A MINIMAL SELF-ORGANIZATION MODEL OF THE GOLGI APPARATUS

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ABSTRACT

The design principles dictating the spatio-temporal organization of eukaryotic cells, and in particular the mechanisms controlling the self-organization and dynamics of membrane-bound organelles such as the Golgi apparatus, remain elusive. Although this organelle was discovered 120 years ago, such basic questions as whether vesicular transport through the Golgi occurs in an anterograde (from entry to exit) or retrograde fashion are still strongly debated. Here, we address these issues by studying a quantitative model of organelle dynamics that includes: *de-novo* compartment generation, inter-compartment vesicular exchange, and biochemical maturation of membrane components. We show that purely anterograde or retrograde vesicular transports are asymptotic behaviors of a much richer dynamical system. Indeed, the structure and composition of cellular compartments and the directionality of vesicular exchange are intimately linked. They are emergent properties that can be tuned by varying the relative rates of vesicle budding, fusion and component maturation.

Keywords Organelles · Golgi Apparatus · Self-organization · Stochastic models

1 Introduction

The Golgi apparatus is an intracellular organelle at the crossroad of the secretory, lysosomal and endocytic pathways [1]. One of its most documented functions is the sorting and processing of many proteins synthesized by eukaryotic cells [2]. Proteins translated in the endoplasmic reticulum (ER) are addressed to the Golgi, where they undergo post-translational maturation and sorting, before being exported to their final destination. During this process, they interact in a predetermined order with Golgi resident enzymes. This is made possible by the peculiar organization of the Golgi, which is composed of distinct sub-compartments, called cisternae, of different biochemical identities [3]. From the ER, cargoes successively reside in cisternae of the *cis*, *medial* and *trans*-identities, after which they exit the Golgi via the *trans*-Golgi network (TGN). In some organisms such as the yeast Saccharomyces cerevisiae, cisternae are dispersed throughout the cell and each cisterna undergoes maturation from a *cis* to a *trans*-identity [4]. In most other eukaryotes, cisternae are stacked in a polarized fashion, with cargoes entering via the *cis*-face and exiting via the *trans*-face [5]. This polarity is robustly conserved over time, despite cisternae constantly exchanging vesicles with each other, the ER and the TGN [2].

The highly dynamical nature and compact structure of a stacked Golgi makes it difficult to determine how cargoes are transported from the *cis* to *trans*-side of the stack and how this transport is coupled to processing. Two major classes of models are proposed [6]. "Vesicular transport" models postulate that cisternae have fixed identities and cargo progresses from one cisterna to the next via anterograde vesicular transport. "Cisternal Maturation" models postulate that cisternae themselves undergo maturation from the *cis* to the *trans*-identity and physically move through the stack, while Golgi

resident proteins remain in the stack by moving toward the *cis*-face by retrograde vesicular transport. Considerable work has been done to resolve this issue [7], but the mechanism dictating the Golgi's organization and dynamics is still an open question. The fact that different studies show contradictory evidences supporting either model [8, 9, 10] suggests that Golgi dynamics might be more versatile, and rely on a combination of anterograde and retrograde transport and maturation, possibly depending on the type of cargo. Although there is a large diversity in Golgi structures and dynamics among different species, the organelle's physiological function as a sorting and processing hub is common to all species, suggesting that important mechanisms controlling Golgi dynamics are conserved. Works in evolutionary biology and biophysics have attempted to describe these mechanisms [11, 12]. Different classes of mathematical models have been proposed, from models of vesicle budding and fusion based on rate equations [13, 14, 15] - some specifically including space [16], to continuous, discrete and stochastic models of protein sorting in the Golgi [17, 18] and computer simulations based on membrane mechanics [19]. To account for the Golgi's ability to reassemble after mitosis [20], many of these studies have sought to describe the Golgi as a self-organized system [13, 21]. However, the manner in which the kinetics of the Golgi two main functions (namely the transport and biochemical maturation of its components) interplay with one another to yield a robust steady-state has so far received much less attention [16, 18].

We propose here a model in which both Golgi self-organization and vesicular transport are solely directed by (local) molecular interactions resulting in composition-dependent budding and fusion. Thus, the directionality of vesicular transport is an emergent property of the self-organization process that co-evolves with the size and number of Golgi sub-compartments, rather than being fixed by arbitrary rules. This approach brings new conceptual insights that highlight the link between the large-scale steady-state organization of the Golgi, the directionality of vesicular transport, and the kinetics of individual processes at the molecular scale. It shows that a wide variety of organelle phenotypes can be observed upon varying the rates of the local processes. Besides, it emphasizes how direct interactions between compartments yield complex vesicular fluxes that correspond neither to the "Cisternal Maturation" or "Vesicular transport" models.

2 Methods

The model of organelle self-organization is shown in Figure 1 (Fig.1), introduced below, and fully described in the Supporting Information (SI). The building blocks are vesicles that can be of three identities, corresponding to the *cis*, *medial* and *trans*-identities of the Golgi. A compartment is an assembly of fused vesicles. The identity of a membrane patch is defined in a very broad sense by its composition in lipids and proteins that influence its interaction with other patches, such as RAB GTPases [22] or fusion proteins such as SNAREs [23]. We describe compartments' dynamics using only three mechanisms - budding, fusion and maturation - defined below:

- **Budding**: Composition-dependent vesicular budding is an important sorting mechanism in cellular traffic [24]. Here we adopt a non-linear budding scheme that relies on both the compartment's size and its contamination state, which promotes efficient sorting [25].
- **Fusion**: Homotypic fusion - the higher probability of fusion between compartments of similar identities - is a feature of cellular organelles in general [26] and the Golgi apparatus in particular [27, 28]. This is controlled here by a fusion rate between compartments proportional to the probability that they present the same identity at the contact site. We stress that this homotypic fusion mechanism (relying on local interactions), allows vesicular transport between compartments of different identities - a process that may be regarded as heterotypic fusion [7]. It merely requires that the receiving compartment contains some membrane patches of identity similar to that of the emitted vesicle.
- **Maturation**: Each membrane patch undergoes stochastic maturation from a *cis* to a *medial* to a *trans*-identity, in agreement with *in vivo* observations [8, 9]. This is a local mechanism of identity conversion, consistent with processes such as the RABs cascade [22, 27]. It is distinct from the maturation of entire compartments, which is also affected by the dynamics of budding and fusion.

