass densities, where $P$ is the space of all probability densities. We want to derive an update equation in the form $\rho_{t+1} = A(\rho_t)$. To be able to extend the update operator $A$ to $P$ or to some its subset by continuity from $P_p$, we have to be sure that 1) $A$ is continuous on $P_p$ with the subset topology and 2) that $P_p$ is dense in $P$. Unfortunately, such direct approach does not work because the operator $A$ constructed in the following appears not to be continuous on $P_p$. However, an additional nonrestrictive assumption on the boundness of the possible phenotypic values removes this obstacle. The exact correct formulation of this assumption will be given in the end of this section.

Let us consider a random variable $\eta$ that describes the total phenotype of a compartment that contains $n$ individuals with phenotypes defined by $n$ random variables $\xi_1, \ldots, \xi_n$ with compactly supported densities $\rho_{\xi_k}$, so $\eta = \sum_k \xi_k$. The joint density of the random “vector” defined by $\xi_k$ is given by $\rho(\xi_1, \ldots, \xi_n) = \bigotimes_k \rho_{\xi_k}$. Using the standard simplification due to the compactness of the supports of $\rho_{\xi_k}$, for any $\varphi \in C_c$, by definition, we can express the action of $\rho(\xi_1, \ldots, \xi_n)$ on $\varphi(\sum_k x_k)$ via the convolution of the individual distributions

\[
\left. \bigotimes_k \rho_{\xi_k}, \varphi(x_1 + \cdots + x_n) \right] = \left. \bigotimes_k \rho_{\xi_k}, \varphi \right].
\] (1)

Therefore, $\rho_\eta = \bigotimes_k \rho_{\xi_k}$. In case when $\forall k \rho_{\xi_k} = \rho$, we have $\rho_\eta = \rho^n$.

In the same way, we can compute the density of a random variable that corresponds to a per-individual fitness in a compartment with $n$ such individuals. This fitness is given by $\zeta = \eta/n$. Its density can be computed noting that it is a pushforward of $\rho_\eta$ with respect to the map $h_n : \mathbb{R} \to \mathbb{R}$, $x \mapsto x/n$, so

\[
\rho_{\zeta} = (h_n)_* \rho_\eta = (h_n)_* \left( \bigotimes_k \rho_{\xi_k} \right).
\] (2)

Here one can understand $(h_n)_* \rho(x)$ as $n \rho(nx)$ with a slight abuse of notations.

In the following we assume that all $\rho_{\xi_k}$ are the same and are equal to $\rho \in P_p$, that is the phenotypic distribution in the population is characterized by the probability density $\rho$.

Let us find the density $\rho_{\xi_1 | k}$ of the per-individual fitness, given that the number of individuals in the compartment is $n$, and $k$ of $\xi_i$ assumed the value $x$ such that $\langle \rho, \chi_{\{x\}} \rangle = p > 0$, while the rest $n - k$ variables assumed any value different from $x$. This is equivalent to say, that first $k$ individuals are independently drawn from the
distribution given by $\delta_x$, and the rest $n - k$ individuals are independently drawn from the distribution given by

$$
\rho_{-x} \overset{\text{def}}{=} \frac{1}{1 - p} (\rho - p \delta_x).
$$

We can immediately conclude that

$$
\rho_{\eta|k} = \delta_x^k \rho_{-x}^{n-k},
$$

and then, as a consequence, the per-individual fitness has the probability density

$$
\rho_{\zeta|k} = \left(n \delta_x^k \rho_{-x}^{n-k}\right)(nx) = (h_n)_* \left(\delta_x^k \rho_{-x}^{n-k}\right) \overset{\text{def}}{=} \sigma^x_{nk}, \quad \text{where } h_n : x \mapsto x/n
$$

The next step is to find the density of fitness distribution of individuals with a given phenotype $x$ in the whole compartmentalized population, given that the phenotype of the initial library has density $\rho \in \mathbb{P}_p$. The density that we want to find describes the distribution of a local fitness (in a compartment) of an individual randomly chosen from all individuals with phenotype $x$. Let $P_n = e^{-\lambda n}/n!$ be the probability to find a compartment with $n$ individuals (assuming the Poisson distribution with the mean number of individuals per compartments $\lambda$). As phenotype $x$ is present in macroscopic quantities in the population, the probability to find a compartment with $n$ individuals, $k$ of which are with phenotype $x$, and the rest have other values (an $nk$-class compartment), randomly drawing it from all compartments is given by

$$
P_{nk} = e^{-\lambda n} \frac{C_n^k p^k (1 - p)^{n-k}}{n!} = P_n C_n^k p^k (1 - p)^{n-k}, \quad \text{where } p = \langle \rho, \chi_{\{x\}} \rangle,
$$

Each such compartment contains $k$ individuals with phenotype $x$, therefore the probability to find an individual that is encapsulated in an $nk$-class compartment randomly drawing it from the subpopulation of all individuals with phenotype $x$ is equal to

$$
P^x_{nk} = \frac{k P_{nk}}{\sum_{m,r} r P_{mr}}.
$$

The normalization constant in the denominator of (7) is equal to

$$
\sum_{n=1}^{\infty} \sum_{k=1}^{n} k P_{nk} = \sum_{n=1}^{\infty} \frac{e^{-\lambda n}}{n!} \sum_{k=1}^{n} k C_n^k p^k (1 - p)^{n-k} = \sum_{n=1}^{\infty} \frac{e^{-\lambda n}}{n!} n p = \lambda p.
$$

In each $nk$-class compartment, the per-individual fitness density of each individual is given by $\sigma^x_{nk}$ from (5), where $\rho_{-x}$ is defined by (3). Therefore, the fitness density $\sigma_x$
of an individual with phenotype $x$ randomly drawn from the whole compartmentalized population is given by

$$
\sigma_x = \sum_{n=1}^{\infty} \sum_{k=1}^{n} p_{nk}^x \sigma_{nk} = \sum_{n=1}^{\infty} \sum_{k=1}^{n} \frac{k P_{nk}}{\lambda p} \sigma_{nk}
$$

$$
= \sum_{n=1}^{\infty} \frac{e^{-\lambda} \lambda^{n-1}}{n!} \sum_{k=1}^{n} k C_n^k p^{k-1} (1-p)^{n-k} (h_n)_* \left( \delta_x^k * \rho_{-x}^{n-k} \right)
$$

$$
= \sum_{n=0}^{\infty} \frac{e^{-\lambda} \lambda^{n}}{n!} \sum_{k=0}^{n} C_n^k p^k (1-p)^{n-k} (h_{n+1})_* \left( \delta_x^{k+1} * \rho_{-x}^{n-k} \right),
$$

where again $h_n : x \mapsto x/n$.

To simplify this expression, we can use the linearity of $(h_i)_*$, the facts that

$$
\sum_{k=0}^{n} C_n^k \rho_1^k \rho_2^{n-k} = (\rho_1 + \rho_2)^n,
$$

that $a(\rho_1 * \rho_2) = (a \rho_1) * \rho_2 = \rho_1 * (a \rho_2)$, where $a$ is some number, and that $p \delta_x + (1-p) \rho_{-x} = \rho$. Finally, we obtain

$$
\sigma_x = \sum_{n=0}^{\infty} \frac{e^{-\lambda} \lambda^{n}}{n!} (h_{n+1})_* \left( \delta_x * \rho^{n+1} \right).
$$

This formula remarkably depends neither on $p$, the probability to encounter the phenotype $x$ in the initial library, nor on $\rho_{-x}$. It is well defined even in the limit $p = 0$. In fact, its structure resembles something expected for a continuous distribution, when the conditional probability to find another individual of phenotype $x$ in a compartment that already contains one is equal to 0.

In the same way we can compute the density of the fitness distribution for the whole population (without conditioning on the phenotype value):

$$
\sigma = \sum_{n=0}^{\infty} \frac{e^{-\lambda} \lambda^{n}}{n!} (h_{n+1})_* \rho^{n+1}.
$$

The fact that $\sigma_x$ and $\sigma$ are indeed generalized functions, that is, that the generalized functional series used to define them [both (9) and (11) for $\sigma_x$ and (12) for $\sigma$] converge to some generalized functions, can be easily established using three facts: 1) the series have the form $\sum_n a_n \rho_n$, where every $\rho_n$ is a compactly supported nonnegative generalized function that obeys $(\rho_n, 1) = 1$, 2) every $\varphi \in C$ can be majorated by a constant in the sense that $\exists B_\varphi \in \mathbb{R}^+ \forall x \mid \varphi(x) \mid \leq B_\varphi$, and 3) the series $\sum a_n$ converges absolutely. It is not difficult to see that these generalized functions are indeed probability densities with the support in the nonnegative semiaxis.
In the following, when the explicit dependence on time (understood as the population number) is required, we will denote it either as a subscript argument, like $\rho_t$, or with an argument in parentheses, like $\sigma_x(t)$. The latter means that we computed $\sigma_x$ in (11) using the value of $\rho$ at generation $t$ (thus, using $\rho_t$). The same meaning will be implied for $\sigma(t)$ and for various expectations. We will omit the time argument when it is not important and no confusion is possible.

As the population is assumed to be infinite, the coefficient in front of each term in $\rho = \sum_k p_k \delta x_k$ deterministically changes according to $p_k(t + 1) = p_k(t) \bar{\bar{w}}_{x_k}(t) / \bar{\bar{w}}(t)$ at any selection step, where $\bar{\bar{w}}_{x_k}(t) = \langle \sigma_{x_k}(t), x \rangle$ is the mean fitness of phenotypes $x_k$ at generation $t$ and $\bar{\bar{w}}(t) = \langle \sigma(t), x \rangle$ is the mean fitness of the whole population at generation $t$.

The mean fitness in the linear selection case is simply given by
\[
\bar{w} = \sum_{n=0}^{\infty} e^{\lambda n} \frac{n+1}{n!} \langle \rho, x \rangle = \langle \rho, x \rangle = \bar{x},
\] (13)
so the population average fitness is exactly equal to the population average phenotype. This property is a specific feature of the phenotype additivity with the linear selection function $f(x) = x$ and with sharing of the reproduction/replicative activity in a compartment among the individuals in it. Indeed, under this condition, the local fitness of any individual in any compartment is exactly equal to the average phenotype in that compartment, and thus the result of (13) is intuitively expected.

The mean fitness of phenotypes $x_k$ is also easily computed
\[
\bar{\bar{w}}_{x_k} = \sum_{n=0}^{\infty} e^{-\lambda n} \frac{x_k + n\bar{x}}{n+1} = \bar{x} + g(\lambda)(x_k - \bar{x}).
\] (14)
Here the factor that depends on $\lambda$ is equal to
\[
g(\lambda) \overset{\text{def}}{=} \frac{1 - e^{-\lambda}}{\lambda}.
\] (15)
Function $g$ is monotonously decreasing with $g(0) = 1$ and $\lim_{\lambda \to \infty} g(\lambda) = 0$. It is asymptotic to $1/\lambda$ at $\lambda \to +\infty$ (see Fig. 2).

Note that the selection with the fitness given by (14) is frequency-dependent. The fitness of a phenotype does not depend only on properties of this phenotype but on properties of the whole population as total via the term with $\bar{x}$.

It follows that a given component $p_k \delta x_k$ of $\rho$ after one round of selection changes to
\[
p_k \delta x_k \mapsto \left( 1 - g(\lambda) + g(\lambda) \frac{x_k}{\bar{x}} \right) p_k \delta x_k = \left( 1 - g(\lambda) + g(\lambda) \frac{x_k}{\bar{x}} \right) p_k \delta x_k.
\] (16)
Fig. 2 The function $g(\lambda)$ and its asymptotic behaviour at large $\lambda$

The update equation for the phenotypic distribution $\rho$ is then written as

$$
\rho_{t+1} = \left(1 - g(\lambda) + g(\lambda) \frac{x}{x_t} \right) \rho_t,
$$

(17)

Where $x$ is the phenotypic value. Note that the influence of the Poisson compartmentalization comes only through the factor $g(\lambda)$ given by (15).

We can rewrite this update rule as $\rho_{t+1} = A(\rho_t)$. The update operator $A$ at the left-hand side is defined for any $\rho \in \mathbb{P}_p$ such that $\langle \rho, x \rangle \neq 0$. As was mentioned before, $A$ is not continuous even on $\mathbb{P}_p$ (see “Appendix A.2” Proposition 1). However, as we demonstrate in “Appendix A.2”, for any closed interval $I = [0, \mathcal{L}]$, this operator is continuous on the space of finite point-mass densities $\mathbb{P}^\mathcal{I}_p \{\delta_0\}$ with the support in $\mathcal{I}$ with the exclusion of the $\delta$-function concentrated at $x = 0$. We also demonstrate that $\mathbb{P}^\mathcal{I}_p$ is dense in $\mathbb{P}^\mathcal{I}$, the space of all probability densities with supports in $\mathcal{I}$. Therefore, $A$ can be extended by continuity to $\mathbb{P}^\mathcal{I}\{\delta_0\}$. Thus, any kind of general nonegatively and compactly supported probability density, that happens to well describe the library at hand, evolves according to (17). The operators that generate $\sigma_x$ in (11) and $\sigma$ in (12) are well defined and continuous on the whole $\mathbb{P}_p$. As $\mathbb{P}^\mathcal{I}_p$ is dense in $\mathbb{P}$, they can be extended by continuity on all probability densities.

3 Selection trajectory

In this section we will assume only the linear selection. The phenotypic density $\rho$ will be however considered to be any compactly supported probability density with $\text{supp } \rho \subset \mathbb{R}_+$ and $\langle \rho, x \rangle \neq 0$.

Let the initial phenotypic distribution be given by $\rho_0$. We will consider its trajectory under the action of operator $A$ defined in the previous section. So, the update Eq. (17) is rewritten as $\rho_{t+1} = A(\rho_t)$ and we have $\rho_t = A^t(\rho_0)$.

Note that the action of $A$ on $\rho$ amounts to a multiplication of $\rho$ by the affine function with the slope $g(\lambda)/\langle \rho, x \rangle$ and with the intercept $1 - g(\lambda)$. Because of the dependence of parameters of this function on the current $\bar{x}_t$, (17) is not solvable in closed form for
The difference in the selection dynamics with \( \lambda > 0 \) and \( \lambda = 0 \). Phenotypic distribution densities \( \rho_{\tau} \) at \( \tau = t, \tau = t + 1, \) and \( \tau = t + 2 \) are shown in thick solid line, thick dashed line, and thick dash-dotted line, respectively. Thin inclined straight lines are graphs of functions \( 1 - g(\lambda) + g(\lambda) x / \bar{x}_{\tau} \), where \( \tau = t \) (solid line), \( t + 1 \) (dashed line), or \( t + 2 \) (dash-dotted line). Note that the affine functions by which the operator \( A \) multiplies the density at each step are different only by a rescaling for \( \lambda = 0 \). In the case of \( \lambda > 0 \), these functions are different both by a rescaling and by a shift. This difference between the cases is responsible for the possibility to solve the case \( \lambda = 0 \) in closed form and for the lack of such solution for \( \lambda > 0 \). The initial density is taken to be \( \rho_t = 2(1-x) \chi_{[0,1]} \). On the left panel, \( g(\lambda) = 0.6, \lambda \approx 1.15 \) a generic \( \rho_0 \) and \( \lambda > 0 \) (see Fig. 3). Nevertheless, we can study some properties of a generic trajectory.

### 3.1 General properties of the trajectories

We will prove two generic properties.

**Property 1** For any \( \rho_0 \) and any \( t \) we have \( \bar{x}_{t+1} - \bar{x}_t \geq 0 \). In other words, the average phenotype does not decrease.

**Proof** To prove the first property it is enough to apply \( \rho_{t+1} \) from (17) to \( x \), which gives

\[
\bar{x}_{t+1} = \bar{x}_t + g(\lambda) \frac{\bar{x}_t^2 - \bar{x}_t}{\bar{x}_t}. \tag{18}
\]

The nonnegativeness of \( \bar{x}_{t+1} - \bar{x}_t \) follows from the positiveness of \( \bar{x} \) and the nonnegativeness of the variance \( \bar{x}_t^2 - \bar{x}_t^2 \).

Formula (18) is reminiscent of Fisher’s fundamental theorem of natural selection in Price’s covariation form (Price 1970). Indeed, if only the individual’s phenotype mattered and was equal the individual’s fitness, Price’s formula for \( \Delta \bar{x}_t \) would be written as

\[
\Delta \bar{x}_t = \frac{\bar{x}_t^2 - \bar{x}_t^2}{\bar{x}_t}, \tag{19}
\]
which is different from (18) only by the factor $g(\lambda)$. Furthermore, any moment of the phenotypic distribution is updated according to

$$
\Delta \bar{x}_t^{m+1} = g(\lambda) \frac{x_t^{m+1} - \bar{x}_t x_t^m}{\bar{x}_t},
$$

which again transforms to Price’s covariation formula for a deterministic phenotype-fitness relation in the limit $\lambda \to 0$.