These mechanisms of compartment self-organization are applied to an open system that interacts with two boundaries: the ER, from which newly synthesized material is injected in the system, and the TGN, through which processed material can leave the system. Vesicles and entire compartments can exit the system by homotypic fusion with either boundaries, following the rules described above. The role of the boundaries' composition is discussed in the SI, where a parameter α (varying between 0 and 1) is introduced to characterize the fraction of *cis* (*trans*) identity in the ER (TGN).

In its simplest form, our model contains only four parameters: the rates of injection, fusion, budding, and maturation ($K_{\mathrm{i}},\ K_{\mathrm{f}},\ K_{\mathrm{b}},\ K_{\mathrm{m}}$). By normalizing time with the fusion rate, we are left with three parameters: $K = K_{\mathrm{b}}/K_{\mathrm{f}}$, $k_{\mathrm{m}} = K_{\mathrm{m}}/K_{\mathrm{f}}$ and $J = K_{\mathrm{i}}/K_{\mathrm{f}}$. The systems' dynamics is entirely governed by these stochastic transition rates, and can be simulated exactly using a Gillespie algorithm [29]

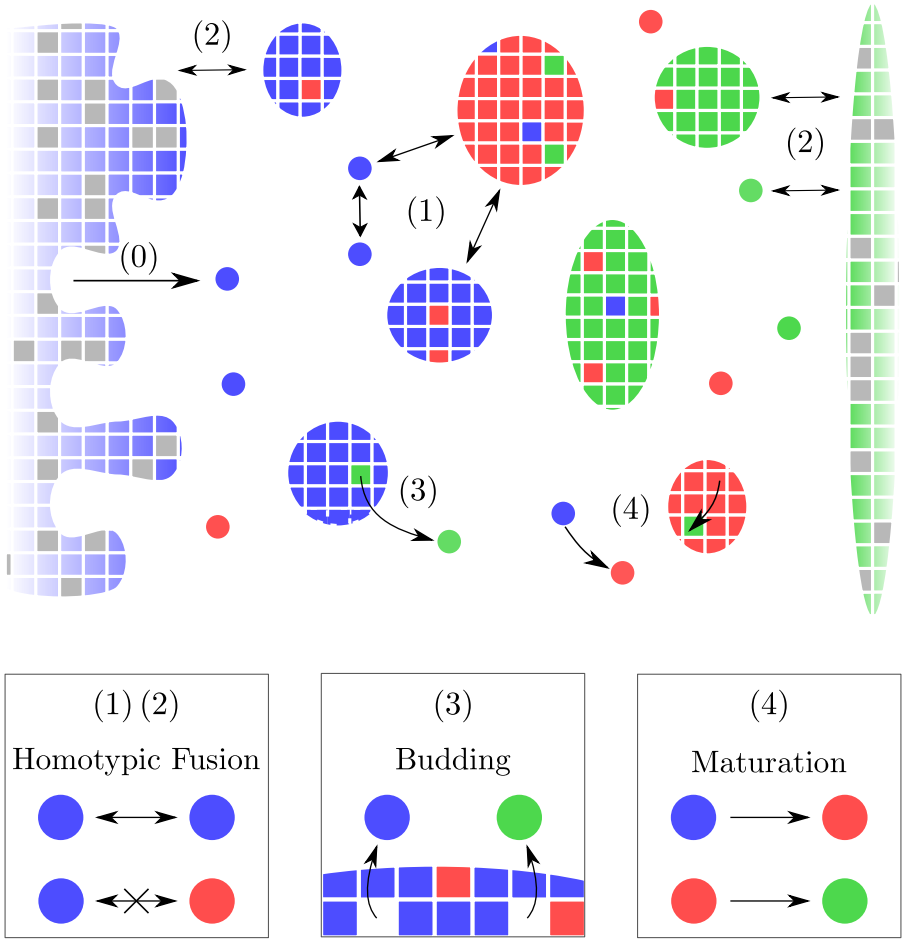

Figure 1: **Model of self-organization of the Golgi apparatus.** The system includes three membrane identities (*cis*: blue, *medial*: red, *trans*: green). It self-organizes between two boundaries: the ER (left) is composed of a *cis*-membrane identity, and the TGN (right) is composed of a *trans*-membrane identity. Golgi's compartments self-organize *via* three stochastic mechanisms: **Fusion**: (1) All compartments can aggregate using homotypic fusion mechanisms: the more identical they are, the higher the fusion rate. (2) Each compartment can exit the system by fusing homotypically with one of the two boundaries. **Budding:** (3) Each compartment larger than a vesicle can create a vesicle by losing a patch of membrane. **Maturation:** (4) Each patch of membrane matures from a *cis* to a *trans*-identity. New *cis*-vesicles (0) bud from the ER at a constant rate. In the sketch, the boundaries also contain neutral (gray) membrane species that dilute their identity (impact of this dilution in Fig.S6 - SI).

3 Results

The steady-state organization and dynamics of the system is described in terms of the average size and purity of compartments, and the directionality of vesicular transport between them (introduced here and detailed in the SI). The typical compartment's size is defined as the ratio of the second over the first moments of the size distribution [30]. The purity of a given compartment is defined as the (normalized) Euclidean distance of the compartment's composition (the fractions ϕ_i in the different i-identities, $i =$ *cis*, *medial* and *trans*) from a perfectly mixed composition. $P = 0,\ 1/2,\ 1$ correspond respectively to compartments that contain the same amount of the three identities, the same amount of two identities, and a single identity (Fig.S2- SI). The system's purity is the average purity of each compartment weighted by its size, ignoring vesicles. Finally, to characterize the directionality of vesicular transport, the dynamics of passive cargoes injected from the ER is followed over time. Each cargo in a compartment of size n has a probability $1/n$ to join a vesicle budding from this compartment. When this vesicle fuses with another compartment or a boundary, the composition difference $\Delta\phi_i$ between the receiving and emitting compartment (a number between -1 and 1) is recorded for the three i-identities ($i =$*cis*, *medial*, *trans*). Averaged over all budding/fusion events, this defines the *enrichment vector* $\vec{E}$, whose three components E_i are normalized for readability so that $\sum_i |E_i| = 1$ (see Fig.S5 - SI

for non-normalized enrichment). Vesicular flux is anterograde if $E_{cis} < 0$ and $E_{trans} > 0$, while it is retrograde if $E_{cis} > 0$ and $E_{trans} < 0$.

3.1 Steady-state organization

The compartment size distribution shows a power law with an exponential cutoff (Fig.2A), as expected for a non-equilibrium steady-state controlled by scission and aggregation. With our definition, the characteristic compartment size is close to half the cut-off size (SI). The way it depends on the model parameters is shown in Fig.2B. Increasing

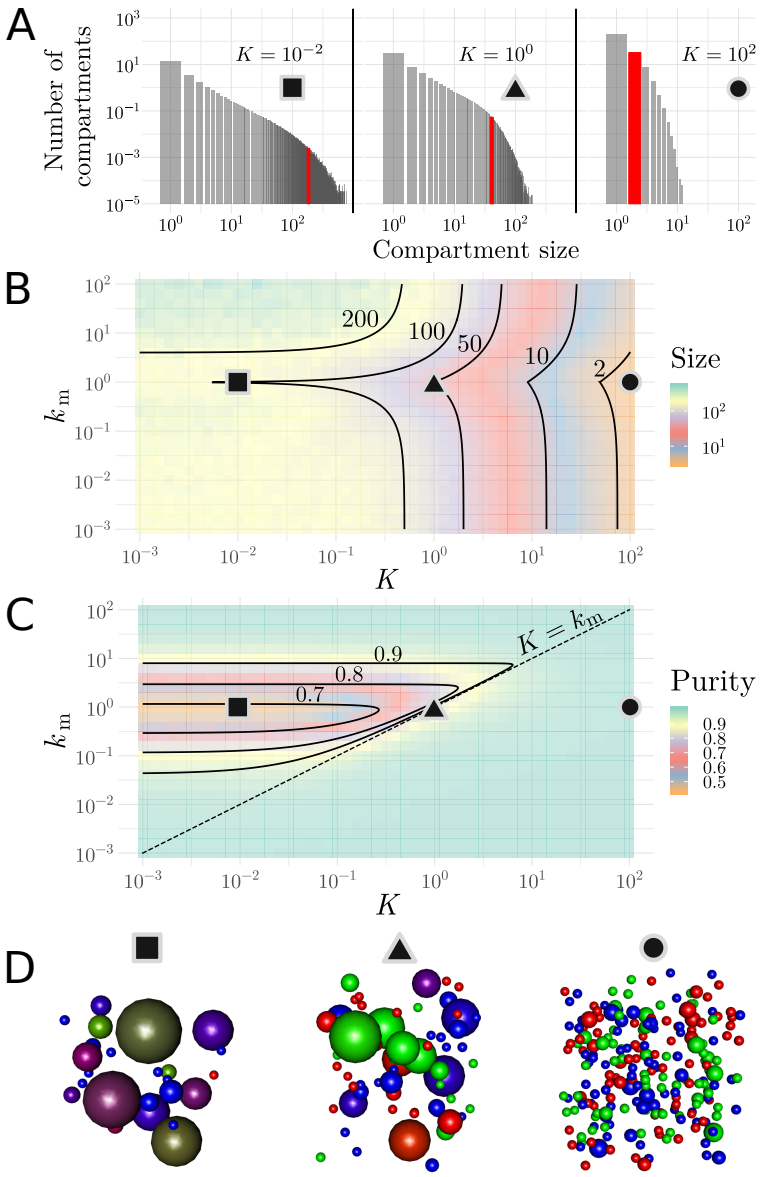

Figure 2: **Steady-state of the self-organized model of Golgi apparatus.** (A) Size distribution of compartments for a maturation rate $k_m = 1$ and different values of the budding rate K. The red bar shows the characteristic compartment size. (B) Phase diagram of the compartments' size as the function of K and k_m. Black lines: theoretical prediction (introduced in text, developed in SI). (C) Phase diagram of the system's purity as the function of K and k_m. Dashed line is $k_m = K$, black lines are a theoretical prediction (see Fig.S3 - SI). (D) System snapshots for $k_m = 1$ showing the *mixed regime* (square - $K = 10^{-2}$), the *sorted regime* (triangle - $K = 1$), and the *vesicular regime* (circle - $K = 10^2$) - see text. See also Fig.S4 for further characterizations of the steady-state organization, Fig.S6 for the role of exits' composition, and Fig.S7 for results with alternative budding and fusion kinetics.

the ratio of budding to fusion rate K decreases the compartment size [31]. The size depends on the maturation rate k_{m}, which controls the compartment's composition and hence the fusion probability, in a non-monotonic manner: for a given budding rate, the size is smallest for intermediate values of the maturation rate ($k_{\mathrm{m}} \simeq 1$). This is well explained by a simple analytical model (discussed in SI) that approximates the system by three pure compartments interacting with a pool of vesicles (solid lines in Fig.2B). The system is mostly composed of *cis* and *medial*-components for low maturation rates and of *trans*-components for high maturation rate. If compartments are well sorted, the mean compartment size is limited by the amount of each species in the system, which is smallest when the three identities are in equal amount, that is when $k_{\mathrm{m}} \simeq 1$.