**Property 2** For any $\rho_0$ we have $\rho_t \to \delta_{x_0}$, as $t \to +\infty$, where $x_0 = \sup \supp \rho_0$. It means that the best mutant is always selected at infinite time.

To prove the second property we first will prove an intuitive lemma.

**Lemma 1** Let $\rho$ be a probability density with bounded support and the cardinality of $\supp \rho$ is greater than 1, then $x_i < \bar{x} < x_s$, where $x_i = \inf \rho$ and $x_s = \sup \supp \rho$.

**Proof** Indeed, as $\rho$ is nonnegative, it is monotone in the following sense: $\forall \varphi_1, \varphi_2 \in C_c \varphi_1 \geq \varphi_2 \Rightarrow \langle \rho, \varphi_1 \rangle \geq \langle \rho, \varphi_2 \rangle$. To show that $\bar{x} \in [x_i, x_s]$, let us take test functions $\varphi_i, \varphi_x, \varphi_s$ such that $\varphi_i|x_i, x_s] = x_i, \varphi_x|x_i, x_s] = x$, and $\varphi_s|x_i, x_s] = x_s$, and they are extended outside $[x_i, x_s]$ to respect $\varphi_i \leq \varphi_x \leq \varphi_s$, which is always possible. Then, by monotonicity and by $\langle \rho, 1 \rangle = 1$, we have $x_i \leq \langle \rho, x \rangle \leq x_s$.

Suppose that $\bar{x} = x_s$, so $\langle \rho, x \rangle = x_s$. Let $\varphi \in C_c$ be a test function with the support in $(-\infty, x_s)$. Then there always exist a positive $\alpha$ and a function $\varphi_\alpha \in C_c$ such that $\varphi_\alpha|x_i, x_s] = \alpha(x_s - x), \varphi_\alpha(x) = 0$ for $x > x_s$, and $-\varphi_\alpha \leq \varphi \leq \varphi_\alpha$. As $\langle \rho, \varphi_\alpha \rangle = 0$, it follows that $\langle \rho, \varphi \rangle = 0$, and therefore, by definition $\supp \rho = \{x_s\}$, which contradicts the premise. In the same way we conclude that if $\bar{x} = x_i$, then $\supp \rho = \{x_i\}$. This proves the lemma. □

**Proof of property 2** Solution of (17) at time $t$ can be written as

$$
\rho_t = \rho_0 \prod_{\tau=0}^{t-1} \left(1 - g(\lambda) + g(\lambda) \frac{x}{\bar{x}_t}\right) \overset{\text{def}}{=} \rho_0 \Pi_t.
$$

For any $t$, $\rho_t$ is obtained from $\rho_0$ as its product with a positive (except possibly at 0, where it is zero for the special case $\lambda = 0$) monotone continuous function $\Pi_t$. Therefore, $\supp \rho_t \subset \supp \rho_0$. More specifically, in most cases $\supp \rho_t = \supp \rho_0$. The only exception corresponds to $\lambda = 0$ and when $0 \in \supp \rho_0$ and it is not a limit point of the support. For this special case the following holds: $\forall t > 0$ supp $\rho_t = \supp \rho_0 \setminus \{0\}$. Therefore, $\forall t$ supp $\rho_t = x_0$.

The monotonously increasing bounded sequence $\{\bar{x}_t\}$ has the limit $\bar{x}_\infty = \lim_{t \to \infty} \bar{x}_t$ with $\bar{x}_\infty \leq x_0$. For any point $x_1$ such that $0 \leq x_1 < \bar{x}_\infty$, there exist $t_0$ such that for any $t \geq t_0$ we have $x_1/\bar{x}_t \leq x_1/\bar{x}_{t_0} < 1$. Let us denote $P_t^f(x) \overset{\text{def}}{=} (1 - g(\lambda) + g(\lambda)x/y)^t$. As $\Pi_t(x_1) \leq \Pi_{t_0}(x_1) P_{\bar{x}_t_0}^{t-t_0}(x_1)$ and $1 - g(\lambda) + g(\lambda)x_1/\bar{x}_{t_0} < 1$. 

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we have $P_{x_0}^{t-t_0}(x_1) \to 0$, and thus, $\Pi_t \to 0$ uniformly on $[0, x_1]$. It follows that $\rho_t|_{(-\infty,x_1)} \to 0$ with $t \to \infty$.

Suppose that $\bar{x}_\infty < x_0$. Then $\Pi_t \to \infty$ uniformly on any $(x_1, x_0) \subset [\bar{x}_\infty, x_0]$, and as $\text{supp} \rho_t \cap (x_1, x_0) \neq \emptyset$, at large enough $t$ the relation $\langle \rho_t, 1 \rangle = \langle \rho_0, \Pi_t \rangle = 1$ is violated. Therefore, $\bar{x}_\infty = x_0$ and $\rho_t \to \delta_{x_0}$.

3.2 Solution in general and explicit solution for $\lambda = 0$

For $\lambda > 0$, the solution of (17) is not representable with a simple closed formula. Nevertheless, it can be always computed by a simple recursion using (20) and (21).

We have $\rho_t = \Pi_t(x) \rho_0$, where $\Pi_0(x) = 1$, $\Pi_{t+1} = \left(1 - g(\lambda) + g(\lambda) \frac{x}{\bar{x}_t}\right)\Pi_t$, and

$\bar{x}_t$ is found by iterations $\bar{x}_m \to (1 - g(\lambda))\bar{x}_{m-1} + g(\lambda)\frac{\bar{x}_{m-1} + 1}{\bar{x}_{m-1}}$. As expected, $\Pi_t$ is a polynomial with coefficients that depend on all the moments of $\rho_0$ from $\bar{x}_0$ up to $\bar{x}_{t+1}$. The case $\lambda = 0$, which effectively corresponds to the case when the number of compartments is much larger than the population size, can be solved in closed form. The idea of the following simple approach to solving (17) for this case was taken from the work by Smerlak and Youssef (2017). At $\lambda = 0$, $g(\lambda) = 1$ and the update Eq. (17) takes the simple form

$$\rho_{t+1} = \frac{x}{\bar{x}_t} \rho_t.$$  

(22)

Note that, at every step, the density from the previous step is multiplied by $x$ and renormalized. Therefore, $\rho_t$ is proportional to $x^t \rho_0$ with some normalization constant, which is trivially reconstructed, and the solution is

$$\rho_t = \frac{x^t}{\bar{x}^t_0} \rho_0, \quad \bar{x}_t = \frac{x^{t+1}_0}{\bar{x}^t_0}.$$  

(23)

An interesting corollary of these formulas is the conclusion that the initial phenotypic probability density of the initial library can be reconstructed from the trajectory of $\bar{x}_t$ during the selection. Indeed, the following equality is a direct consequence of (23):

$$\bar{x}^m_0 = \prod_{i=0}^{m-1} \bar{x}_i.$$  

(24)

As the knowledge of all moments of $\rho_0$ allows to reconstruct the density itself, the phenotypic distribution in a library can be in principle obtained by tracking the mean population phenotype during selection process instead of direct measurement.

As an illustration, we will apply (23) to a library that contains only two classes of phenotypes, $x_1$ and $x_2$, and to a library that is described by a homogeneous distribution.
of phenotype on the interval \([x_1, x_2]\). In the former case, we have \(\rho_0 = p \delta_{x_1} + (1-p) \delta_{x_2}\) and, thus, if we denote \(\rho_t = p_t \delta_{x_1} + (1-p_t) \delta_{x_2}\), the solution is \(p_t = \frac{p x_1^t + (1-p) x_2^t}{p x_1^t + (1-p) x_2^t}\).

In the latter case, \(\rho_0 = \frac{1}{x_2 - x_1} \chi_{[x_1, x_2]}\) and, thus, \(\rho_t = \frac{x_t'(x_2^{t+1} - x_1^{t+1})}{(x_2 - x_1)^2 (t+1)} \chi_{[x_1, x_2]}\).

### 3.3 Exact solution for continuous time

Unlike (17), its continuous time counterpart can be solved exactly for any value of \(\lambda\). If we rewrite (17) as

\[
\rho_{t+1} - \rho_t = g(\lambda) \left( \frac{x}{x_t} - 1 \right) \rho_t, \tag{25}
\]

and assume that at each selection step the changes are small (which is true, for example, for \(\rho_0\) with small diameter of the support in comparison to \(\tilde{x}_0\), or for very large \(\lambda\)), then the selection dynamics can be approximated by

\[
\frac{d\rho_t}{dt} = g(\lambda) \left( \frac{x}{x_t} - 1 \right) \rho_t. \tag{26}
\]

Here \(\{\rho_t\}\) is understood as a \(C^1\) one-parameter family of generalized functions and \(d\rho_t/dt\) is understood as

\[
\frac{d\rho_t}{dt} = \lim_{\Delta t \to 0} \frac{\rho_{t+\Delta t} - \rho_t}{\Delta t} \tag{27}
\]

in the topology of \(C'_c\). We will prove the existence of solution of (26) by construction.

Let us first assume that a solution of (26) with given \(\rho_0\) exists for any compactly supported phenotypic probability density \(\rho_0\), \(\text{supp} \ \rho_0 \subset \mathbb{R}_+\). Then, for a given solution, we can assume \(\tilde{x}_t\) to be a given function of time that allows us to reparametrize \(t\) in (26) by the introduction a new independent variable \(\tau\) such that

\[
d\tau = g(\lambda) \frac{dt}{\tilde{x}_t}. \tag{28}\]

This transforms (26) to

\[
\frac{d\rho_{\tau}}{d\tau} = (x - \tilde{x}_\tau) \rho_{\tau}. \tag{29}\]

Let us introduce a new family of generalized functions \(n_{\tau}\) that solves the following Cauchy problem

\[
\frac{dn_{\tau}}{d\tau} = (x - 1)n_{\tau}, \quad n_0 = \rho_0. \tag{30}\]
One can look at $n$ as at the population density in the case when the normalization is not performed at the end of each selection cycle (the population is let to grow freely).

A solution of this problem corresponds to a solution of (29) by $\rho_\tau = \frac{n_\tau}{\langle n_\tau, 1 \rangle}$.

Indeed,

$$\frac{d \rho_\tau}{d \tau} = \frac{\dot{n}_\tau}{\langle n_\tau, 1 \rangle} - \frac{n_\tau \langle \dot{n}_\tau, 1 \rangle}{\langle n_\tau, 1 \rangle^2} = (x - 1) \rho_\tau - \left( \frac{\langle xn_\tau, 1 \rangle}{\langle n_\tau, 1 \rangle} - 1 \right) \rho_\tau = (x - \bar{x}_\tau) \rho_\tau,$$

(31)

where $\dot{n}_\tau = d n_\tau / d \tau$.

The solution of (30) exists, unique and is very easy to find. Namely, it is

$$n_\tau = e^{x \tau - \tau} \rho_0.$$

(32)

The fact of its uniqueness can be established by passing to a Laplace transform of (30) with respect to $x$, which gives a linear PDE with constant coefficients, uniqueness of solution of which can be conventionally established by the method of characteristics and by the uniqueness and smooth dependence theorems for ODEs. A relevant condition here is the compactness of $\text{supp} \, \rho_0$.

As $\langle n_\tau, 1 \rangle = \psi(\tau) e^{-\tau}$, where $\psi(y) = \langle \rho_0, e^{xy} \rangle$ is the so-called moment generating function of $\rho_0$ (it is its Laplace transform evaluated at $-\tau$), the corresponding solution of (29) is

$$\rho_\tau = \frac{e^{x \tau}}{\psi(\tau)} \rho_0.$$

(33)

It follows that $\bar{x}_\tau = \psi'(\tau) / \psi(\tau)$, and thus, taking into account (28), the corresponding implicit solution of (26) is given by

$$\rho_t = \frac{e^{x \tau}}{\psi(\tau)} \rho_0, \quad \bar{x}_t = \frac{\psi'(\tau)}{\psi(\tau)}, \quad t = \ln \frac{\psi(\tau)}{g(\lambda)}, \quad \psi(\tau) = \langle \rho_0, e^{x \tau} \rangle.$$

(34)

This solution can be rewritten in an explicit form

$$\rho_t = e^{x \psi^{-1}(e^{g(\lambda)t})-g(\lambda)t} \rho_0, \quad \bar{x}_t = e^{-g(\lambda)t} \psi'(\psi^{-1}(e^{g(\lambda)t})).$$

(35)

however, this explicit form is not practical, as for a generic case, the inverse of $\psi$ is impossible to compute explicitly. Even in a simple case of $\rho_0 = p \delta_{x_1} + (1 - p) \delta_{x_2}$, for which $\psi(\tau) = p e^{x_1 \tau} + (1 - p) e^{x_2 \tau}$, the inversion requires the solution of a transcendental functional equation.

The existence of a solution of (26) follows from the explicit construction. It is also unique. Indeed, suppose that there are two solutions of (26) $\rho^1_\tau$ and $\rho^2_\tau$ such that $\rho^1_0 = \rho^2_0 = \rho_0$. Then, with a solution dependent time rescaling, they both obey the same Eq. (29) with the same initial data but with possibly different $\tau$ (we denote them $\tau_1$ and $\tau_2$). The corresponding $n^i_\tau$ are uniquely constructed as $n^i_\tau = \psi(\tau_i) e^{-\tau_i} \rho_0$, and
it follows that the values of \( t \) that correspond to \( \tau_1 \) and \( \tau_2 \) such that \( \tau_1 = \tau_2 \) are the same. Finally, we conclude that \( \rho_1^0 = \rho_2^1 \).

The solution (33) of (29), along with the proof of its existence and uniqueness, was essentially obtained by different methods by Alfaro and Carles (2014) for a class of regular probability densities (absolutely continuous distributions) and by Martin and Roques (2016) for general densities. In particular, both works considered selection with mutations, while we left the question of mutations completely out of the scope. It is instructive to compare our explicit solution with the aforementioned results.

Alfaro and Carles found the exact solution for a more general “replicator-mutator” equation

\[
\partial_t u = (x - \bar{x})u + a^2 \partial_{xx} u, \quad \bar{x}(t) \overset{\text{def}}{=} \int x u(x, t) \, dx, \tag{36}
\]

where \( u(x, t) \) is the phenotypic distribution and the Laplacian represents an approximation of mutation process. It is shown in this work that the exact solution to (36) with the initial condition \( u(x, 0) = u_0(x) \) is given by

\[
u(x, t) = \frac{v(x, t)}{1 + \int_0^t \int \xi v(\xi, \tau) \, d\xi \, d\tau},
\]

where \( v \) solves \( \partial_t v = a^2 \partial_{xx} v + xv, \quad v(x, 0) = u_0(x) \). \tag{37}

The Eq. (29) with a regular \( \rho_0 \) corresponds to a degenerate case with \( a = 0 \). Then, \( v \) is simply given by \( v(x, t) = e^{xt} u_0(x) \) and \( u(x, t) = e^{xt} u_0(x) / e^{\tilde{x}t} u_0(\tilde{x}) \int d\xi, \) which is identical to (33).

Martin and Roques studied an approximate equation for the evolution of the cumulant generating function of the phenotypic distribution for a wide class of mutation processes

\[
\partial_t C_t(z) = \alpha(z) C_t'(z) - C_t'(0) + \beta(z), \tag{38}
\]

where \( C_t(z) \) is the expected cumulant distribution function under the deterministic approximation \( (C(z) = \ln \psi(z)) \), \( \alpha \) and \( \beta \) characterize the mutation process. This equation is equivalent to (29), if one assumes \( \alpha(z) = 1, \beta(z) = 0 \), and applies (29) on \( e^{xz} \) with the subsequent division by \( \psi_t(z) \) (note also that \( C_t'(0) = \psi_t'(0) = \tilde{x}_t \), as \( \psi_t(0) = 1 \)). It is demonstrated by Martin and Roques that the solution of (38) with the initial condition \( C_0 \) is given by

\[
C_t(z) = C_0 \left( y \left( y^{-1}(z) + t \right) \right) - C_0 \left( y(t) \right)
+ \int_0^t \left( \beta \left( y \left( y^{-1}(z) + s \right) \right) - \beta \left( y(s) \right) \right) \, ds, \tag{39}
\]

where \( y \) solves \( y'(z) = \alpha(y(z)), \quad y(0) = 0 \). In our case, \( y(z) = y^{-1}(z) = z \), and therefore \( C_t(z) = C_0(z + t) - C_0(t) \), or \( \psi_t(z) = \psi_0(z + t) / \psi_0(t) \), which is equivalent to (33).
4 Nonlinear phenotype-fitness dependence

In this section, we suppose that the local fitness of any individual inside a compartment with \( n \) individuals is given by

\[
h_n(x_1 + \cdots + x_n) = \frac{f(x_1 + \cdots + x_n)}{n},
\]

where the function \( f \) associates the total number of new individuals generated in the compartment with the total phenotype in the compartment, and \( x_i \) are phenotypes of the individuals in the compartment. In the following, we will call function \( f \) a selection function. The linear selection corresponds to the case \( f = \text{Id}_\mathbb{R} \).