The dependency of the compartments' purity with the parameters is shown in Fig.2C. As for the compartments' size, it is non-monotonic in the maturation rate, and regions of lowest purity are found for intermediate maturation rates $k_{\mathrm{m}} \simeq 1$, when all three identities are in equal amount. The purity increases with the budding rate K in a sigmoidal fashion (see Fig.3). This can be understood by analyzing the processes involved in mixing and sorting of different identities. In a first approximation, mixing occurs by maturation and sorting occurs by budding of contaminating species, suggesting a transitions from low to high purity when the budding rate reaches the maturation rate ($K \simeq k_{\mathrm{m}}$). A "purity transition" is indeed observed in an intermediate range of maturation rates ($0.1 < k_{\mathrm{m}} < 10$, see Fig.2C). It is most pronounced for $k_{\mathrm{m}} \simeq 1$. Beyond this range, the system is dominated by one identity and compartments are always pure. The purity variation can be qualitatively reproduced by an analytical model (see Fig.S3- SI) that accounts for the competition between maturation and budding, but also includes fusion between compartments and with the exits (solid lines in Fig.2C.).

In summary, three types of organization can be found, mostly controlled by the ratio of budding over fusion rate K: a *mixed regime* at low K, where compartments are large and contain a mixture of identities, a *vesicular regime* at high K, where compartments are made of only a few vesicles and are very pure, and a *sorted regime* for intermediate values of K, where compartments are both rather large, and rather pure. Snapshots of these three steady-states are shown in Fig.2D. Non-linear budding is essential to the existence of the intermediate *sorted* regime. With a linear budding scheme (Fig.S7 - SI), the purity transition occurs for larger values of the budding rate K, where compartments are already small.

3.2 Vesicular Transport

We compute the mean enrichment in *cis*, *medial* and *trans*-identities during vesicular exchange from a donor to a receiving compartment. We focus on systems containing comparable amounts of each species at steady-state ($k_{\mathrm{m}} = 1$ - SI). The enrichment in *cis*, *medial* and *trans*-identities are shown in Fig.3 as a function of the budding rate K. The most striking result is the high correlation between compartment's purity and the directionality of vesicular transport. For low values of the budding rate ($K \ll 1$), compartments are well mixed (purity $\simeq 1/2$) and the vesicular flux is retrograde, with a gain in *cis*-identities and a loss in *trans*-identities. For high values of the budding rate ($K \gg 1$), compartments are pure (purity $\simeq 1$) and the vesicular flux is anterograde, with a gain in *trans*-identities and a loss in *cis*-identities. The archetypal behaviors most often discussed in the literature are thus, within the limits of our model, asymptotic regimes for extreme values of the ratio of budding to fusion rates. Remarkably, the cross over between these two asymptotic behaviors is rather broad ($K \sim 0.1$) and displays a more complex vesicular transport dynamics, mostly oriented toward medial compartments.

In our model organelle, vesicle budding and fusion are biased solely by local compositions, which is subjected to irreversible biochemical maturation. The interplay between these microscopic processes gives rise to an irreversible flux of vesicles across the system. To better understand the directionality of vesicular exchanges, we represent on Fig.4 the vesicular fluxes between compartments as a vector field on the compartments' composition space. This is shown as a triangle plots in which each point corresponds to a fraction of *cis* (100% at the top), *medial* (100% at the bottom right) and *trans* (100% at the bottom left) components in a compartment (Fig.S2 - SI). The net vesicular flux leaving compartments with a given composition is represented as a vector field. The variation of the vesicular flux with the budding rate K (Fig.4) is consistent with that of the composition enrichment in the different identities (Fig.3). As K increases, the flux evolves from being mostly retrograde at low K (from *trans*-rich toward less mature compartments) to being anterograde at high K (from *cis* to *medial*, and *medial* to *trans*-compartments). In between, the vesicular flux is centripetal toward mixed compartments, leading to a net enrichment in *medial*-identity. The relationship between structure and transport is explained below, and a dissection of this fluxes with respect to the vesicles' composition is shown in Fig.S5 - SI. The net flux leaving a particular region of the triangular phase space is proportional to the total mass (number of compartments times their size) with this composition, while the flux arriving at a particular region depends on the number of compartments with that composition. Both quantities are shown on Fig.4.

At low budding rate ($K \ll 1$ - top row in Fig.4), the system is in the mixed regime, where compartments are large (hundreds of vesicles in size) but mixed (purity $\sim 1/2$). The system is dominated by maturation, and the

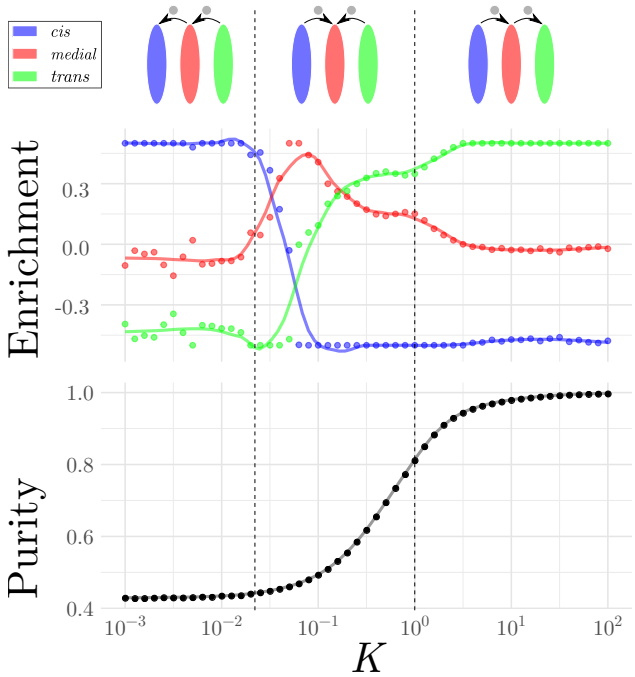

Figure 3: **Directionality of vesicular transport,** displayed varying the budding rate K for a maturation rate $k_{\mathrm{m}} = 1$ (equal amount of all species in the system). Middle panel: normalized enrichment in *cis* (blue), *medial* (red) and *trans* (green) identities between the acceptor and donor compartments during vesicular exchange (solid lines are guides for the eyes). See Fig.S5 for non-normalized fluxes. Top panel: sketches showing the direction of vesicular transport consistent with these data. Bottom panel: system purity for the same parameters. Before the purity transition (low values of K - purity ~ 0.5) the vesicular flux is retrograde. After the transition (high values of K - purity ~ 1) the vesicular flux is anterograde. Around the crossover ($K \sim 0.1$), the vesicular flux is centripetal and oriented toward *medial*-compartments. The centripetal flux disappears if inter-compartments fusion is prohibited - see Fig.S7.

compartment distribution is spread around a path going from purely *cis* to purely *trans*-compartments, consistent with a dynamics where each compartment maturates independently from the other ($\phi_A = e^{-k_{\mathrm{m}}t}$, $\phi_B = k_{\mathrm{m}} t e^{-k_{\mathrm{m}}t}$ and $\phi_C = 1 - \phi_A - \phi_B$, where t is the typical age of the compartment - see top left panel of Fig.4 and SI). The majority of the transport vesicles are emitted by *trans*-rich compartments, which concentrate most of the system's mass, leading to a retrograde vesicular flux.

At high budding rate ($K \gg 1$ - bottom row in Fig.4), the system is in the vesicular regime, where compartments are fairly pure, but rather small, with a size equivalent to a few vesicles. Compartments are exclusively distributed along the *cis/medial* and *medial/trans*-axes of composition, and accumulate at the triangle's apexes. Membrane patches that just underwent maturation bud quickly from the donor compartment and fuse with pure compartments with the appropriate *medial* or *trans*-identity, explaining the clear anterograde vesicular transport. This feature is further reinforced under very high budding rate, where the vesicular flux is dominated by vesicles undergoing maturation *after* budding, which prohibits their back fusion with the donor compartment.

In the intermediate regime (middle row in Fig.4), the distribution of mixed compartments is more homogeneous along the line describing the maturation of individual compartments - both in terms of mass and number of compartments. *Cis*-rich and *trans*-rich compartments emit comparable amount of transport vesicles, while large *medial*-rich compartments are absent. This leads to a centripetal vesicular flux with an enrichment in *medial* identity (see Fig.3). The lack of large *medial*-rich compartment is due to the fact that they can be contaminated by either *cis* or *trans*-species and fuse with *cis*-rich or *trans*-rich compartments. If inter-compartment fusion is prohibited, the enrichment in medial-identity in the intermediate regime is abolished, with a direct transition from anterograde to retrograde vesicular transport upon increasing K (see Fig.S7 - SI and the Discussion section).

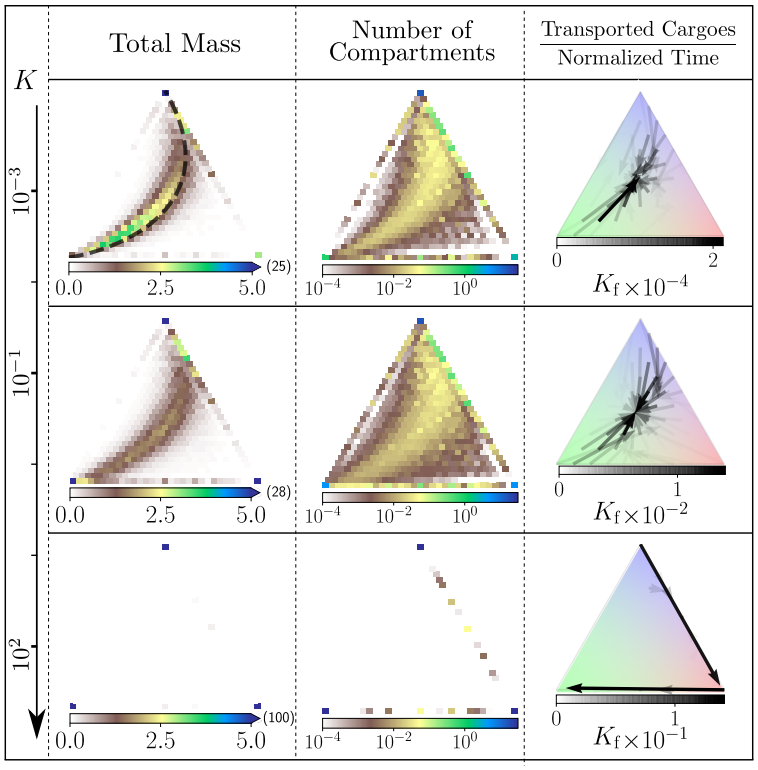

Figure 4: **Relationship between the system's structure and the vesicular fluxes.** Distribution of the total mass (truncated scale, maximum value in brackets) and number of compartments, and the vesicular flux between them, shown in a triangular composition space. Each point represents a given compartment's composition in *cis* (100% at the top), *medial* (100% at the bottom right) and *trans* (100% at the bottom left) identities. Arrows show the vesicular flux. The base of each arrow is the composition of the donor compartment and the tip is the average composition of receiving compartments (ignoring back fusion). The Arrows' opacity is proportional to the flux of transported cargoes going through this path per unit time ($1/K_{\mathrm{f}}$), normalized by the total number of cargoes. The dashed line on the top-left triangle is a theoretical prediction discussed in the text. See also Fig.S5 for further characterizations of the vesicular transport, and Fig.S7 to discuss the impact of other budding and fusion implementations on the vesicular fluxes.