If the dependence of the total in-compartment fitness on the total in-compartment phenotype is nonlinear, the update equation is still given by the formula

\[
\rho_{t+1} = \frac{\langle \sigma_x(t), y \rangle}{\langle \sigma(t), y \rangle} \rho_t,
\]

where \( \sigma_x \) and \( \sigma \) are given by (11) and (12) with the only difference that \( h_n \) is now taken to be \( h_n(x) = f(x)/n \) as in (40). Its analysis, however, becomes much harder. The main problem is the inability to compute the evolution operator \( A \) in closed form. Even in a case when each individual term in both \( \langle \sigma_x, y \rangle \) and \( \langle \sigma, y \rangle \) can be explicitly computed for any density \( \rho \), it is difficult (and impossible in general) to find the explicit sum of these whole series. Another complication comes from the observation that although every term in these series is a compactly supported density, the support of the series themselves may happen to be unbound due to the expansion of the support of \( (h_{n+1})*\delta x \ast \rho^*n \) and \( (h_{n+1})*\rho^*n+1 \) in case when \( h_n \) grows sufficiently fast. This may lead to the divergence of the expectation for the fitness in the compartmentalized population. It is possible to show, however, that if \( f \) is majorated by an exponential function \( a e^{bx} \) (or more generally, by \( a \chi bx \), which means that \( f \) does not grow faster than some exponential from both sides), then not only are \( \langle \sigma_x, y \rangle \) and \( \langle \sigma, x \rangle \) well defined finite numbers, but also the maps \( \rho \to \sigma_x \) and \( \rho \to \sigma \) are continuous on \( \mathbb{P} \) and the update operator \( A \) is continuous on the set of probability densities with the support in some arbitrary large closed interval \( I \) (see “Appendix A.3”).

If the selection function \( f \) is continuous, the update equation can be rewritten in different terms using the cross-correlation

\[
\rho_{t+1} = \frac{1}{N} \left( \sum_{n=0}^{\infty} \frac{e^{-\lambda} \lambda^n}{(n + 1)!} \text{Corr}(\rho_i^{*n}, f) \right) \rho_t,
\]

where \( N \) is the normalization constant and \( \text{Corr}(\rho_1, \rho_2) \) is the cross-correlation of densities \( \rho_1 \) and \( \rho_2 \). It is defined by its action on any function \( \varphi \) from \( C_c \):

\[
\langle \text{Corr}(\rho_1, \rho_2), \varphi \rangle = \langle \rho_1(x_1), (\rho_2(x_2), \varphi(x_2 - x_1)) \rangle.
\]
When one of $\rho_i$ is continuous, $\text{Corr}(\rho_1, \rho_2)$ is continuous, too. Therefore, (42) is defined correctly for a continuous $f$. The only potential problem of this expression is when $n = 0$. We can define $\rho^{*0} = \delta_0$, which can be understood rigorously. Then $\text{Corr}(\rho^{*0}, f)(x) = \text{Corr}(\delta_0, f)(x) = f(x)$.

If $\rho$ is also continuous, then one can use the classical formulas

$$\rho * \rho(x) = \int_{\mathbb{R}} \rho(y)\rho(x - y)\,dy, \quad \text{and} \quad \text{Corr}(\rho, f)(x) = \int_{\mathbb{R}} \rho(y)f(x + y)\,dy.$$  

We can also rewrite the update equation in a form that demonstrates its meaning more intuitively and transparently:

$$\rho_{t+1}(x) = \frac{1}{\mathcal{N}} \left( \sum_{n=0}^{\infty} \frac{e^{-\lambda \lambda_n}}{(n + 1)!}(f(x + x_1 + \cdots + x_n))_{x_1, \ldots, x_n} \right) \rho_t(x), \quad \text{(45)}$$

where all $x_i$ are independently drawn from $\rho_t$ and we used an averaging notation common to physical and statistical literature.

In the following, we will consider two special cases that make it possible to compute $\rho_{t+1}$ based on $\rho_t$ in closed form, namely when $f$ is a polynomial and when $f$ is a linear combination of exponential functions.

4.1 Polynomial $f$

Not only does this case allow to compute $\rho_{t+1}$ in closed form, but it also allows to derive the explicit expression for the update operator $A$, action of which is still a multiplication of $\rho_t$ by a polynomial function (coefficients of which nonlinearly depend on $\rho_t$).

Let $f(x) = a_0 + a_1x + \cdots + a_mx^m$, $a_i \in \mathbb{R}$. Then every $h_n$ is a polynomial, too. Therefore, to compute $\langle \sigma_x, y \rangle$ and $\langle \sigma, y \rangle$, it is enough to find $(s^k_n)_x \overset{\text{def}}{=} \langle \delta_x * \rho^{*n}, y^k \rangle$ and $s^k_n \overset{\text{def}}{=} \langle \rho^{*n+1}, y^k \rangle$ for any $k$, $0 \leq k \leq m$, and then to find the sums $\sum_n \frac{P_n}{n+1}(s^k_n)_x$ and $\sum_n \frac{P_n}{n+1} s_n^k$, where $P_n = e^{-\lambda \lambda_n}/n!$. Indeed, for any density $\nu$ we have

$$\langle (h_{n+1})_x v, x \rangle = \frac{1}{n + 1} \langle v, a_0 + a_1x + \cdots + a_mx^m \rangle = \frac{1}{n + 1} (a_0 + a_1 \langle v, x \rangle + \cdots + a_m \langle v, x^m \rangle). \quad \text{(46)}$$

The computation of the closed forms of $\langle \sigma_x, y \rangle$ and $\langle \sigma, y \rangle$, and therefore, of $A$, can in principle be performed in an algorithmic way (see “Appendix A.4”). As an example, we will treat the simplest nonlinear case $f : x \mapsto x^2$. There we have

$\text{Corr}(\rho, f)(x) = \int_{\mathbb{R}} \rho(y)f(x + y)\,dy$.}
Finally, the update equation takes the form
\[ \langle \delta_x * \rho^{*n}, y^2 \rangle = (y^2 \delta_x * \rho^{*n}, 1) = \langle (y^2 \delta_x) * \rho^{*n} + 2n(y\delta_x) * \rho^{*n-1} * (y\rho) + n\delta_x * \rho^{*n-1} * (y^2 \rho) + n(n-1)\delta_x * \rho^{*n-2} * (y\rho)^2, 1 \rangle = x^2 + 2nx\bar{x} + nx^2 + n(n-1)\bar{x}^2. \] (47)

Therefore
\[ \langle \sigma_x, y \rangle = g_0x^2 + g_1 \left( 2x\bar{x} + x^2 \right) + g_2\bar{x}^2, \] (48)
where \( g_i = e^{-\lambda_i} \frac{d}{d\lambda} \frac{e^\lambda - 1}{\lambda} \), so \( g_0 = g(\lambda) \) (see “Appendix A.4”). In the same way we find
\[ \langle \rho^{*n+1}, y^2 \rangle = (y^2 \rho^{*n+1}, 1) = \langle (n + 1)\rho^{*n} * (y^2 \rho) + (n + 1)n\rho^{*n-1} * (y\rho)^2, 1 \rangle = (n + 1)x^2 + (n + 1)n\bar{x}^2 \] (49)
and
\[ \langle \sigma, y \rangle = x^2 + \lambda\bar{x}^2. \] (50)

Finally, the update equation takes the form
\[ \rho_{t+1} = \frac{g_0x^2 + g_1 \left( 2x\bar{x}_t + x^2_t \right) + g_2\bar{x}_t^2}{\bar{x}_t^2 + \lambda\bar{x}_t^2} \rho_t. \] (51)

### 4.2 \( f \) as a linear combination of exponentials

Another simple case is when the selection function \( f \) is a linear combination of exponentials with constant coefficients, \( f(x) = \sum_{k=1}^{m} a_k e^{bkx} \). As in the case of a polynomial \( f \), it is enough to compute \( \langle \delta_x * \rho^{*n}, e^{by} \rangle \) and \( \langle \rho^{*n+1}, e^{by} \rangle \) for an arbitrary \( b \), which is, of course, a trivial problem. Indeed, \( \langle \delta_x * \rho^{*n}, e^{by} \rangle = e^{bx} \psi(b)^n \) and \( \langle \rho^{*n+1}, e^{by} \rangle = \psi(b)^{n+1} \), where, as before, \( \psi(s) \equiv \langle \rho, e^{sx} \rangle \) is the moment generating function. The relevant sums are easily computable, too:

\[ \sum_{n=0}^{\infty} \frac{e^{-\lambda} \lambda^n}{(n+1)!} e^{bx} \psi(b)^n = \frac{e^{bx} \psi(b) - 1}{\lambda \psi(b) e^{bx}}, \] (52)
\[ \sum_{n=0}^{\infty} \frac{e^{-\lambda} \lambda^n}{(n+1)!} \psi(b)^{n+1} = \frac{e^{bx} \psi(b) - 1}{\lambda e^{bx}}. \] (53)

Therefore, the update equation takes the form
\[ \rho_{t+1} = \frac{\sum_{k=1}^{m} a_k \frac{e^{bx} \psi(b_k) - 1}{\psi(b_k)} e^{bkx}}{\sum_{k=1}^{m} a_k \frac{e^{bx} \psi(b_k) - 1}{\psi(b_k)}} \rho_t. \] (54)
Interestingly, when \( f(x) = e^{bx} \), the selection dynamics does not depend on \( \lambda \), as in this case \( \rho_t = \frac{e^{bx}}{\psi_0(bt)} \rho_0 \).

Finally, the form of \( f \) that has both polynomials and linear combinations of exponential with constant coefficients as its special cases is a linear combination of exponentials with polynomial coefficients

\[
f(x) = \sum_{k=1}^{m} p_k(x)e^{bx_k}, \quad \text{where} \quad p_k(x) = \sum_{j=0}^{n_k} a_{jk}x^j.
\]  

(55)

Using a combinations of the approaches considered in the current and the preceding sections, one can obtain the update equation for this function in closed form (see “Appendix A.4”).

### 4.3 Any continuous exponentially majorated \( f \)

In principle, the action of \( \rho \) can be extended on complex-valued test functions with values in \( \mathbb{C} \) by a literal repetition of all the constructions outlined in “Appendix A.1”. This gives a hope to use the result of the previous section to approximate an arbitrary \( f \) by its truncated Fourier series on some large enough interval. As any such approximation is a sum of exponentials, the update equation for them can be written in closed form. In this case, all \( b_k \) (in the notation of the previous section) are imaginary and \( \psi(b_k t) = \varphi(ib_k t) \), where \( \varphi \) is the characteristic function of \( \rho \). However, one cannot, in general, select an interval once and then build approximations to any precision. The problem comes from the growing, in general, support of the growing convolution powers \( \rho^n \) in \( \sigma_x \).

We demonstrate in “Appendix A.5” that for any continuous exponentially bound \( f \) and for any compactly supported density \( \rho \) with \( \langle \sigma_x^f, y \rangle \neq 0 \) there is a sequence of (possibly ever growing) intervals and trigonometric approximations \( p_k \) of \( f \) on these intervals based on the Fourier series that generates \( \sigma_x^{p_k} \) and \( \sigma_y^{p_k} \) such that \( A^{p_k}(\rho) \rightarrow A^f(\rho) \), \( \sigma_x^{p_k} \rightarrow \sigma_x^f \), and \( \sigma_y^{p_k} \rightarrow \sigma_y^f \), where \( \sigma_x^f, \sigma_y^f, \) and \( A^f \) denote the corresponding \( \sigma_x, \sigma_y, \) and \( A \) generated by a selection function \( F \) and the density \( \rho \).

### 5 Generalization to a non-Poissonian distribution in compartments, to nonadditive phenotype, and to multi-trait phenotype cases

In this section, we do not aim for the proof of existence, continuity, etc of the relevant operators and operations or for the study of the generality of the results. We will just point out a possible generalization of the framework developed so far.
5.1 Non-Poissonian distribution

It is possible to generalize $\sigma_x$ and $\sigma$, and thus the update operator, to an arbitrary repartition of individuals in the compartments. A non-Poissonian distributions can arise, for example, if bacteria are used as an intermediate vehicle for a genome and its protein products before the compartmentalization. If the bacteria have a tendency to stick to each other it would in this case disturb the Poisson distribution of the bacteria in the compartments. In any case, this deviation is expressed in the fact that the probability to find a compartment with $n$ individuals $P_n$ is different from $e^{-\lambda} \lambda^n / n!$.

Following the same path as in Sect. 2, it can be demonstrated that for arbitrary $P_n$ we have

$$
\sigma_x = \frac{\sum_{n=1}^{\infty} n P_n(h_n) \ast (\delta_x \ast \rho^{*n-1})}{\sum_{n=1}^{\infty} n P_n}, \quad \sigma = \frac{\sum_{n=1}^{\infty} n P_n(h_n) \ast \rho^{*n}}{\sum_{n=1}^{\infty} n P_n}.
$$

(56)

Of course, it is enough to know only $\sigma_x$ and $\langle \sigma, x \rangle$ can always be computed as $\langle \langle \sigma_x, y \rangle, \rho \rangle, 1$. However, as it is seen from practical examples, a separate computation of $\sigma$ can be simpler. In addition, $\sigma$ has its own physical significance.

The update equation is, as before, $\rho_{t+1} = \frac{\langle \sigma_x(t), y \rangle}{\langle \sigma(t), y \rangle} \rho_t$. The cross-correlation form of it is represented by

$$
\rho_{t+1} = \frac{1}{\mathcal{N}} \left( \sum_{n=1}^{\infty} P_n \text{Corr}(\rho_t^{*n-1}, f) \right) \rho_t,
$$

(57)

where $\mathcal{N}$ is the normalization constant.

As before, we provide a form of this equation written in conventional notations of physical and applied statistical literature:

$$
\rho_{t+1}(x) = \frac{1}{\mathcal{N}} \left( \sum_{n=1}^{\infty} P_n \langle f(x + x_1 + \cdots + x_{n-1}) \rangle_{x_1, \ldots, x_{n-1}} \right) \rho_t(x),
$$

(58)

where all $x_i$ are independently drawn from $\rho_t$.

5.2 Nonadditive phenotype

The assumption of the additivity of the phenotype, although reasonable in some cases, is far from being universal. For example, if the fitness value of a compartment is defined by some enzymatic kinetics, say, Michaelis-Menten reaction, the effective kinetic parameters may not be additive. If the fitness is defined by the total enzymatic reaction rate (encoded by the genomes of individuals) under assumption of the excess of the substrate, then we have the additivity. However, if enzymes belonging to different phenotypes have different affinity to the substrate and the substrate is not in excess, the additivity is lost.
The most general way to describe a nonadditive phenotype is to declare what happens to the local fitness of individuals in a compartment when different number of individuals with different phenotypes are enclosed together. This implies a definition of a family of selection functions \( \{ f_n \} \), where \( f_n : \mathbb{R}^n \to \mathbb{R} \), so if a compartment contains \( n \) individuals with individual phenotypes \( x_1, \ldots, x_n \), then the per-individual fitness in the compartment is given by \( f_n(x_1, \ldots, x_n) \). When the local fitness of individuals is defined by the total phenotype of their compartment, functions \( f_n \) have the form of compositions \( f_n = h_n \circ f \circ c_n \) of the sharing function \( h_n : x \mapsto x/n \), of the selection function \( f : \mathbb{R} \to \mathbb{R} \) that maps the total phenotype in the compartment to the total number of offspring, and of the “combination” functions \( c_n : \mathbb{R}^n \to \mathbb{R} \) that define the total phenotype from the phenotypes of individuals. Even more general case, when \( f_n \) do not have the structure of the composition, corresponds to situations, when the very notion of the total phenotype is inapplicable.

All functions \( f_n \) are naturally symmetric in the sense that if we denote by \( \varpi \) a permutation of the set \( \{1, \ldots, n\} \), then

\[
\forall \varpi \quad f_n(x_{\varpi(1)}, \ldots, x_{\varpi(n)}) = f_n(x_1, \ldots, x_n). \tag{59}
\]

This symmetry comes from the fact that all individuals in a compartment are equivalent and it does not matter which one we consider to be the first one, which one to be the second one, and so on. If the compartments had some internal structure, or the total phenotype depended on the order of entry of individual (think of the order of infection of the same bacterium by multiple phages), then the symmetry would be lost.

Let us first consider \( \rho \in \mathbb{P}_p \). Then the probability density of the local fitness of individuals with phenotype \( x \) in an \( nk \)-class compartment, analogously to (5), is given by

\[
\sigma^x_{nk} = (f_n)_*(\delta^\otimes k \otimes \rho^\otimes n-k), \tag{60}
\]

with the same notations as in Sect. 2. Here \( (f_n)_* : C'_c(\mathbb{R}^n) \to C'_c(\mathbb{R}) \) is the pushforward generated by \( f_n \).