4 Discussion

The genesis and maintenance of complex membrane-bound organelles such as the Golgi apparatus rely on self-organization principles that are yet to be understood. Here we present a minimal model in which steady-state Golgi-like structures spontaneously emerge from the interplay between three basic mechanisms: the biochemical maturation of membrane components, the composition-dependent vesicular budding, and inter-compartment fusion. As our model does not include space, we do not refer to the spatial structure of a stacked Golgi, but rather to the self-organization of Golgi components into compartments of distinct identities undergoing vesicular exchange. Despite the absence of spatial information, we observe the emergence of directional vesicular exchanges between compartments of different identities. This suggests that spatial information is less crucial than biochemical ones, and is in line with observations that modifying the spatial structure of the Golgi preserves its functionality [10]. Our main result is that the steady-state organization of the organelle, in terms of size and composition of its compartments, is intimately linked to the directionality of vesicular exchange between compartments through the mechanism of homotypic fusion. Both aspects are controlled by a kinetic competition between vesicular exchange and biochemical maturation.

The results presented in Fig.2 can be used to explain a number of experimental observations. The predicted role of the budding and fusion rates is in agreement with the phenotype observed upon deletion of Arf1 (a protein involved in vesicle budding), which decreases the number of compartments and increases their size, particularly that of *trans*-compartments which seem to aggregate into one major cisterna [28]. A comparable phenotype has been observed upon mutation of NSF (a fusion protein), which produces extremely large, but transient, *trans*-compartments [32], thereby increasing the system's stochasticity (see Fig.S4 - SI, for a quantification of the fluctuations in our model). This is consistent with an increase of the fusion rate according to our model. Furthermore, the predicted correlation between

compartments' size and purity is in agreement with the observation that decreasing the budding rate by altering the activity of COPI (a budding protein) leads to larger and less sorted compartments [33]. Modifying the expression of *VPS74* (yeast homolog of *GOLPH3*) leads to an altered Golgi organization, comparable to the *ARF1* deletion phenotype [34]. Both Δ*arf1* and Δ*vps74* present an enlargement of the Golgi cisternae and a disruption of molecular gradients in the system, which we interpret, in the limits of our system, as a lower purity due to slower sorting kinetics following an impairment of the budding dynamics.

The directionality of vesicular transport is intimately linked to the steady-state organization of the organelle; vesicular transport is anterograde when compartments are pure, and it is retrograde when they are mixed. This can be intuitively understood with the notion of "contaminating species". If a compartment enriched in a particular molecular identity emits a vesicle of that identity, the vesicle will likely fuse back with the emitting compartment by homotypic fusion, yielding no vesicular exchange. One the other hand, an emitted vesicle that contains a minority (contaminating) identity will homotypically fuse with another compartment. If budding is faster than maturation, the contaminating species is the one that just underwent maturation, and its budding and fusion with more mature compartment leads to a anterograde transport. In such systems, compartments are rather small and pure. If maturation is faster than budding, the contaminating species is the less mature one, leading to a retrograde transport. Such systems are rather mixed with large compartments. Thus, the directionality of vesicular exchange is an emergent property intimately linked to the compartments' purity. The *medial*-rich compartments are special in that regard: for intermediate values of the purity ($K \sim k_{\mathrm{m}}$), they may be contaminated both by yet to be matured *cis*-species and already matured *trans*-species and can fuse both with *cis*-rich and *trans*-rich compartments. If inter-compartment fusion is allowed, large *medial*-rich compartments are relatively scarce, and emit few vesicles. On the other hand, *medial*-vesicles emitted by *cis/medial* and *medial/trans*-compartments may fuse together to form (small) *medial*-rich compartments. For intermediate values of the budding rates, this vesicular flux dominates vesicular exchange and leads to a centripetal vesicular flux towards *medial*-compartments (details in Fig.S5 - SI). If inter-compartment fusion is prohibited, which for instance corresponds to a situation where Golgi cisternae are immobilized [10], *medial*-rich compartments are present at steady-state, the centripetal vesicular flux is less intense, and the purity transition is accompanied with a direct transition from retrograde to anterograde vesicular flux (Fig.S7 - SI).

Regardless of the model's details, we find that systems showing a well defined polarity with well sorted cisternae exhibit anterograde vesicular fluxes, whereas systems with mixed compartments exhibit retrograde fluxes. The former dynamics is expected when biochemical maturation is the slowest kinetic process and compartments are long-lived, while the latter is expected when vesicular exchange is slow and the system is composed of transient compartments undergoing individual maturation. One can relate this prediction to the difference in organization and dynamics between the Golgi of S. cerevisiae and the more organized Golgi of higher organisms such as vertebrates. Maturation of Golgi cisternae has been directly observed in S. cerevisiae [8], with colocalization of different identity markers within single cisternae (low purity) during maturation, whereas vesicular transport phenotypes have been indirectly observed [10] or inferred through modeling [35] in mammalian cells. Consistent with our predictions, Golgi dynamics is one to two orders of magnitude faster in S. cerevisiae Golgi cisternae (maturation rate $\sim 1/\mathrm{min}$. [8, 9]) than in mammalian cells (export rate $\sim 1/30\mathrm{min}$. [36]). At this point, one may speculate a link between structure and function through kinetics. A well sorted and polarized Golgi is presumably required to accurately process complex cargo. The glycosylation of secreted cargo is key to the interaction of a vertebrate cell with its organism's immune system [37], and glycans appear to be more diverse in higher eukaryotes - which also possess a highly organized Golgi - than in unicellular eukaryotes like yeast [38]. The Golgi organization in S. cerevisiae could thus be the result of an adaptation that has favored fast transport over robustness of processing, leading to a less organized Golgi characterized by cisternal maturation and retrograde vesicular transport. Remarkably, the slowing down of Golgi transport when S. cerevisiae is starved in a glucose-free environment coincides with the Golgi becoming more polarized [39], strengthening the proposal derived from our model of a strong connection between transport kinetics and steady-state Golgi structure.