Note that for any \( \rho_1, \ldots, \rho_n \) and any permutation \( \varpi \) we have

\[
(f_n)_*(\rho_{\varpi(1)} \otimes \ldots \otimes \rho_{\varpi(n)}) = (f_n)_*(\rho_1 \otimes \ldots \otimes \rho_n). \tag{61}
\]

Indeed, for any \( \varphi \in C'_c(\mathbb{R}) \) we have

\[
((f_n)_*(\rho_1 \otimes \ldots \otimes \rho_n), \varphi) = \langle \rho_1(x_1) \otimes \ldots \otimes \rho_n(x_n), \varphi(f_n(x_1, \ldots, x_n)) \rangle \\
= \langle \rho_1(x_1), \ldots, \langle \rho_n(x_n), \varphi(f_n(x_{\varpi(1)}, \ldots, x_{\varpi(n)})) \rangle \rangle \\
= \langle \rho_{\varpi(1)}(x_{\varpi(1)}), \ldots, \langle \rho_{\varpi(n)}(x_{\varpi(n)}), \varphi(f_n(x_{\varpi(1)}, \ldots, x_{\varpi(n)})) \rangle \rangle \\
= \langle (f_n)_*(\rho_{\varpi(1)} \otimes \ldots \otimes \rho_{\varpi(n)}), \varphi \rangle \tag{62}
\]

Because of this and because \( (f_n)_* \) is a linear operator, the following binomial identity is valid
\[
\sum_{k=0}^{n} C_n^k p^k (1 - p)^{n-k} (f_n)_* \left( \delta_x^\otimes k \otimes \rho^\otimes n-k \right) = (f_n)_* \rho^\otimes n. \tag{63}
\]

Here we identify \( \rho^\otimes 0 \) with \( 1 \in \mathbb{R} \), \( \rho^\otimes 1 \) and \( 1 \otimes \rho \) with \( \rho \).

Following the same reasoning as in Sect. 2 and in Sect. 5.1, we can conclude that the fitness densities \( \sigma_x \) and \( \sigma \) are given by

\[
\sigma_x = \sum_{n=1}^{\infty} \frac{n P_n (f_n)_* (\delta_x \otimes \rho^\otimes n-1)}{\sum_{n=1}^{\infty} n P_n}, \quad \sigma = \frac{\sum_{n=1}^{\infty} n P_n (f_n)_* \rho^\otimes n}{\sum_{n=1}^{\infty} n P_n}, \tag{64}
\]

or, in the case of Poissonian \( P_n = e^{-\lambda} \lambda^n / n! \),

\[
\sigma_x = \sum_{n=0}^{\infty} \frac{e^{-\lambda} \lambda^n (f_{n+1})_* (\delta_x \otimes \rho^\otimes n)}{n!}, \quad \sigma = \sum_{n=0}^{\infty} \frac{e^{-\lambda} \lambda^n (f_{n+1})_* \rho^\otimes n+1}{n!}. \tag{65}
\]

These expressions can be extended by continuity to all \( \rho \in \mathbb{P} \). The proof of continuity is analogous to the proof of Theorem 3 in “Appendix A.2”. The only essential additional fact to be used is the continuity of the tensor product. The continuity of the update operator is a more delicate issue and we will not study it here.

As before, we provide a conventional intuitively transparent form of the corresponding update equation for a general \( P_n \) (all \( x_i \) are independently drawn from \( \rho_t \)):

\[
\rho_{t+1} (x) = \frac{1}{\mathcal{N}} \left( \sum_{n=1}^{\infty} n P_n (f_n (x, x_1, \ldots, x_{n-1}))_{x_1,\ldots,x_{n-1}} \right) \rho_t (x). \tag{66}
\]

### 5.3 Multiple traits

In the same way we can consider not only a single trait phenotype \( x \), but also multiple traits \( x_1, \ldots, x_m \). We can organize them in a tuple \( \xi = (x_1, \ldots, x_m) \in \mathbb{R}^m \) \( \text{def} = \mathbb{X} \). The distribution of the traits at the beginning of each selection cycle is now given by a generalized function from \( \mathbb{P}(\mathbb{X}) \subset C'_c(\mathbb{X}) \).

In the classical case, when no co-compartmentalization occurs, the selection process is ignorant about how exactly the fitness is defined by the underlying traits. Therefore, we could abstract from these traits once by using a pushforward of the trait distribution with the selection function. If the fitness is given by the selection function on the trait space \( f : \mathbb{X} \rightarrow \mathbb{R} \) and the initial trait distribution is given by \( \rho \in \mathbb{P}(\mathbb{X}) \), then the initial fitness distribution is given by \( \tilde{\rho} = f_* \rho \in \mathbb{P}(\mathbb{R}) \). As the resulting fitness distribution after \( t \) cycles of selection is given by the initial fitness distribution multiplied by some continuous function of the fitness values, namely by \( \tilde{\rho}_t = \frac{\chi^t}{(\tilde{\rho}, \chi^t)} \tilde{\rho} \) [see (23)], the final trait distribution could be reconstructed as the initial trait distribution multiplied by the pullback of this function by the selection function. Indeed, the pullback of any \( \varphi \in C_c(\mathbb{R}) \) to \( C_c(\mathbb{X}) \) by \( f \) is defined by \( f^* \varphi \) \( \text{def} = \varphi \circ f \in C_c(\mathbb{X}) \). Using the identity
\( \varphi(f \ast \rho) = f^*(f^* \varphi) \ast \rho \), which is easy to verify, one recovers \( \rho_t = \frac{f^* (\xi)^t}{\langle \rho, f(\xi)^t \rangle} \). This expression goes well with the intuition [see the derivation of (23)].

Unfortunately, the co-compartmentalization complicates this picture. As previously, we define the rules of how the phenotypic parameters (the traits) are related to the local fitness of individuals by a family of functions \( \{f_n\} \) on Cartesian powers of the trait space \( f_n : \mathbb{X}^n \to \mathbb{R} \). Functions \( f_n \) assign the per-individual fitness \( f_n(\xi_1, \ldots, \xi_n) \) to a compartment with \( n \) individuals with phenotypes \( \xi_1, \ldots, \xi_n \). In a case of a non-additive but still well defined total phenotypic traits in a compartment, each function \( f_n \) is in fact a composition \( f_n = h_n \circ f \circ c_n \) of the sharing functions \( h_n : x \mapsto x/n \), the selection function \( f : \mathbb{X} \to \mathbb{R} \) that operates on the combined phenotype of the compartment, and of the functions \( c_n : \mathbb{X}^n \to \mathbb{X} \) that define how individual phenotypic traits are combined when multiple individuals are mixed in one compartment. In a more general case \( f_n \) do not have this structure.

Analogously to the single trait case, we can start from \( \mathbb{P}_p(\mathbb{X}) \) and obtain the expressions

\[
\sigma_{\xi} = \frac{\sum_{n=1}^{\infty} n P_n(f_n)^* (\delta_{\xi} \otimes \rho^\otimes n-1)}{\sum_{n=1}^{\infty} n P_n}, \quad \sigma = \frac{\sum_{n=1}^{\infty} n P_n(f_n)^* \rho^\otimes n}{\sum_{n=1}^{\infty} n P_n}, \quad (67)
\]

or, in the case of Poissonian \( P_n = e^{-\lambda \lambda^n} / n! \),

\[
\sigma_{\xi} = \sum_{n=0}^{\infty} \frac{e^{-\lambda \lambda^n}}{n!} (f_{n+1})^* (\delta_{\xi} \otimes \rho^\otimes n), \quad \sigma = \sum_{n=0}^{\infty} \frac{e^{-\lambda \lambda^n}}{n!} (f_{n+1})^* \rho^\otimes n+1. \quad (68)
\]

These formulas are again extended by continuity to all \( \rho \in \mathbb{P}(\mathbb{X}) \). Note that \( \sigma_{\xi} \) and \( \sigma \) are probability densities on the space of the per-individual fitness \( \sigma_{\xi}, \sigma \in \mathbb{P}(\mathbb{R}) \). The update operator \( A : \mathbb{P}(\mathbb{X}) \to \mathbb{P}(\mathbb{X}) \) is given by \( A(\rho) = \frac{\langle \sigma_{\xi}, x \rangle}{\langle \sigma, x \rangle} \). We will discuss neither its domain nor the condition of its continuity.

### 6 Numerical simulations

#### 6.1 Linear selection with large number of compartments and large population

To test the prediction given by the update Eq. (17) for the additive linear case, we carried out numerical simulations of the compartmentalized selection. The simulations were performed with Wolfram Mathematica. The corresponding notebook is provided in Online Resource. The number of compartments was fixed at \( 10^6 \). An initial set of \( 2 \cdot 10^5 \) or \( 2 \cdot 10^6 \) phenotypes (depending on \( \lambda \)) was then drawn from a Gaussian distribution (centered at 1, with variance 1/2) on the interval \([0, 4]\).

One generation is implemented using the following loop, which is then repeated \( n \) times.
Natural selection in compartmentalized environment with reshuffling

- Each value from the set is randomly assigned to a compartment.
- Each compartment is given the local fitness as the mean of the encapsulated phenotypes (with rounding, when needed).
- An updated weight for each phenotype value is obtained by summing the local fitness of each compartment in which it was present (taking into account the multiplicity of that phenotype in each compartment, i.e. having in the end the number of offspring it was able to create overall).
- A new set of $2 \cdot 10^5$ or $2 \cdot 10^6$ phenotype values (depending on $\lambda$) is drawn randomly from the list of the present phenotypes, using the updated weight list.

The result and its comparison to the theoretical prediction are shown on Fig. 4. One can see that the agreement is very good.

### 6.2 The limit of small number of compartments and small population

Although the predictions of the deterministic theory agree well with the individual-based simulation for reasonably large populations (populations of $10^5$–$10^6$ or more are typical for viral infections and for directed evolution experiments in emulsions), we also investigated their applicability to small populations and to small number of compartments. In a standard selection scenario (like the Fisher-Wright process), the only relevant effect observed in small populations is the genetic drift—a stochastic deviation from the deterministic selection due to random sampling effect in the construction of a new generation of a finite number of individuals. The compartmentalization adds another source of stochasticity, and thus it is interesting to compare these two cases. This additional randomness comes from a probabilistic connection between the phenotype of an individual and its fitness. Different realizations of the finite population packing into compartments result in different fitness assignments to individuals. Additional complication comes from the fact that this fitness assignment is not independent for different individuals. This implies a possible difference between the stochastic selection in a finite population with and without compartmentalization. As the formal treatment of the compartmentalized case in a finite population is very difficult and deserves a separate investigation, we compared the two cases by numerical simulations reducing both the population size $N$ (relevant for both cases) and the number of compartments $M$ (which may give a nonnegligible effect).

Individual realizations of simulations become more and more random as $N$ decreases, both because of strong genetic drift of phenotypes with low frequencies and because of randomness in the initial drawing from the continuous distribution. This is especially important for the high-value tail of the initial phenotypic distribution. Indeed, the irregularities due to the randomness are already seen on Fig. 4. And as the high phenotype value front of the distribution is crucially important for the result of long-term selection, effectively defining the final state, it makes more sense to look at the average of an ensemble of trajectories instead of individual trajectories, when the stochasticity becomes too strong. Figure 5 shows the ensemble average of individual-based simulations (phenotypic distributions are averaged for each time frame) for various $N$ and $\lambda$ (and thus for various $M$, as $\lambda = N/M$) for the same simulation algorithm and for the first generation drawn from the same phenotypic
Fig. 4 Numerical simulations and their comparison to the theory. Individual trajectories are shown for two different values of $\lambda$. The phenotypic distribution in the population at each round of selection is shown by the histogram. The theoretical prediction obtained by iterating Eq. (17) is shown as the solid line. The initial phenotypic distribution in each case is $\rho(x) = \frac{1}{\mathcal{N}} e^{-(x-1)^2} I_{[0,4]}(x)$, where $\mathcal{N}$ is the normalization constant. The horizontal axis represents the phenotypic value distribution, as in the previous section. To keep the statistics comparable, for each $M$ we ran simulations $m$ times to keep $mM = 10^6$ constant for values of $M = 10^5, 10^6, \ldots, 10^2$. As before, we used $\lambda = 2$ and $\lambda = 0.2$ for every given $M$, thus changing $N$ from $2 \times 10^6$ to 20.

The limit distribution of the averaged trajectory for small populations observed on these results is the distributions of the final fixed phenotypes of individual trajectories. This spreading can have three sources: 1) the initial sampling from the tail of $\rho_0(x) = \frac{1}{\mathcal{N}} e^{-(x-1)^2} I_{[0,4]}(x)$, which may result in the maximal value phenotype in the sampling smaller than 4, 2) the standard genetic drift, which may result in the fixation of a phenotype different from the maximal one in the initial sampling, and 3) the stochastic effects due to compartmentalization.

To distinguish these effects, we performed the same individual-based simulation but without random compartmentalization, where each individual directly acquired the fitness equal to its phenotype. Focusing on the cases that corresponded to $M = 100$ on Fig. 5, where the compartmentalized case showed fixation of a single phenotype in individual trajectories, we ran the noncompartmentalized simulations with populations sizes $N = 200$ and $N = 20$ to be compared with $\lambda = 2$ and $\lambda = 0.2$, respectively.
Fig. 5 The same numerical experiments (the same initial distribution, the same selection algorithm) as on Fig. 4 performed with different number of compartments $M$ and for two values of $\lambda$ for each value of $M$. The ensemble average phenotypic distributions are shown for each indicated number of repeats. The black line shows the theoretical prediction by Eq. (17). Numbers in boxes indicate rounds of selection. The horizontal axis represents the phenotypic value.

Fig. 6 Distribution of fixed phenotypes in individual-based simulations of compartmentalized and classical linear selection for two small values of the population size. The corresponding $M$ and $\lambda$ for the compartmentalized case are shown in parentheses. The distributions are based on $10^4$ trajectories. The red line shows the empirical distribution of the maximal phenotypic value in the first generation (a sample of size $N$ drawn from the distribution $\rho_0$).

The result of these simulations is shown on Fig. 6, where again a mean distribution of the fixed phenotypes are shown averaged for $10^4$ individual trajectories. One can see that the main part of the variability in the noncompartmentalized case comes from the sampling of the initial population, as the distribution of the fixed phenotypes closely matches the (numerically computed) distribution of the maximum phenotypic value in a sample of size $N$ from $\rho_0$. The final distribution, however, is broadened and slightly shifted to low values of fitness. We attribute this to the genetic drift. Note how the
compartmentalization does not affect this process in case of small $\lambda$ but significantly worsens the final distribution in case of large $\lambda$—the effect that, in contrast to the regular genetic drift, becomes stronger with the increase of the population size. This effect may be a manifestation of the increase in the correlations between the fitness of individuals. Another possible explanation could be the weakening of the deterministic part of the selection process (predicted by Eq. (17) for large values of $\lambda$). This could give more chances for stronger mutants to be lost before their frequency grows high enough. It is difficult to separate these effects from this kind of data so we performed an additional numerical experiment designed specially for this purpose.

6.3 Stochasticity in a bivariate population under linear and nonlinear selection

To distinguish the stochastic effect of compartmentalization from the deterministic reduction of the selection pressure predicted by (17), we performed individual-based simulations on a population of much simpler structure (a bivariate case). More specifically, we took the initial distribution with density $\rho = p\delta_{x_1} + (1 - p)\delta_{x_0}$, where we assume $x_0$ to be the wild type phenotype, $x_1$ to be an invading mutant, and $p$ to be the gene frequency if this mutant.

The case of a bivariate population makes the comparison simpler, as in the classical situation, the mean change of the frequency in one cycle of selection is given by the deterministic part of selection obtained in the infinite population limit, while the variance of this change results from the sampling stochasticity. To quantify the additional stochastic effect of the compartmentalization, we compared the mean frequency change to the value predicted by (17) for the corresponding value of $\lambda$ and its variance with the sampling variance (classical genetic drift) for the corresponding population size. The results are depicted on Fig. 7. Interestingly, the combination of the deterministic theory developed in our work with the sampling variance due to drawing of the new population (the standard genetic drift) completely explains the observed stochasticity in the compartmentalized selection with the linear selection function.

We also performed the same simulations for nonlinear selection functions theoretically studied in this article: a quadratic selection, an exponential one, and $f(x) = 1 - e^{-x}$ [see (51) and (54) with the following remark]. The latter function, being a sum of two exponentials, $e^{0x}$ and $e^{-x}$, and thus completely described by (54), is an example of a saturating selection: it monotonously increases but is limited from above by 1. The results of these simulations are shown on Fig. 7, too. Although the quadratic case, and to even lesser extent the saturating selection, shows a wider variance of the gene frequency in one cycle than predicted by the genetic drift, the effect is very weak. Only the exponential case strongly demonstrates the effect of the compartmentalization when the number of compartments decreases ($\lambda$ increases). Interestingly, the deviation of the noise from the genetic drift happens along with the deviation of the mean from the prediction based on the infinite population theory.

We can conclude that the deterministic theory with genetic drift works well even in very small populations at least for not very strongly growing selection functions. However, the behaviour of the exponential selection clearly demonstrates some new effect that deserves a separate investigation.

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Fig. 7 Mean gene frequency $p$ of the more active mutant with phenotype $x_1$ (resident phenotype $x_0$) in a small population of size $N$ and its standard deviation after one selection cycle with and without compartmentalization ($M$ compartments) and with various selection functions $f$. Statistical properties are computed on a sample of $10^5$ individual numerical experiments (in blue). These values are compared to the prediction of the deterministic infinite population theory and with the standard deviation given by a simple genetic drift due to sampling (in red). The case marked “classic” corresponds to the classical selection without any compartmentalization, where the selection function directly defines the fitness.

Discussion and conclusion

In this work we considered a model of the group selection with no intragroup competition, where grouping happens by a random encapsulation of the individuals in a collection of compartments at the beginning of each replication cycle. This study was initially motivated by experiments from the field of artificial molecular evolution. In particular, compartmentalized in vitro directed evolution protocols provide a very pure implementation of our model, and may in the future be used to test its predictions.

We note however that our framework can also apply to some situations of natural evolution. Viruses, for example, exist as highly polymorphic populations, or quasi-species (Domingo et al. 2001), which randomly infect individual cells (Manrubia and Lázaro 2006) to carry their replication cycle. These cells thus serve as genotype-phenotype linkage-maintaining compartments. The possibility of co-infection of a given cells by multiple, possibly non-clonal viral particles is largely acknowledged, and generally parametrized by the averaged “multiplicity of infection” (González-Jara et al. 2009; Frank 2001; Novella et al. 2004). Additionally, inside the cells,
the genetic and viral replication is carried according to the phenotypic activity of gene products from the viral genome (Manrubia and Lázaro 2006). This mode of reproduction naturally involves template indiscriminateness, and is thus accurately described by our model.

In the context of the origin of life, it is also assumed that ancestral replicators were RNA molecules functioning as universal RNA-dependent RNA-polymerases with activity \textit{in trans}. These primordial replicators possibly used naturally formed vesicles, coacervates, pores in regolith, or other microcompartmentalized niches. This implies a random compartmentalization—hence frequency-dependent—stage in their replication cycle. In a similar way, modern parasites may also fall in our conceptual scheme, because multiple unrelated individuals may infect a single host and modify their joint virulence/survival as a result of interaction (Davies et al. 2002; Fried and Alenick 1981; Fried et al. 1990).

Finally, the process is similar to selection in polyploid organisms under random mating. The analogy becomes exact if one considers zygotes to be compartments and the haploid genomes to be the individuals under selection. There are two particularities though: the fixed group size in the polyploids (the ploidy of the organism) and the presence of the genetic cross-over, not considered in our model. Thus, the problem considered in the present article can be viewed as a generalization of natural selection theory to organisms with variable ploidy, in the absence of recombination.

Our model includes a number of simplifications: no selection inside compartments (pure group selection), infinitely large populations, no overlap between generations, completely random occupation of compartments by individuals, absence of mutations, deterministic phenotypic expression, and (in most parts) a single additive phenotypic trait. Many of these assumptions are relevant to the initial motivation of the work (in vitro micro-compartmentalized evolution).

The most restrictive condition is the demand of the additivity of the phenotype in a compartment. In the context of directed evolution of enzymes, when the phenotypic trait is the enzymatic activity, this hypothesis neglects any effect of the activity saturation. Another case of failure of the additivity is brought by the independence of the total phenotype in a compartment on the number of individuals in it \textit{per se}. This situation is encountered, for example, when individuals are bacteria that contain a plasmid (plasmids) with the gene and the appropriate protein product, like in the work by Ghadessy et al. (2001). If the content of a bacterial cell has an effect on critical reactions during the reproduction phase, the additivity of per-individual activities fails.

The deterministic infinite population limit is not very restrictive, as we demonstrated by numerical simulations in small finite populations. First, typical population sizes in some potential domains of application for our theory (high throughput \textit{in vitro} evolution, viral evolution) are very large. Second, when the population is small, the developed theory well predicts the average result of selection. The stochastic component, in turn, can be almost completely attributed to and taken in to account by the standard genetic drift, despite the compartmentalization. This situation is similar to the standard mathematical treatment of the selection process in diploid organisms, where the Hardy–Weinberg equilibrium followed by the deterministic selection is coupled to the haploid genetic drift (Ewens 2004). In the light of the analogy between a compartmentalized population and a population of organisms with variable ploidy, the
infinite population limit is equivalent to the Hardy-Weinberg equilibrium assumption. This approximation fails, however, if the selection function grows too fast, like in the exponential selection.

The initial motivating question for our work was the influence of the average number of individuals per compartment ($\lambda$) on the selection dynamics. We answered this question using distribution theoretical tools. The main result is a quantitative characterization of the co-encapsulation effect in the form of a discrete-time dynamical equation for the phenotypic distribution that uses $\lambda$ as the main parameter. Using this dynamical description, we thoroughly analyzed the simplest case: additive phenotype with linear selection function under Poisson partitioning in the compartments. The main conclusion is that the selection process, contrary to a common belief in the field of directed evolution, is still effective even in the context of mixing many individuals with different phenotypes in the same compartment. In quantitative terms, random co-encapsulation slows down the selection dynamics (the rate of the mean fitness change) by a factor that decays approximately as $1/\lambda$. With ten individuals per compartment in average, the selection is roughly ten-fold slower than that for the extremely diluted case, when any individual is alone in its compartment. In all cases, however, independent of $\lambda$, the most active mutant is still eventually selected for.

We also developed a framework to treat more complicated cases like nonlinear fitness, nonadditive phenotype, and multiple trait phenotype. For some special cases (polynomial and exponential selection), we obtained dynamical equations in closed form with an explicit effect of $\lambda$. The striking observation here is that different nonlinearities have very different resistance to random co-encapsulation. In particular, the exponential selection is completely immune to this effect (in the infinite population approximation). This is especially interesting as some important cases of evolving replicators can be approximated by this regime, e.g. when the phenotypic trait directly controls the catalytic rate of the replication.

Although this is not the first theoretical work on compartmentalized selection, we greatly generalized previous research, which only considered populations of few phenotypes. Our model is suitable for the treatment of arbitrary phenotypic distributions, discrete and continuous alike, and arbitrary laws of distribution of individuals among the compartments. Thanks to the advanced mathematical tools applied in this work, the computations became more transparent and simple in comparison to the traditional combinatorial approach, which gets very cumbersome already for a quadratic selection function, even in the case of a simple bivariate population.

Finally, one practical implication of this study concerns the design of in vitro evolution experiments. While many practitioners tend to empirically select very small values of $\lambda$, we show that higher library concentration will not drastically affect the performance of the experiment. The corresponding increase of the throughput is, however, paid for by the decrease of the selection pressure, and more rounds may become necessary to fix the best genotypes. A more comprehensive assessment of the cost-benefit balance for directed evolution experiments will be published elsewhere.

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**Appendices**

**A.1 Random variables and generalized functions**

This appendix does not contain new results. Its purpose is to be an introduction to the framework used in the article.

A random variable $X$ on $\mathbb{R}$ is associated with a positive Radon measure $\mu_X$, which can be understood as a non-negative generalized function (linear continuous functional) $\rho_X$ defined on the space of continuous functions with compact supports $C_c$ endowed with the appropriate topology [see, for example, the book by Schwartz (1997)]. This topology is conventionally induced by the following convergence rule: we say $\varphi_n \to \varphi$ in $C_c$, if there is a compact $K$ such that $\forall n \sup \{\text{supp } \varphi_n \subset K \}$ and $\varphi_n \to \varphi$ homogeneously. We will denote the topological linear space of generalized functions on $C_c$ endowed with the weak topology as $C'_c$, so the topology on $C'_c$ is induced by the convergence $\rho_n \to \rho \iff \forall \varphi \in C_c \langle \rho_n, \varphi \rangle \to \langle \rho, \varphi \rangle$. Such convergence is equivalent to the weak convergence of the corresponding measures in the measure-theoretical language. As this is the only convergence of generalized functions that we consider in the article, the use of the sign $\to$ to denote it does not bring any confusion.

A generalized function $\rho$ is called non-negative, if for any non-negative $\varphi \in C_c$ (that is $\forall x \in \mathbb{R} \varphi(x) \geq 0$) we have $\langle \rho, \varphi \rangle \geq 0$. We will denote the subset of non-negative generalized functions with the subset topology as $C'_c+$.

It is possible to extend the action of $\rho$ from functions in $C_c$ to any indicator function of a Borel set $\chi_B$, $B \in \mathcal{B}$, where $\mathcal{B}$ is the Borel $\sigma$-algebra. This is done using the so-called upper and lower value of $\rho$ on an indicator of a set [in the book by Schwartz (1997) this corresponds to the upper and the lower measure of a set]. The upper value of $\chi_A$ is defined by

$$\langle \hat{\rho}, \chi_A \rangle = \inf_{U \subset A} \sup_{\sup \{\text{supp } \varphi \subset U \}} \sup_{0 \leq \varphi \leq 1} \langle \rho, \varphi \rangle,$$

where $U$ are open sets in the standard topology on $\mathbb{R}$, and $\varphi \in C_c$. The lower value is defined as

$$\langle \check{\rho}, \chi_A \rangle = \sup_{K \subset A} \inf_{\varphi \geq 0, \varphi|_K = 1} \langle \rho, \varphi \rangle,$$

where $K$ are compact. If, for compactly supported non-negative $\rho$, $\langle \hat{\rho}, \chi_A \rangle = \langle \check{\rho}, \chi_A \rangle$, the set $A$ is called $\rho$-measurable and the value $\langle \rho, \chi_A \rangle$ is defined as this common value and is called the measure of $A$. It can be proven that for any compactly supported non-negative generalized function $\rho$ any Borel set is $\rho$-measurable [it is also $\sigma$-regular, see the book by Schwartz (1997)]. In particular, the whole $\mathbb{R}$ is measurable, and in fact, for
\( \rho \) to be a (generalized) probability density functions, we require \( \langle \rho, \chi \rangle \overset{\text{def}}{=} \langle \rho, 1 \rangle = 1. \) For non-negative \( \rho \) that have \( \langle \rho, 1 \rangle < \infty \), any set \( A \) such that \( \langle \hat{\rho}, \chi_A \rangle = \langle \hat{\rho}, \chi_A \rangle \) is called \( \rho \)-measurable, whether \( \rho \) is compactly supported or not. Note that for a non-negative \( \rho \) and a continuous \( f, \langle \rho, f \rangle \) is well defined as \( \langle f \rho, 1 \rangle \) and one does not need any further development of the theory.

Any point-set is measurable, too, and we can compute a measure of a point \( x \) as

\[
\langle \rho, \chi_{\{x\}} \rangle = \inf_{\varphi \geq 0, \varphi(x) = 1} \langle \rho, \varphi \rangle. \tag{71}
\]

From now on we will call non-negative generalized functions with \( \langle \rho, 1 \rangle = 1 \) (probability) densities (of random variables). The meaning of a density \( \rho_\xi \) of a random variable \( \xi \) is that the probability to find the value of \( \xi \) in a set \( A \) is equal to \( \langle \rho_\xi, \chi_A \rangle \). A (cumulative) distribution function of \( \xi \) is the function \( F_\xi: x \mapsto \langle \rho_\xi, \chi_{(-\infty,x]} \rangle \). The density is the (generalized) derivative of its own distribution function. The mathematical expectation (the mean) of the random variable is then computed as \( \bar{x} = \langle \rho, x \rangle \).

Any density \( \rho \) can be uniquely decomposed into a sum

\[
\rho = \rho_a + \rho_p + \rho_r, \tag{72}
\]

where \( \rho_a \) is the regular part, \( \rho_p \) is the point-mass part, and \( \rho_r \) is the residual singular part.

The regular part \( \rho_a \) is a regular generalized function, i.e. its action on any \( \varphi \in C_c \) can be represented by \( \langle \rho_a, \varphi \rangle = \int f \varphi \, dx \) for a unique \( f \in L^1_{\text{loc}} \) (the integration is in the sens of Lebesgue). The regular part is also called the absolutely continuous part (hence the notation \( \rho_a \)). A density that has only this part is also called absolutely continuous. Any point has a zero measure in respect to an absolutely continuous density. It is convenient to identify (a representative of) \( f \) with \( \rho_a \) and to write \( \rho_a \) instead of \( f \).

The point-mass part \( \rho_p \) is an at most countable sum of \( \delta \)-functions

\[
\rho_p = \sum_{n \in \mathbb{N}} a_n \delta_{x_n}, \quad a_n \geq 0, \quad n \neq m \Rightarrow x_n \neq x_m, \quad \sum_{n \in \mathbb{N}} a_n \leq 1. \tag{73}
\]

It follows that \( \forall x \neq x_n \langle \rho_p, \chi_{\{x\}} \rangle = 0 \), and \( \forall x_n \langle \rho_p, \chi_{\{x_n\}} \rangle = a_n \). Physically speaking, \( \rho_p \) represents all fitness values that are present in macroscopic quantities in the population.

Finally, the residual singular part \( \rho_r \) is characterized by the zero (Lebesgue) measure of its support and, at the same time, \( \forall x \langle \rho_r, \chi_{\{x\}} \rangle = 0 \). Its support is Cantor set-like and its distribution function is Cantor function-like.

The regular and the residual singular parts form together the continuous part \( \rho_c = \rho_a + \rho_r \). The sum of the point-mass and the residual singular parts is the singular part \( \rho_s = \rho_p + \rho_r \). It is tempting to disregard \( \rho_r \) as unphysical. However, it may turn to be a good tool to model libraries obtained by a random mutagenesis from a single mutant in a very rugged fitness landscape. It might be possibly a good approximation to a library generated on a smooth landscape but by an error-prone PCR with large number of cycles.
A.2 Continuity of operator $A$ and of the operators that generate $\sigma_x$ and $\sigma$ for the linear fitness case

Let $\mathbb{P}$ be the space of probability densities with, so $\mathbb{P} = \{ \rho \in C^\prime_c \mid \langle \rho, 1 \rangle = 1 \}$. Let $\mathbb{P}_p$ be the space of finite point-mass probability densities, so $\mathbb{P}_p = \{ \rho \in \mathbb{P} \mid \exists n \in \mathbb{N}, \rho = \sum_{k=0}^{n} a_k \delta_{x_k} \}$. Let us fix some very large positive number $L$. Let us denote $I \text{ def } = [0, L]$. Let $\mathbb{P}_I$ be the space of probability densities concentrated in $I$, so $\mathbb{P}_I = \{ \rho \in \mathbb{P} | \text{supp } \rho \subset I \}$. Let us denote $N \text{ def } = \{ \rho \in \mathbb{P} | \langle \rho, x \rangle = 0 \}$. Let us also denote $N_p \text{ def } = N \cap \mathbb{P}_p$. If $N_p$ was nowhere dense in $\mathbb{P}_p$, if, in addition, $\mathbb{P}_p$ was dense in $\mathbb{P}$ and (17) happened to be continuous both in $\mathbb{P}_p \setminus N_p$ and $\mathbb{P} \setminus N$, we could take this expression as an extension by continuity of $A$ from $\mathbb{P}_p$ to $\mathbb{P}$. Unfortunately, this assertion is not true.

**Proposition 1** For any $\lambda > 0$, the operator $A: \mathbb{P}_p \setminus N_p \to \mathbb{P}_p$ defined by (17) is nowhere continuous.

**Proof** Take any $\rho \in \mathbb{P}_p \setminus N_p$. By the definition of $\mathbb{P}_p$, we have $0 < |\langle \rho, x \rangle| < \infty$. Consider the sequence

$$\rho_n = \frac{n}{n+1} \rho + \frac{1}{n+1} \delta_{x^2}. \quad (74)$$

For any large enough $n$, $\rho_n \in \mathbb{P}_p \setminus N_p$. Furthermore, $\rho_n \to \rho$ in the topology of $\mathbb{P}_p \setminus N_p$. Indeed, for any test function $\varphi \in C_c$ there is a number $n_0$ such that for any $n > n_0$ the point $n^2$ does not belong to the support of $\varphi$. Therefore, for $n > n_0$ we have

$$\langle \rho_n, \varphi \rangle = \frac{n}{n+1} \langle \rho, \varphi \rangle \to \langle \rho, \varphi \rangle. \quad (75)$$

From the other hand, $\rho_n$ does not converge to $\rho$ in mean. Indeed,

$$\langle \rho_n, x \rangle = \frac{n}{n+1} \langle \rho, x \rangle + \frac{n^2}{n+1} \to +\infty. \quad (76)$$

Therefore,

$$A(\rho_n) \to \left( 1 - g(\lambda) \right) \rho \neq A(\rho) = \left( 1 - g(\lambda) + g(\lambda) \frac{x}{\langle \rho, x \rangle} \right) \rho. \quad (77)$$

\[ \square \]

This deplorable fact, however, can be remedied by the restriction of the space of the considered probability densities to $\mathbb{P}_I$ for some $\mathcal{I}$. This restriction has purely
technical meaning and does not reflect any physical constraints. Nevertheless, some justification comes from the fact that there is a universal upper bound on the activity of any enzyme of the considered class of enzymes. This upper bound is reflected by the number \( \mathcal{L} \) that defines \( \mathcal{I} \). Note that \( \mathbb{P}^{\mathcal{I}} \cap N = \mathbb{P}^p_0 \cap N = \{ \delta_0 \} \).