In summary, we have analyzed a model of self-organization and transport in cellular organelles based on a limited number of kinetic steps allowing for the generation and biochemical maturation of compartments, and vesicular exchange between them. Although we kept the complexity of individual steps to a minimum, our model gives rise to a rich diversity of phenotypes depending on the parameter values, and reproduces the effect of a number of specific protein mutations. We identify the concomitance of a structural transition (from large and mixed to small and pure sub-compartments) and a dynamical transition (from retrograde to anterograde vesicular exchange). Within our model, anterograde vesicular transport is accompanied by many spurious events of vesicle back-fusion. For very high budding rates, compartments are very pure and anterograde transport is dominated by vesicles undergoing maturation *after* budding from a compartment. In our model, such vesicle maturation events, which have been described in the secretory pathway as a way to direct vesicular traffic and to prevent vesicles back-fusion [40], only occur when compartments are very small (high budding rate). Adding composition-dependent feedback involving specific molecular actors to tune budding and fusion rates - for instance to reduce or prevent the budding of the majority species - will displace the

purity transition to lower budding rates and thus extend the regime of anterograde transport to larger sub-compartment steady-state sizes. Cisternal stabilizers like GRASP or vesicles careers like Golgins, known to already be present in the ancestor of eukaryotes [41], are obvious candidates. Although more complex models, and in particular the inclusion of spatial dependencies, are surely relevant to organelle's dynamics, the fundamental relationship between kinetics, structure and transport highlighted by our model is a universal feature of the interplay between biochemical maturation and vesicular exchange in cellular organelles.

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References

[1] Rebecca Heald and Orna Cohen-Fix. Morphology and function of membrane-bound organelles. *Current Opinion in Cell Biology*, 26:79–86, February 2014.

[2] J. Lippincott-Schwartz, T. H. Roberts, and K. Hirschberg. Secretory protein trafficking and organelle dynamics in living cells. *Annual Review of Cell and Developmental Biology*, 16:557–589, 2000.

[3] Pamela Stanley. Golgi glycosylation. *Cold Spring Harbor Perspectives in Biology*, 3(4), April 2011.

[4] Suda Yasuyuki and Nakano Akihiko. The Yeast Golgi Apparatus. *Traffic*, 13(4):505–510, December 2011.

[5] Gaelle Boncompain and Franck Perez. The many routes of Golgi-dependent trafficking. *Histochemistry and Cell Biology*, 140(3):251–260, September 2013.

[6] Vivek Malhotra and Satyajit Mayor. Cell biology: The Golgi grows up. *Nature*, 441(7096):939–940, June 2006.

[7] Benjamin S. Glick and Alberto Luini. Models for Golgi Traffic: A Critical Assessment. *Cold Spring Harbor Perspectives in Biology*, 3(11), November 2011.

[8] Eugene Losev, Catherine A. Reinke, Jennifer Jellen, Daniel E. Strongin, Brooke J. Bevis, and Benjamin S. Glick. Golgi maturation visualized in living yeast. *Nature*, 441(7096):1002–1006, June 2006.

[9] Kumi Matsuura-Tokita, Masaki Takeuchi, Akira Ichihara, Kenta Mikuriya, and Akihiko Nakano. Live imaging of yeast Golgi cisternal maturation. *Nature*, 441(7096):1007–1010, June 2006.

[10] Myun Hwa Dunlop, Andreas M. Ernst, Lena K. Schroeder, Derek K. Toomre, Grégory Lavieu, and James E. Rothman. Land-locked mammalian Golgi reveals cargo transport between stable cisternae. *Nature Communications*, 8(1):432, 2017.

[11] Mary J. Klute, Paul Melançon, and Joel B. Dacks. Evolution and diversity of the Golgi. *Cold Spring Harbor Perspectives in Biology*, 3(8):a007849, August 2011.

[12] Pierre Sens and Madan Rao. (Re)modeling the Golgi. *Methods in Cell Biology*, 118:299–310, 2013.

[13] Bernd Binder, Andrean Goede, Nikolaus Berndt, and Hermann-Georg Holzhütter. A conceptual mathematical model of the dynamic self-organisation of distinct cellular organelles. *PloS One*, 4(12):e8295, December 2009.

[14] Iaroslav Ispolatov and Anne Müsch. A model for the self-organization of vesicular flux and protein distributions in the Golgi apparatus. *PLoS computational biology*, 9(7):e1003125, 2013.

[15] Shankar Mukherji and Erin K. O'Shea. Mechanisms of organelle biogenesis govern stochastic fluctuations in organelle abundance. *eLife*, 3:e02678, June 2014.

[16] Himani Sachdeva, Mustansir Barma, and Madan Rao. Nonequilibrium description of de novo biogenesis and transport through Golgi-like cisternae. *Scientific Reports*, 6:38840, December 2016.

[17] Haijun Gong, Yusong Guo, Adam Linstedt, and Russell Schwartz. Discrete, continuous, and stochastic models of protein sorting in the Golgi apparatus. *Physical Review. E, Statistical, Nonlinear, and Soft Matter Physics*, 81(1 Pt 1):011914, January 2010.

[18] Quentin Vagne and Pierre Sens. Stochastic Model of Vesicular Sorting in Cellular Organelles. *Physical Review Letters*, 120(5):058102, February 2018.

[19] Masashi Tachikawa and Atsushi Mochizuki. Golgi apparatus self-organizes into the characteristic shape via postmitotic reassembly dynamics. *Proceedings of the National Academy of Sciences of the United States of America*, 114(20):5177–5182, May 2017.