**Theorem 1** For any \( \mathcal{I} \), the operator \( A : \mathbb{P}^{\mathcal{I}} \setminus \{ \delta_0 \} \to \mathbb{P}^{\mathcal{I}} \) defined by the formula (17) is continuous.

**Proof** Let \( \rho_n \) be some sequence from \( \mathbb{P}^{\mathcal{I}} \setminus \{ \delta_0 \} \) that converges to some \( \rho \in \mathbb{P}^{\mathcal{I}} \setminus \{ \delta_0 \} \). Then

\[
|\langle \rho_n, x \rangle - \langle \rho, x \rangle| = |\langle \rho_n - \rho, x \rangle| = |\langle \rho_n - \rho, \eta \rangle| \to 0,
\]

where \( \eta \) is some function from \( C_c \) such that \( \forall x \in \mathcal{I} \eta(x) = x \).

It follows that

\[
\left( 1 - g(\lambda) + g(\lambda) \frac{x}{\langle \rho_n, x \rangle} \right) \to \left( 1 - g(\lambda) + g(\lambda) \frac{x}{\langle \rho, x \rangle} \right)
\]

point-wise on \( \mathcal{I} \). As all the involved functions are also continuous on the compact set \( \mathcal{I} \), the convergence is uniform.

Let us denote the function on the right-hand side of (79) as \( \tilde{\psi} \) and the functions on the left-hand side as \( \tilde{\varphi}_n \) (for each \( n \), \( \tilde{\varphi}_n \) corresponds to the function generated by \( \rho_n \)). Then it is always possible to find functions \( \varphi, \varphi_n \in C_c \) such that \( \varphi_n \to \varphi \) in \( C_c \) and \( \varphi|_{\mathcal{I}} = \tilde{\varphi}|_{\mathcal{I}}, \varphi_n|_{\mathcal{I}} = \tilde{\varphi}_n|_{\mathcal{I}} \) and, therefore, \( \tilde{\varphi}_n \rho = \varphi_n \rho, \tilde{\varphi} \rho = \varphi \rho \).

As \( A(\rho_n) = \varphi_n \rho_n \) and \( A(\rho) = \varphi \rho \), what is left to be proven is that given \( \rho_n \to \rho \) and \( \varphi_n \to \varphi \) we have \( \varphi_n \rho_n \to \varphi \rho \). First notice that \( \sup |\varphi_n \psi - \varphi \psi| \to 0 \) for any \( \psi \in C_c \) (from which it follows that \( \varphi_n \psi \to \varphi \psi \) in \( C_c \)). Therefore, for any \( \epsilon > 0 \) there is \( n_0 \) such that for any \( n > n_0 \) we have \( \sup |\varphi_n \psi - \varphi \psi| < \epsilon \). We have, therefore, for any \( n > n_0 \)

\[
|\langle \varphi_n \rho_n - \varphi \rho, \psi \rangle| \leq |\langle \rho_n - \rho, \varphi \psi \rangle| + |\langle \rho_n, \varphi_n \psi - \varphi \psi \rangle| \\
\leq |\langle \rho_n - \rho, \varphi \psi \rangle| + \sup |\varphi_n \psi - \varphi \psi| < |\langle \rho_n - \rho, \varphi \psi \rangle| + \epsilon.
\]

But \( \rho_n \to \rho \) means that there is \( n_1 > n_0 \) such that for any \( n > n_1 \) we have \( |\langle \rho_n - \rho, \varphi \psi \rangle| < \epsilon \). Therefore, for any \( \epsilon \) and for any \( \psi \) we have \( |\langle A(\rho_n) - A(\rho), \psi \rangle| < 2\epsilon \) starting from \( n_1 \).

As it is seen from the proof, the statement of the theorem stays correct, if we replace \( \mathcal{I} \) with any compact set \( K \) with nonempty interior and if we replace \( \mathbb{P}^{\mathcal{I}} \setminus \{ \delta_0 \} \) with \( \mathbb{P}^K \setminus (\mathbb{P}^K \cap N) \), where \( \mathbb{P}^K = \{ \rho \in \mathbb{P} | \sup \rho \subset K \} \). Furthermore, the set \( \mathbb{P}^K_0 \cap N \) is nowhere dense in \( \mathbb{P}^K_0 \) and the set \( \mathbb{P}^K \cap N \) is nowhere dense in \( \mathbb{P}^K \).

The only thing that is left to be proven is that the space of finite discrete densities is dense in the space of general densities.
Theorem 2 For any $\mathcal{I}$, the space $\mathbb{P}^{\mathcal{I}}_p$ is dense in $\mathbb{P}^{\mathcal{I}}$ as its subset.

Proof First let us prove that the space $\mathbb{P}^{\mathcal{I}}_p \cap C_c$ is dense in $\mathbb{P}^{\mathcal{I}}$, where $C_c$ is understood as being naturally embedded into $C'_c$. That is, any density from $\mathbb{P}^{\mathcal{I}}$ can be approximated by a sequence of densities from $C_c$ with the supports in $\mathcal{I}$.

Consider some $\omega \in C_{c+}$ such that $\forall x \omega (-x) = \omega (x)$ and $\int \omega (x) \, dx = 1$. Let us denote $r = \text{diam supp } \omega$ and $\omega_n(x) = \frac{1}{n} \omega \left( \frac{x}{n} \right)$. Let us also consider the sequence of mappings $F_n : \mathbb{R} \rightarrow \mathbb{R}, x \mapsto \frac{\mathcal{L}}{\mathcal{L} + 2 \frac{r}{n}} \left( x + \frac{r}{n} \right)$. For each $n$, $F_n$ bijectively maps the interval $\left[ -\frac{r}{n}, \mathcal{L} + \frac{r}{n} \right]$ to the interval $[0, \mathcal{L}] = \mathcal{I}$.

For any generalized function $\rho \in \mathbb{P}^{\mathcal{I}}$, take the sequence of $\psi_n = (F_n)_*(\rho \ast \omega_n) \in \mathbb{P}^{\mathcal{I}} \cap C_c$. Since $\omega_n \rightarrow \delta_0$ in $\mathbb{P}$, as $n \rightarrow \infty$, we have $\rho \ast \omega_n \rightarrow \rho$ in $\mathbb{P}$. Let us prove that $\psi_n \rightarrow \rho$.

For any $\varphi \in C_c$ we have $\langle (F_n)_*(\rho \ast \omega_n), \varphi \rangle = \langle \rho \ast \omega_n, \varphi \circ F_n \rangle$. It is not difficult to show that $\varphi \circ F_n \rightarrow \varphi$ in $C_c$. The point $x_0 = \mathcal{L}/2$ is the stationary point for all $F_n$. All $F_n$ are affine and contracting with the contraction coefficient $\mathcal{L}/(\mathcal{L} + 2r/n)$. Their inverses $F_n^{-1}$ are expanding with the expansion coefficient $\kappa_n \equiv 1 + 2r/(n\mathcal{L})$. Note that $\forall n, \kappa \equiv \kappa_1 \geq \kappa_n$. Let $\Delta \equiv \max \left( |x_0 - \inf \text{ supp } \varphi|, |x_0 - \sup \text{ supp } \varphi| \right)$ and $K \equiv [x_0 - \kappa \Delta, x_0 + \kappa \Delta]$. Then $\forall n > 0$, $\sup \varphi \circ F_n \subset K$ and $\sup \varphi \subset K$. As $F_n \rightarrow \text{Id}_\mathbb{R}$ pointwise on $\mathbb{R}$ and $F_n$ are continuous, this convergence is uniform on $K$. That is $\sup_{x \in K} |F_n(x) - x| \rightarrow 0$. As $\varphi$ is continuous and compactly supported, it is also uniformly continuous. Therefore,

$$\sup_{x \in \mathbb{R}} \left| \varphi \left( F_n(x) \right) - \varphi(x) \right| \rightarrow 0.$$  \hspace{1cm} (81)

But together with $\sup \varphi \circ F_n \subset K$ this proves that $\varphi \circ F_n \rightarrow \varphi$ in $C_c$.

We have $\rho \ast \omega_n \rightarrow \rho$ and $\varphi \circ F_n \rightarrow \varphi$. Using the same reasoning as in the proof of Theorem 1, we conclude that $\langle \psi_n, \varphi \rangle = \langle \rho \ast \omega_n, \varphi \circ F_n \rangle \rightarrow \langle \rho, \varphi \rangle$, and thus, $\psi_n \rightarrow \rho$.

From the other hand, any $\psi \in \mathbb{P}^{\mathcal{I}}_p \cap C_c$ can be approximated by a sequence from $\mathbb{P}^{\mathcal{I}}_p$. Indeed, we can select some sequence of conventionally ordered Darboux partitions $\{\Delta_n\}$ of some interval $[a, b]$ that contains $\operatorname{supp} \psi$ (one can take the whole $\mathcal{I}$) with the graininess of the partitions going to 0 with $n \rightarrow \infty$, where $\Delta_n = \{x^{(n)}_k\}, \, k \in \{0, 1, \ldots, K_n\}, \, x^{(n)}_n < x^{(n)}_{k+1}, \, x^{(n)}_0 = a, \, x^{(n)}_{K_n} = b, \, \Delta x^{(n)}_k = x^{(n)}_{k+1} - x^{(n)}_k, \, \text{ and } \max_{k<n} \Delta x^{(n)}_k \rightarrow 0, \, \text{ when } n \rightarrow \infty$. Then we can take the sequence of $\rho_n \in \mathbb{P}^{\mathcal{I}}_p$ of the following form

$$\rho_n = \sum_{k=0}^{K_n-1} \Delta x^{(n)}_k \psi(\xi^{(n)}_k) \delta^{(n)}_{\xi^{(n)}_k}, \hspace{1cm} (82)$$
where \( \xi_k^{(n)} \in [x_k^{(n)}, x_{k+1}^{(n)}] \) such that

\[
\psi(\xi_k^{(n)}) \Delta x_k^{(n)} = \int_{x_k^{(n)}}^{x_{k+1}^{(n)}} \psi(x) \, dx.
\] (83)

Such \( \xi_k^{(n)} \) always exist by the mean value theorem. Their role is to enforce \( \langle \rho_n, 1 \rangle = 1 \).

Then for any \( \varphi \in \mathcal{C}_c \) we have

\[
\langle \rho_n, \varphi \rangle = \left\langle \sum_{k=0}^{K_n-1} \Delta x_k^{(n)} \psi(\xi_k^{(n)}) \delta_{\xi_k^{(n)}}, \varphi \right\rangle
= \sum_{k=0}^{K_n-1} \Delta x_k^{(n)} \psi(\xi_k^{(n)}) \varphi(\xi_k^{(n)}) \to \int_a^b \psi(x) \varphi(x) \, dx = \langle \psi, \varphi \rangle,
\] (84)
as both \( \psi \) and \( \varphi \) are continuous, and thus, \( \psi \varphi \) is Riemann integrable. As \( \mathbb{P}^\mathcal{G} \cap \mathcal{C}_c \) is dense in \( \mathbb{P}^\mathcal{G} \), it follows that \( \mathbb{P}^\mathcal{G}_\rho \) is dense in \( \mathbb{P}^\mathcal{G} \).

Note that the proof of the theorem is easily extended to probability densities on \( \mathbb{R}^n \), the case important for a multitrait selection considered in Sect. 5.3. The only difference is that in this case Riemann sums are built on the base of Jordan partitions of a Jordan-measurable set (a simple rectangular parallelepiped is enough) that contains \( \text{supp} \psi \).

We now will prove that the operators that generate \( \sigma_x \) and \( \sigma \) from \( \rho \) defined by formulas (11) and (12) are continuous, too, and that they can be thus extended to any probability density. It means that they can be used independently of \( A \), if the situation demands it. Let us denote these operators from \( \mathbb{P} \) to itself as \( \Sigma_x \) and \( \Sigma \). Let us also denote \( \sigma_x \rho \overset{\text{def}}{=} \Sigma_x(\rho) \) and \( \sigma \rho \overset{\text{def}}{=} \Sigma(\rho) \) to be able to distinguish fitness distributions generated by different phenotypic distributions.

**Theorem 3** For any \( x \in \mathbb{R} \), the operators \( \Sigma_x, \Sigma : \mathbb{P} \to \mathbb{P} \) are continuous.

**Proof** We will prove the theorem only for \( \Sigma_x \). The proof for \( \Sigma \) is analogous. The proof is essentially based on the absolute convergence of all the involved numerical series.

Let us choose any sequence of \( \rho_n \in \mathbb{P} \) that converges to some \( \rho \in \mathbb{P} \) in \( \mathbb{P} \). We need to prove that \( \Sigma_x(\rho_n) \to \Sigma_x(\rho) \). Let us choose some \( \varphi \in \mathcal{C}_c \). The value of \( \langle \Sigma_x(\rho), \varphi \rangle \) is equal to

\[
\langle \Sigma_x(\rho), \varphi \rangle = \sum_{k=0}^{\infty} P_k \langle \delta_x \ast \rho^{*k}, \varphi \circ h_{k+1} \rangle,
\] (85)

where \( P_k = e^{-\lambda} \lambda^k / k! \) and \( h_k : x \mapsto x/k \).
First note that \( \sup_x |\varphi \circ h_k(x)| \leq \sup_x |\varphi(x)| \). Let us denote \( \Phi \overset{\text{def}}{=} \sup_x |\varphi(x)| \). Then the following estimate is correct

\[
|\langle \delta_x \ast \rho_n \ast^k - \delta_x \ast \rho^{*k}, \varphi \circ h_{k+1} \rangle| \leq |\langle \delta_x \ast \rho_n^{*k}, \varphi \circ h_{k+1} \rangle| + |\langle \delta_x \ast \rho^{*k}, \varphi \circ h_{k+1} \rangle| \leq \Phi \left( \langle \delta_x \ast \rho_n^{*k}, 1 \rangle + \langle \delta_x \ast \rho^{*k}, 1 \rangle \right) = 2\Phi. \tag{86}
\]

This, in turn, implies

\[
|\langle \Sigma_x(\rho_n) - \Sigma_x(\rho), \varphi \rangle| \leq \sum_{k=0}^{\infty} P_k |\langle \delta_x \ast \rho_n^{*k} - \delta_x \ast \rho^{*k}, \varphi \circ h_{k+1} \rangle| \leq 2\Phi \sum_{k=0}^{\infty} P_k = 2\Phi < \infty. \tag{87}
\]

Therefore, for any \( \epsilon > 0 \) there is \( k_0 \) such that \( \sum_{k=k_0}^{\infty} 2\Phi P_k < \epsilon \), and, thus, by (86), for any \( n \)

\[
\sum_{k=k_0}^{\infty} P_k |\langle \delta_x \ast \rho_n^{*k} - \delta_x \ast \rho^{*k}, \varphi \circ h_{k+1} \rangle| < \epsilon. \tag{88}
\]

From the other hand, for any \( k, \rho_n \rightarrow \rho \) implies \( \delta_x \ast \rho_n^{*k} \rightarrow \delta_x \ast \rho^{*k} \). Therefore, for any \( \epsilon > 0 \) and any \( k_0 \) there is \( n_0 \) such that for any \( n > n_0 \)

\[
\sum_{k=0}^{k_0-1} P_k |\langle \delta_x \ast \rho_n^{*k} - \delta_x \ast \rho^{*k}, \varphi \circ h_{k+1} \rangle| < \epsilon. \tag{89}
\]

Using the following intuitive logical formula

\[
(\forall x \forall y \forall z A(x, y, z)) \land (\forall x \forall y \exists z B(x, y, z)) \Rightarrow \forall x \forall y \exists z \left( A(x, y, z) \land B(x, y, z) \right), \tag{90}
\]

where \( A \) and \( B \) are some propositional functions in three variables, we conclude that for any \( \epsilon > 0 \) there exists \( n_0 \) such that for any \( n > n_0 \)

\[
\sum_{k=0}^{\infty} P_k |\langle \delta_x \ast \rho_n^{*k} - \delta_x \ast \rho^{*k}, \varphi \circ h_{k+1} \rangle| < 2\epsilon, \tag{91}
\]

and thus, \( |\Sigma_x(\rho_n) - \Sigma_x(\rho)| < 2\epsilon. \) \( \square \)

The only assertion that is left to be proven to justify the extension of \( \Sigma_x \) and \( \Sigma \) from \( \mathbb{P}_p \) to \( \mathbb{P} \) is that \( \mathbb{P}_p \) is dense in \( \mathbb{P} \). This theorem can be proven in the same way as Theorem 2 but simpler. One can simply take \( \psi_n = \rho \ast \omega_n \).
The assumption of the Poisson distribution of the individuals in the compartments is essential here. We also assume that the phenotype-fitness relation is defined by a continuous selection function \( f \). By its meaning, \( f \) is expected to be nonnegative on the positive semiaxis. We, however, will treat a more general case, which will be useful for the question of an approximation of \( f \). The phenotype is considered to be additive.

The notations are the same as in “Appendix A.2”, except that by \( \sigma_{f,\rho} \) and \( \sigma_{f,\rho} \) we will denote the expressions (11) and (12), respectively, where \( h_n : x \mapsto f(x)/n \), and we explicitly indicate the dependence on \( f \) and \( \rho \). By \( \Sigma_{f,\rho} \) and \( \Sigma_{f,\rho} \) we will denote the operators \( \Sigma_{f,\rho} : \rho \mapsto \sigma_{f,\rho} \) and \( \Sigma_{f,\rho} : \rho \mapsto \sigma_{f,\rho} \). We will also denote \( N_{f,\rho} = \{ \rho \in \mathbb{P} | \langle \sigma_{f,\rho}, x \rangle = 0 \} \), \( N_{f,\rho} = \mathbb{P} \cap \mathbb{P}_{f,\rho} \), and \( N_{f,\rho} = \mathbb{P} \cap \mathbb{P}_{f,\rho} \).

With a nonlinear selection function \( f \), the situation becomes more complicated. In general, both \( \sigma_{f,\rho} \) and \( \sigma_{f,\rho} \) are not compactly supported densities anymore. Thus, the update operator \( A_{f,\rho} : \rho \mapsto \langle \sigma_{f,\rho}, y \rangle / \langle f, y \rangle \rho \) may not even be defined on all densities from \( \mathbb{P} \setminus N_{f,\rho} \) or even from \( \mathbb{P}_{f,\rho} \setminus N_{f,\rho} \).

We will prove first that the operator in question is, indeed, well defined for some class of functions \( f \).

**Theorem 4** Let \( f \) be a continuous function. Let \( a \) and \( b \) be positive real numbers such that \( |f(x)| \leq a \) \( c h x \) for all \( x \in \mathbb{R} \). Then for any compactly supported probability density \( \rho \) and for any \( \lambda > 0 \) the expectations of \( \sigma_{f,\rho} \) and \( \sigma_{f,\rho} \) are finite.

**Proof** To prove the theorem we will show that the series involved in \( \langle \sigma_{f,\rho}, y \rangle \) and in \( \langle \sigma_{f,\rho}, y \rangle \) converge absolutely, namely that

\[
\sum_{n=0}^{\infty} \frac{e^{-\lambda} a^n}{(n+1)!} |\langle \sigma_{f,\rho}, y \rangle| < \infty, \quad \sum_{n=0}^{\infty} \frac{e^{-\lambda} a^n}{(n+1)!} |\langle \sigma_{f,\rho}, y \rangle| < \infty. (92)
\]

First, for any compactly supported probability density \( \nu \) we have \( |\langle \nu, f(x) \rangle| \leq \langle \nu, f(x) \rangle \). Then, using the estimate \( |f(x)| \leq a \) \( c h x \), the expression for the moment generating function \( \psi_{\rho}(s) \) def \( \psi_{\sigma_{f,\rho}}(s) = e^{as} \), and the fact that \( \psi_{\delta_{0}}(s) = e^{as} \), we obtain the estimates

\[
\sum_{n=0}^{\infty} \frac{e^{-\lambda} a^n}{(n+1)!} |\langle \sigma_{f,\rho}, y \rangle| \leq \sum_{n=0}^{\infty} \frac{e^{-\lambda} a^n}{(n+1)!} \left( e^{bx} \psi_{\rho}(b)^n + e^{-bx} \psi_{\rho}(-b)^n \right)
\]

\[
\leq \sum_{n=0}^{\infty} \frac{e^{-\lambda} a^n}{(n+1)!} b e^{bx} \psi_{\rho}(b)^n = a e^{bx} - \frac{\psi_{\rho}(b) - 1}{\lambda} < \infty, \quad (93)
\]
where \( \tilde{\psi}_\rho(x) = \max\left(\psi_\rho(x), \psi_\rho(-x)\right) \), and

\[
\sum_{n=0}^\infty \frac{e^{-\lambda} \lambda^n}{(n+1)!} \left| (\rho^{n+1}, f(y)) \right| \leq \sum_{n=0}^\infty \frac{e^{-\lambda} \lambda^n}{(n+1)!} \frac{a}{2} \left( \psi_\rho(b)(n+1) + \psi_\rho(-b)(n+1) \right)
\leq \sum_{n=0}^\infty \frac{e^{-\lambda} \lambda^n}{(n+1)!} a \tilde{\psi}_\rho(b)(n+1) = a e^{\lambda \tilde{\psi}_\rho(b)} - 1 \frac{\lambda e^\lambda}{\lambda e^\lambda} \leq \infty.
\] (94)

The last relations in the chains follow from the fact that for any compactly supported probability density \( \rho \) its moment generating function \( \psi_\rho \) is positive and finite for any value of the argument.

A counterexample to the theorem’s statement with the dropped condition \( |f(x)| \leq a \text{ch} \ b x \) is given by the function \( e^{x^2} \) and the density \( \delta_1 \). Indeed, in this case the expressions for \( \sigma_x \) and \( \sigma \) coincide and we have

\[
\langle \sigma_{e^{x^2}, \delta_1}, y \rangle = \langle \sigma_{e^{x^2}, \delta_1}, y \rangle = \sum_{n=0}^\infty \frac{e^{-\lambda} \lambda^n}{(n+1)!} e^{(n+1)^2}.
\] (95)

This series is divergent as its positive terms increase with the increase of \( n \).

The statement of theorem can be extended to a wider class of functions \( f \) than merely continuous (keeping the majorating condition). This requires a construction of the fully developed theory of integration for Radon measures, which is possible but is not of an interest in this work.

The next two theorem establish the continuity of \( A^f, \Sigma_x^f \), and \( \Sigma^f \).

**Theorem 5** For any interval \( \mathcal{I} \), under conditions of Theorem 4, the operator \( A^f \) is continuous on the subset \( \mathbb{P}^f \setminus N^f, \mathcal{I} \).

**Proof** The proof essentially repeats the proof of Theorem 3.

Let \( \rho_n \in \mathbb{P}^f \) be a sequence that approaches some \( \rho \in \mathbb{P}^f \) in \( \mathbb{P}^f \). We will prove that \( \langle \sigma_x^{f, \rho_n}, y \rangle \), as functions of \( x \), converge to \( \langle \sigma_x^{f, \rho}, y \rangle \) uniformly on \( \mathcal{I} \). The convergence \( \langle \sigma_x^{f, \rho_n}, y \rangle \to \langle \sigma_x^{f, \rho}, y \rangle \) is proven analogously. The both facts will imply that, if \( \rho \in \mathbb{P}^f \setminus N^f, \mathcal{I} \), then

\[
\frac{\langle \sigma_x^{f, \rho_n}, y \rangle}{\langle \sigma_x^{f, \rho_n}, y \rangle} \to \frac{\langle \sigma_x^{f, \rho}, y \rangle}{\langle \sigma_x^{f, \rho}, y \rangle}
\] (96)

uniformly on \( \mathcal{I} \), and thus, \( A^f(\rho_n) \to A^f(\rho) \) in \( \mathbb{P}^f \setminus N^f, \mathcal{I} \).

As \( \text{supp} \rho_n \subset \mathcal{I} \) and \( \text{supp} \rho \subset \mathcal{I} \), we have the pointwise convergence \( \psi_{\rho_n} \to \psi_\rho \). Indeed, for any \( t \in \mathbb{R} \) we have

\[
\psi_{\rho_n}(t) - \psi_\rho(t) = \langle \rho_n - \rho, e^{xt} \rangle = \langle \rho_n - \rho, \eta_t(x) \rangle \to 0,
\] (97)

where \( \eta_t \in C_{c+} \) such that \( \forall x \in \mathcal{I} \eta_t(x) = e^{xt} \).

\( \square \) Springer
Therefore, for any $n$, we have the following estimate

$$|\langle \delta_x \ast (\rho^*_n - \rho^*_k), f(y) \rangle| \leq a e^{b x} \left( \left( \sup_m \tilde{\psi}_{\rho_m}(b) \right)^k + \tilde{\psi}_\rho(b)^k \right)^k \leq a e^{b x} \left( \sup_m \tilde{\psi}_{\rho_m}(b) + \tilde{\psi}_\rho(b) \right)^k,$$

where the notation is the same as in the proof of Theorem 4.

Let us denote $c = \sup_m \tilde{\psi}_{\rho_m}(b) + \tilde{\psi}_\rho(b)$. It follows that for any $n$

$$\sum_{k=0}^{\infty} \frac{e^{-\lambda^k}}{(k+1)!} \sup_{\delta_x} |\langle \delta_x \ast (\rho^*_n - \rho^*_k), f(y) \rangle| \leq \sum_{k=0}^{\infty} \frac{a e^{b x} \lambda^k}{(k+1)!} \leq \sum_{k=0}^{\infty} \frac{a e^{b x} \lambda^k}{c \lambda} < \infty,$$

and therefore, for any $\epsilon > 0$ there is $k_0$ such that $\sum_{k=k_0}^{\infty} \frac{a e^{b x} \lambda^k}{(k+1)!} < \epsilon$, and, thus, for any $n$

$$\sum_{k=k_0}^{\infty} \frac{e^{-\lambda^k}}{(k+1)!} \sup_{\delta_x} |\langle \delta_x \ast (\rho^*_n - \rho^*_k), f(y) \rangle| < \epsilon.$$

From the other hand, as $\rho_n \to \rho$, for any $\epsilon > 0$ and any $k_0$ there is $n_0$ such that for any $n > n_0$

$$\sum_{k=0}^{k_0-1} \frac{e^{-\lambda^k}}{(k+1)!} \sup_{\delta_x} |\langle \delta_x \ast (\rho^*_n - \rho^*_k), f(y) \rangle| < \epsilon.$$

Therefore, by (90), it follows that for any $x$, $\langle \sigma_x^{F, \rho_n}, y \rangle \to \langle \sigma_x^{F, \rho}, y \rangle$.

For any $\rho \in \mathbb{P}$ with bounded support the function $\langle \delta_x \ast \rho, f(y) \rangle$ is continuous in $x$. Indeed, we have $\langle (\delta_x - \delta_x) \ast \rho, f(y) \rangle = \langle \rho, f(y + x') - f(y + x) \rangle$ and $f(y + x') \to f(y + x)$ uniformly on $\text{supp} \rho$ as $x' \to x$. Therefore, $\langle \sigma_x^{F, \rho_n}, y \rangle$ and $\langle \sigma_x^{F, \rho}, y \rangle$ are continuous in $x$ as absolutely convergent series of continuous functions. This, in turn, implies that $\langle \sigma_x^{F, \rho_n}, y \rangle \to \langle \sigma_x^{F, \rho}, y \rangle$ uniformly on $\mathcal{F}$ as functions of $x$. \hfill \Box

Note that $N^{F, \mathcal{F}}$ is nowhere dense in $\mathbb{P}^{\mathcal{F}}$ and $N^{F, \mathcal{F}}_p$ is nowhere dense in $\mathbb{P}^{\mathcal{F}}_p$. Indeed, this follows from the implication $\neg((\sigma^{F, \rho_n}, x) \to (\sigma^{F, \rho}, x)) \Rightarrow \neg(\rho_n \to \rho)$, which, in turn, is equivalent to the proven implication $\rho_n \to \rho \Rightarrow (\sigma^{F, \rho_n}, x) \to (\sigma^{F, \rho}, x)$. Note also that the statement of the theorem can be extended to $\mathbb{P}^K$, where $K$ is any compact set with nonempty interior. In the case when $\forall x > 0$, $f(x) > 0$, we have
either $N^f = \{\delta_0\}$ or $N^f = \emptyset$, so all these subtleties become irrelevant for the extensions by continuity of $A^f$ from $\mathbb{P}_p^\mathcal{F}$ to $\mathbb{P}^\mathcal{F}$.

**Theorem 6** Under conditions of Theorem 4, the operators $\Sigma^f_x$ and $\Sigma^f$ are continuous on $\mathbb{P}$.

**Proof** Note that for any $\varphi \in C_c$ we have $\sup_x |\varphi \left( \frac{f(x)}{n} \right) | \leq \sup_x |\varphi(x)|$. After that, the proof literally repeats the proof of Theorem 3. \hfill \square

**A.4 Polynomial selection function and sums of exponential with polynomial coefficients**

Let the total fitness in a compartment with $n$ individuals characterized by phenotypes $x_1, \ldots, x_n$ be given by $f(x_1 + \cdots + x_n)$, where

$$f(x) = a_0 + a_1 x + \cdots + a_m x^m. \quad (102)$$

As it is shown in Sect. 4.1, to find $\sigma_x$ and $\sigma$ it is enough to consider $(s_n^k)_x = \langle \delta_x * \rho^n, y^k \rangle$ and $s_n^k = \langle \rho^{n+1}, y^k \rangle$ for any $k$, $0 \leq k \leq m$, and then to find the sums $\sum_n \frac{P_n}{n+1} s_n^k$ and $\sum_n \frac{P_n}{n+1} s_n^k$, $P_n = e^{-\lambda} \frac{n^k}{n!}$.

Note that the operation of the multiplication of a generalized function $\rho$ by the parameter $(x_\rho)$ is a derivation on a convolution algebra, that is it is linear and obeys the Leibniz rule:

$$x(\rho_1 * \rho_2) = (x \rho_1) * \rho_2 + \rho_1 * (x \rho_2). \quad (103)$$

Indeed, for any $\varphi \in C_c$ we have

$$\langle x(\rho_1 * \rho_2), \varphi \rangle = (\rho_1 * \rho_2, x \varphi) = (\rho_1 \otimes \rho_2, (x_1 + x_2) \varphi(x_1 + x_2))$$

$$= (x_1 \rho_1 \otimes \rho_2 + \rho_1 \otimes (x_2 \rho_2), \varphi(x_1 + x_2)) = ((x \rho_1) * \rho_2 + \rho_1 * (x \rho_2), \varphi). \quad (104)$$

As $\langle \rho, y^m \rangle = x^m$ and $\langle \delta_x, y^m \rangle = x^m$, $(s_n^k)_x = \langle y^k(\delta_x * \rho^n), 1 \rangle$ is a linear combination of all expressions of the form $x^n \prod_i \hat{x}^{\beta_i} \hat{y}^{\gamma_i}$, where all $\beta_i$ are different and $\alpha + \sum_i (\beta_i + \gamma_i) = k$. For example, for $k = 2$ we have $x^2$, $x \hat{x}$, $\hat{x}^2$, $\overline{x_2}$, for $k = 3$ we have $x^3$, $x^2 \hat{x}$, $x \overline{x_2}$, $\hat{x}^2$, $\overline{x_3}$, $\overline{x_2}$, $\hat{x} \overline{x_2}$, $\overline{x_3}$, $\overline{x_2} \overline{x_2}$, $\hat{x} \overline{x_3} \overline{x_2} \overline{x_2}$, $\overline{x^4}$, $\overline{x^2 \overline{x_2}}$, $\overline{x^3}$, $\overline{x \overline{x_2}}$, $\overline{x^2 \overline{x_2}}$, $\overline{x^3} \overline{x^2}$, $\overline{x^4}$, $\overline{x^2 \overline{x_2}}$, $\overline{x^3}$, $\overline{x_2}$, $\overline{x^4}$, etc. These expressions enter $(s_n^k)_x$ with coefficients that are (nonnegative) polynomials in $n$ of the form $n(n-1) \ldots (n-l+1) a_l(k)$ or just $a_0(k)$ ($a_l(k) \in \mathbb{N}$). The polynomial (in $k$) functions $a_l(k)$ can be in principle found using some combinatorics. At the very least, they are algorithmically computable.
The sums $\sum_n P_n(s_n^k)x/(n + 1)$ can be evaluated using the identity

$$\sum_{n=0}^{\infty} \lambda^n \frac{n(n-1)(n-2)\ldots(n-p+1)}{n!} = \lambda^p \sum_{n=0}^{\infty} \frac{\lambda^n}{n!} \frac{1}{n + p + 1} = \lambda^p \frac{d}{d\lambda^p} \frac{e^\lambda - 1}{\lambda}. \quad (105)$$

Likewise, $s_n^k$, being $\langle \lambda^k \rho^{*n+1}, 1 \rangle$, is a linear combination of all expressions of the form $\prod_i x^{\beta_i} y_i$, where all $\beta_i$ are different and $\sum_i (\beta_i + \gamma_i) = k$. The coefficients in front these expressions in $s_n^k$ are of the form $(n + 1)n(n-1)(n-2)\ldots(n-p+1)$, $l \in \mathbb{N}$. The summation of $\sum_n P_n s_n^k/(n + 1)$ is trivial.