[20] Jen-Hsuan Wei and Joachim Seemann. Golgi ribbon disassembly during mitosis, differentiation and disease progression. *Current Opinion in Cell Biology*, 47:43–51, August 2017.

[21] Jens Kühnle, Julian Shillcock, Ole G. Mouritsen, and Matthias Weiss. A modeling approach to the self-assembly of the Golgi apparatus. *Biophysical Journal*, 98(12):2839–2847, June 2010.

[22] Bianka L. Grosshans, Darinel Ortiz, and Peter Novick. Rabs and their effectors: Achieving specificity in membrane traffic. *Proceedings of the National Academy of Sciences*, 103(32):11821–11827, August 2006.

[23] Huaqing Cai, Karin Reinisch, and Susan Ferro-Novick. Coats, tethers, Rabs, and SNAREs work together to mediate the intracellular destination of a transport vesicle. *Developmental Cell*, 12(5):671–682, May 2007.

[24] Juan S. Bonifacino and Benjamin S. Glick. The mechanisms of vesicle budding and fusion. *Cell*, 116(2):153–166, January 2004.

[25] S. Dmitrieff and P. Sens. Cooperative protein transport in cellular organelles. *Physical Review. E, Statistical, Nonlinear, and Soft Matter Physics*, 83(4 Pt 1):041923, April 2011.

[26] W. Antonin, C. Holroyd, R. Tikkanen, S. Höning, and R. Jahn. The R-SNARE endobrevin/VAMP-8 mediates homotypic fusion of early endosomes and late endosomes. *Molecular Biology of the Cell*, 11(10):3289–3298, October 2000.

[27] Suzanne R. Pfeffer. How the Golgi works: a cisternal progenitor model. *Proceedings of the National Academy of Sciences of the United States of America*, 107(46):19614–19618, November 2010.

[28] Madhura Bhave, Effrosyni Papanikou, Prasanna Iyer, Koushal Pandya, Bhawik Kumar Jain, Abira Ganguly, Chandrakala Sharma, Ketakee Pawar, Jotham Austin, Kasey J. Day, Olivia W. Rossanese, Benjamin S. Glick, and Dibyendu Bhattacharyya. Golgi enlargement in Arf-depleted yeast cells is due to altered dynamics of cisternal maturation. *Journal of Cell Science*, 127(Pt 1):250–257, January 2014.

[29] Daniel T. Gillespie. Exact stochastic simulation of coupled chemical reactions. *The Journal of Physical Chemistry*, 81(25):2340–2361, December 1977.

[30] Quentin Vagne, Matthew S. Turner, and Pierre Sens. Sensing Size through Clustering in Non-Equilibrium Membranes and the Control of Membrane-Bound Enzymatic Reactions. *PloS One*, 10(12):e0143470, 2015.

[31] Matthew S. Turner, Pierre Sens, and Nicholas D. Socci. Nonequilibrium Raftlike Membrane Domains under Continuous Recycling. *Physical Review Letters*, 95(16):168301, October 2005.

[32] Sayuri Tanabashi, Keiko Shoda, Chieko Saito, Tomoaki Sakamoto, Tetsuya Kurata, Tomohiro Uemura, and Akihiko Nakano. A missense mutation in the NSF gene causes abnormal Golgi morphology in Arabidopsis thaliana. *Cell Structure and Function*, February 2018.

[33] Effrosyni Papanikou, Kasey J. Day, Jotham Austin, and Benjamin S. Glick. COPI selectively drives maturation of the early Golgi. *eLife*, 4, December 2015.

[34] Prasanna Iyer, Madhura Bhave, Bhawik Kumar Jain, Sudeshna RoyChowdhury, and Dibyendu Bhattacharyya. Vps74p controls Golgi size in an Arf1-dependent manner. *FEBS letters*, October 2018.

[35] Serge Dmitrieff, Madan Rao, and Pierre Sens. Quantitative analysis of intra-Golgi transport shows intercisternal exchange for all cargo. *Proceedings of the National Academy of Sciences of the United States of America*, 110(39):15692–15697, September 2013.

[36] L. Bonfanti, A. A. Mironov, J. A. Martínez-Menárguez, O. Martella, A. Fusella, M. Baldassarre, R. Buccione, H. J. Geuze, A. A. Mironov, and A. Luini. Procollagen traverses the Golgi stack without leaving the lumen of cisternae: evidence for cisternal maturation. *Cell*, 95(7):993–1003, December 1998.

[37] Sean O. Ryan and Brian A. Cobb. Roles for major histocompatibility complex glycosylation in immune function. *Seminars in Immunopathology*, 34(3):425–441, May 2012.

[38] Peng Wang, Hong Wang, Jiangtao Gai, Xiaoli Tian, Xiaoxiao Zhang, Yongzhi Lv, and Yi Jian. Evolution of protein *N*-glycosylation process in Golgi apparatus which shapes diversity of protein *N*-glycan structures in plants, animals and fungi. *Scientific Reports*, 7:40301, January 2017.

[39] Stephanie K. Levi, Dibyendu Bhattacharyya, Rita L. Strack, Jotham R. Austin, and Benjamin S. Glick. The Yeast GRASP Grh1 Colocalizes with COPII and Is Dispensable for Organizing the Secretory Pathway. *Traffic (Copenhagen, Denmark)*, 11(9):1168–1179, September 2010.

[40] Christopher Lord, Deepali Bhandari, Shekar Menon, Majid Ghassemian, Deborah Nycz, Jesse Hay, Pradipta Ghosh, and Susan Ferro-Novick. Sequential interactions with Sec23 control the direction of vesicle traffic. *Nature*, 473(7346):181–186, May 2011.

[41] Lael D. Barlow, Eva Nývltová, Maria Aguilar, Jan Tachezy, and Joel B. Dacks. A sophisticated, differentiated Golgi in the ancestor of eukaryotes. *BMC Biology*, 16:27, March 2018.