Let us now consider that the total fitness in a compartment with $n$ individuals with phenotypes $x_1, \ldots, x_n$ is given by $f(x_1 + \cdots + x_n)$, where $f$ is a linear combination of exponential functions with polynomial coefficients:

$$f(x) = \sum_{j=1}^{m} p_j(x)e^{a_j x}, \quad (106)$$

where $p_j$ are polynomials. It is again sufficient to find $\langle \delta_x \ast \rho^{*n}, y^k e^{a y} \rangle$ and $\langle \rho^{*n+1}, y^k e^{a y} \rangle$ for any $k$ and $a$. Note that $\langle x \rho, e^{ax} \rangle = \frac{d}{ds} \psi(s) \bigg|_{s=a}$, where $\psi(a) = \langle \rho, e^{ax} \rangle$ [this is directly related to (103)]. Therefore, $\langle \delta_x \ast \rho^{*n}, y^k e^{a y} \rangle = \frac{d^k}{ds^k} \psi(s^n) \bigg|_{s=a}$ and $\langle \rho^{*n+1}, y^k e^{a y} \rangle = \frac{d^k}{ds^k} \psi((s+1)^{n+1}) \bigg|_{s=a}$. These expressions can be summed with $e^{-\lambda^n/(n + 1)!}$ in the same way as in the pure polynomial case. Thus, every function $f$ of the form (106) allows to write the update equation in closed form.

### A.5 Approximation of $f$ using truncated Fourier series

We assume the Poisson distribution of the individuals in the compartments. We also assume that the phenotype-fitness relation is defined by a continuous selection function $f$. The activity is considered to be additive. By $\sigma_f^x, \rho$ and $\sigma_f^y, \rho$ we will denote the expressions (11) and (12), respectively, where $h_n : x \mapsto f(x)/n$, and we explicitly indicate the dependence on $f$ and $\rho$.

We will call a trigonometric polynomial of order $n$ and period $T$ any function from $\mathbb{R}$ to $\mathbb{C}$ of the form

$$p(x) = \sum_{k=-n}^{n} c_k e^{2\pi i k x/T}, \quad c_k \in \mathbb{C}, \quad c_{-n} c_n \neq 0. \quad (107)$$

The trigonometric polynomial $p$ is called real if $p(\mathbb{R}) \subset \mathbb{R}$, where the second $\mathbb{R}$ is understood as the natural embedding in $\mathbb{C}$. $p$ is real if and only if $\forall k \ c_{-k} = \bar{c}_k$. 

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Let $L(I)$ mean the length of the interval $I$. We call the truncated to order $n$ Fourier series of the function $f$ on the interval $I$ the trigonometric polynomial

$$f_n(x) = \sum_{k=-n}^{n} a_k e^{2\pi ikx/L(I)}, \quad a_k = \frac{1}{L(I)} \int_I f(x) e^{-\frac{2\pi ikx}{L(I)}} dx. \quad (108)$$

We need the following known fact.

**Theorem 7** For any $\epsilon > 0$ and any periodic continuous function $f$ with period $T$ there exists a trigonometric polynomial $p$ with period $T$ such that $\sup_x |f(x) - p(x)| < \epsilon$. Furthermore, $p$ can be constructed from the Fourier series of $f$, namely if $f_n$ is the truncated to order $n$ Fourier series of $f$, then, for large enough $n$, one can take $p = (f_0 + f_1 + \cdots + f_n)/(n+1)$.

The proof can be found, for example, in the book by Rudin (1976). The last statement is known as Fejér’s theorem. Note that all $p$ constructed in this way for a real function $f$ are real.

We start with an observation that for any probability density $\rho$ the following holds.

If $\text{supp } \rho \subset [-d, d]$, then $\text{supp } \rho^{*n} \subset [-nd, nd]$ and $\text{supp } \delta_x * \rho^{*n-1} \subset [-nd, nd]$ for any $x \in \text{supp } \rho$. For any $\rho$ we will denote $I^\rho$ some interval $I^\rho = [-d, d]$ such that $\text{supp } \rho \subset I^\rho$. For example, one can use the smallest interval with these properties. We will also introduce intervals $I^\rho_n = [-nd, nd]$ (not to be confused with $I_k$ used later) for the same $d$ that defines $I^\rho$.

We will prove the following main theorem.

**Theorem 8** Let $f$ be a continuous function $f: \mathbb{R} \to \mathbb{R}$ exponentially bounded in the following sense: There are positive numbers $a$ and $b$ such that $|f(x)| \leq a \, \text{ch} \, bx$ for all $x$. Then for any compactly supported probability density $\rho$ there exists a sequence $k \mapsto (I_k, p_k)$ of pairs of closed intervals $I_k$ and of real trigonometric polynomials $p_k$, where $p_k$ approximates $f$ on $I_k$, such that $\langle \sigma_{\lambda}^{p_k, \rho}, y \rangle \to \langle \sigma^{f, \rho}, y \rangle$ homogeneously on $\text{supp } \rho$ as a function of $x$, $\langle \sigma_{\lambda}^{p_k, \rho}, y \rangle \to \langle \sigma^{f, \rho}, y \rangle$ as a sequence of numbers, while $\sigma_{\lambda}^{p_k, \rho} \to \sigma^{f, \rho}$ for any $x \in \text{supp } \rho$ and $\sigma^{p_k, \rho} \to \sigma^{f, \rho}$ in the sense of generalized functions. $p_k$ can be constructed using Fourier series approximations of $f$ on the appropriate intervals.

As the logic of the proof is slightly convoluted, we will first formulate and prove two auxiliary lemmas.

**Lemma 2** Under conditions of Theorem 8, for any $\epsilon > 0$ there exists $n_0 > 0$ such that for any function $p$ that is bound by $\sup_{x \in \mathbb{R}} |p(x)| < \sup_{x \in I^\rho_0} |f(x)| + \epsilon$ the following holds:

$$\forall x \in \text{supp } \rho \quad \left| \sum_{n=n_0}^{\infty} \frac{e^{-\lambda n}}{(n+1)!} \langle \delta_x * \rho^{*n}, f - p \rangle \right| < \epsilon \quad \text{and} \quad (109)$$

$$\left| \sum_{n=n_0}^{\infty} \frac{e^{-\lambda n}}{(n+1)!} \langle \rho^{*n+1}, f - p \rangle \right| < \epsilon. \quad (110)$$
Proof By the virtue of the estimate on $f$ from the conditions of Theorem 8, the relations of the statement of the lemma follow from the relations

$$\forall x \in \text{supp } \rho \sum_{n=n_0}^{\infty} \frac{e^{-\lambda \lambda n}}{(n+1)!} \langle \delta_x * \rho^{*n}, |f| + a \text{ ch } (b L(I_{n_0}^\rho)) \rangle < \epsilon \quad \text{and (11)}$$

$$\sum_{n=n_0}^{\infty} \frac{e^{-\lambda \lambda n}}{(n+1)!} (\rho^{*n+1}, |f| + a \text{ ch } (b L(I_{n_0}^\rho))) < \epsilon. \quad (112)$$

Let us prove (111). Indeed, using the same reasoning as in the proof of Theorem 4, we have $\forall x \in \text{supp } \rho$

$$\sum_{n=n_0}^{\infty} \frac{e^{-\lambda \lambda n}}{(n+1)!} \langle \delta_x * \rho^{*n}, |f(y)| + a \text{ ch } (b L(I_{n_0}^\rho)) + \epsilon \rangle$$

$$\leq \sum_{n=n_0}^{\infty} \frac{e^{-\lambda \lambda n}}{(n+1)!} \langle \delta_x * \rho^{*n}, a \text{ ch } (b y) + a \text{ ch } (b L(I_{n_0}^\rho)) + \epsilon \rangle$$

$$< \sum_{n=n_0}^{\infty} \frac{e^{-\lambda \lambda n}}{(n+1)!} \left( a e^{b|x|} \psi_{\rho}(b)^n + a \left( e^{L(I^\rho)} \right)^n_0 + \epsilon \right)$$

$$\leq a \sum_{n=n_0}^{\infty} \frac{e^{-\lambda \lambda n}}{(n+1)!} e^{b x_0} \psi_{\rho}(b)^n + a \sum_{n=n_0}^{\infty} \frac{e^{-\lambda \lambda n}}{(n+1)!} \left( e^{L(I^\rho)} \right)^n + \epsilon \sum_{n=n_0}^{\infty} \frac{e^{-\lambda \lambda n}}{(n+1)!}, \quad (113)$$

where $\psi_{\rho}(t) = \langle \rho, e^{\lambda t} \rangle, \tilde{\psi}_{\rho}(t) = \max \left( \psi_{\rho}(t), \psi_{\rho}(-t) \right)$, and $x_0 = \max(| \inf \text{supp } \rho |, | \sup \text{supp } \rho |)$.

The statement (111) follows from the fact that the series in the last expression have positive terms and converge to finite numbers, if summed started from $n = 0$. Indeed, this means that for any $\epsilon > 0$ there is $n_0$ such that the last expression is smaller than $\epsilon$. It can be proven analogously that the same $n_0$ fulfills the statement (112).

Lemma 3 Under conditions of Theorem 8, for any $\epsilon > 0$ and any $n_0 > 0$ there exists a real trigonometric polynomial $p$ such that $\sup_{x \in I_{n_0}^\rho} |f(x) - p(x)| < \epsilon$, $\sup_{x \in \mathbb{R}} |p(x)| < \sup_{x \in I_{n_0}^\rho} |f(x)| + \epsilon$, as well as

$$\forall x \in \text{supp } \rho \sum_{n=0}^{n_0-1} \frac{e^{-\lambda \lambda n}}{(n+1)!} \langle \delta_x * \rho^{*n}, f - p \rangle < \epsilon \quad \text{and (144)}$$

$$\sum_{n=0}^{n_0-1} \frac{e^{-\lambda \lambda n}}{(n+1)!} \langle \rho^{*n+1}, f - p \rangle < \epsilon. \quad (115)$$

Proof Let $d$ be the number such that $I^\rho = [-d, d]$. For any $n \leq n_0 - 1$, we have $\text{supp } \delta_x * \rho^{*n} \subset I_{n_0}^\rho$ and $\text{supp } \rho^{*n+1} \subset I_{n_0}^\rho$. Let us extend $f |_{I_{n_0}^\rho}$ to $I_{n_0+1}^\rho$ by a function
Let \( \hat{f} \) such that \( \hat{f}(x) = \alpha x + \beta_1 \) for any \( x \in \left[ -(n_0 + 1)d, -n_0 df \right] \) and \( \hat{f}(x) = \alpha x + \beta_2 \) for any \( x \in [n_0 d, (n_0 + 1)d] \), where \( \alpha, \beta_1, \) and \( \beta_2 \) are selected to fulfill

\[
\hat{f}'(-(n_0 + 1)d) = \hat{f}'((n_0 + 1)d) = \frac{f(-n_0 d) + f(n_0 d)}{2},
\]

\[
\hat{f}'(-n_0 d) = f(-n_0 d), \quad \hat{f}(n_0 d) = f(n_0 d). \tag{116}
\]

Function \( \hat{f} \) is continuous on \( I_{n_0+1}^\rho \) and can be extended to the whole \( \mathbb{R} \) as a periodic continuous function \( \tilde{f} \) with period \( L(I_{n_0+1}^\rho) \). By Theorem 7, using the truncations of the Fourier series for \( \tilde{f} \) up to some order, we can construct a real trigonometric polynomial \( p \) such that for any \( \epsilon > 0 \) \( \sup_{x \in \mathbb{R}} |\hat{f}(x) - p(x)| < \epsilon \). It follows that for any \( \epsilon > 0 \) we can find \( p \) such that \( \sup_{x \in I_{n_0}^\rho} |f(x) - p(x)| < \epsilon \). Furthermore, by construction, \( \sup_{x \in \mathbb{R}} |p(x)| < \sup_{x \in I_{n_0}^\rho} |f(x)| + \epsilon \).

Let us consider the right-hand side of the statement (114). For any \( x \in \supp \rho \) we have

\[
\sum_{n=0}^{n_0-1} e^{-\lambda n} \frac{\lambda^n}{(n+1)!} \langle \delta_x * \rho^{*n}, f - p \rangle < \epsilon \sum_{n=0}^{n_0-1} e^{-\lambda n} \frac{\lambda^n}{(n+1)!} < \epsilon g(\lambda) \leq \epsilon. \tag{117}
\]

Here we essentially used the inclusion \( \supp \delta_x * \rho^{*n} \subset I_{n_0}^\rho \) for any \( x \in \supp \rho \). The relation (117) implies the relation (114).

The statement (115) can be proven analogously, using the fact that \( \supp \rho^{*n+1} \subset I_{n_0}^\rho \) for any \( n < n_0 \).

Proof of Theorem 8 Let us symbolically rewrite the statement of Lemma 2 as \( \forall \epsilon > 0 \exists \rho: A(\epsilon, n_0, p) \). In the same manner, let us symbolically rewrite the statement of Lemma 3 as \( \forall \epsilon > 0 \exists n_0 > 0 \exists \rho: B(\epsilon, n_0, p) \). Then (90) implies \( \forall \epsilon > 0 \exists n_0 > 0 \exists \rho: A(\epsilon, n_0, p) \land B(\epsilon, n_0, p) \). As the statement \( A \) estimates the sums from 0 to \( n_0 - 1 \), and the statement \( B \) estimates the sums from \( n_0 \) to \( \infty \), the statement \( A \land B \) gives estimates on the sums from 0 to \( \infty \). Therefore, the consequence of the above is explicitly read as following. For any positive \( \epsilon \) there exists a number \( n_0 \) and a trigonometric polynomial \( p \) (which can be constructed based on an approximation of \( f \) by truncated Fourier series, as in the proof of Lemma 3) such that \( \sup_{x \in I_{n_0}^\rho} |f(x) - p(x)| < \epsilon \) and

\[
\forall x \in \supp \rho \quad \left| \sum_{n=0}^{\infty} e^{-\lambda n} \frac{\lambda^n}{(n+1)!} \langle \delta_x * \rho^{*n}, f - p \rangle \right| < \epsilon \quad \text{and} \tag{118}
\]

\[
\left| \sum_{n=0}^{\infty} e^{-\lambda n} \frac{\lambda^n}{(n+1)!} \langle \rho^{*n+1}, f - p \rangle \right| < \epsilon. \tag{119}
\]

The part of the theorem with \( \langle \sigma_x^{p_k, \rho}, y \rangle \to \langle \sigma_x^{f, \rho}, y \rangle \) and \( \langle \sigma_{x}^{p_k, \rho}, y \rangle \to \langle \sigma_{x}^{f, \rho}, y \rangle \) is proven, if we take, for example, \( \epsilon_k = 1/(1 + k), k \in \mathbb{N} \), and if we take as \( I_k \) the
interval $I_{n_0}$ and as $p_k$ the trigonometric polynomial $p$, where $n_0$ and $p$ are provided by the above statement for $\epsilon = \epsilon_k$.

Let us choose any $\varphi \in C_c$. Let us denote $\Phi \overset{\text{def}}{=} \sup x |\varphi(x)|$ and $s_k \overset{\text{def}}{=} \sup_{x \in I_k} |\varphi (p_k(x)) - \varphi (f(x))|$. As $\varphi$ is continuous and compactly supported it is also uniformly continuous on $\mathbb{R}$. Therefore, we have $s_k \rightarrow 0$, as $k \rightarrow \infty$. Furthermore, for any $n$ we have

$$\sup_{x \in I_k} \left| \varphi \left( \frac{p_k(x)}{n+1} \right) - \varphi \left( \frac{f(x)}{n+1} \right) \right| \leq s_k$$ \hspace{1cm} (120)

and

$$\sup_{x} \left| \varphi \left( \frac{p_k(x)}{n+1} \right) - \varphi \left( \frac{f(x)}{n+1} \right) \right| \leq 2\Phi.$$ \hspace{1cm} (121)

Using the same technique of splitting the series into two parts by $n_0(k) = n_0(\epsilon_k)$ and using the former estimate for the initial part of the sum of the series and the latter estimate for the rest of the series by the same logic and taking into account that $n_0(\epsilon)$ provided by the Lemma 2 ever grows with the decay of $\epsilon$, we conclude that

$$|\langle \sigma_{pk}^{p_k,\rho} - \sigma_{k}^{f,\rho}, \varphi \rangle| \leq s_k + 2\Phi e^{-\lambda} e_{n_0}(\lambda) \rightarrow 0, \quad k \rightarrow \infty,$$ \hspace{1cm} (122)

where $e^\infty_m(x) \overset{\text{def}}{=} \sum_{j=m}^{\infty} \frac{x^j}{j!}$.

In the same way we prove $\sigma_{pk}^{p_k,\rho} \rightarrow \sigma_{f}^{f,\rho}$. \hfill $\square$

This theorem implies that for any compactly supported $\rho$ with $\langle \sigma_{f}^{f,\rho}, x \rangle \neq 0$ we have the convergence $A_{pk}(\rho) \rightarrow A_{f}(\rho)$, as $k \rightarrow \infty$. Note that the sequence $p_k$ depends on $\rho$. This is due to the involvement of $\psi_{\rho}$ in (113). However, if $f$ is a bounded function, then this dependence can be dropped from (113), and the choice of $n_0$, and thus of $p_k$, becomes independent of the current distribution. In this case we can state the pointwise convergence $A_{pk} \rightarrow A_{f}$ (on $\mathbb{P}_K$ for some compact $K$). The proof of the theorem is constructive. However, it is not optimized for applications.

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