experiments by observing microtubule assembly on the micropedestals, fabricated on the surface of the coverslip. This novel assay reveals microtubule dynamics away from contact with any surfaces, making it possible to avoid non-specific effects that appear when microtubules come into contact with the coverslip. We have observed a significant decrease in the frequency of incorporation of GTP-tubulins into the sites of microtubule lattice defects and a decrease in the frequency of rescues compared to the results of the conventional assay. These results are in agreement with our Monte Carlo model's predictions. Overall, our study provides a unified framework for describing the processes of microtubule assembly and their nonequilibrium dynamics.

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S1.72. On the codon usage bias

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In modern genomics, the question of the codon usage bias in the functioning of a living organism remains very important and intriguing.

Traditionally, here the general position is used about the determining role of the GC composition in the formation of the main structural properties of genomic DNA. According to this concept, assessments of the contribution of guanine and cytosine to the third position of the codon (GC3) in the composition of protein-coding genes are often used as a possible measure of codon bias. However, in real circumstances, based, for example, on the specific composition of the human genome, such a "GC3 preference" turns out to be no more than 25%! Therefore, at the moment, many are still inclined to believe that under the existing conditions of the degeneracy of the genetic code, the observed inequality of synonymous codons in both pro and eukaryotes is a consequence of a non-trivial balance of two factors - the influence of natural selection and mutational predisposition.

Earlier in our work, we have already pointed out the important contribution of the "hidden" ambiguity of the initial form of complementary H-pairing of nitrous bases in initiating the observed features of the structural and functional organization of nucleic acid molecules. This ambiguity is caused by the bi-stable nature of the pyramidal structure of the exocyclic amino groups of adenine, guanine, and cytosine involved in the hydrogen bonding. The reduced, 2-fold polymorphism of the structure of AT-pairs compared to the 4-fold polymorphism of GC-pairs determines the observed dominance of A/T tracks over G/C tracks in the DNA double helix of any organisms. Thus, the natural selection of pairs is realized in ensuring the reliability of the processes of preservation and transmission of genetic information in the cell.

There is reason to expect that the increased ambiguity of GC base pairing form of compared to AT-pairing should somehow be reflected in the case under discussion, when forming the preferred composition of nucleotide triplets in the "codon-anticodon" structure of the nucleic acid binding center to maintain high accuracy and stability of the course of genetic processes.

Using the GenBank data and the www.kazusa.or.jp/codon resource, we performed a spectral analysis of the frequencies of occurrence of all 64 types of codons in the genes of a wide representation of pro- and eukaryotes, covering a scale range of sizes of the studied genomes (from 1.6 Mb to 140 000 Mb) with different GC-content. The genomes of amoeba (Amoeba proteus), tardigrade (Targidrada), horseshoe crab (Limulus polyphemus), and mollusk (Nautilus pompilius) were studied as an example of relic eukaryotes. Other eukaryotes as well as prokaryotes were represented by the genomes of a human (Homo sapiens), a chimpanzee (Pan triglodites), a mouse (Mus musculis), a marble lungfish (Protopterus aethiopicus), a frog (Xenopus tropicalis), a fly (Drosophila melanogaster), a flower (Arabidopsis thaliana), slime mold (Dictyostelium discoideum), parasite (Leishmania major), yeast (Saccharomyces cerevisiae), malaria parasite (Plasmodium farciparum), bacteria (Escherichia coli), and very small bacteria (Candidatus Pelagibacter). The spectral analysis itself was carried out on the basis of a developed own computer program with a description of the algorithm in [A.A.Samchenko et al, Biophysics,2016, 61 (6),813-824].

The results obtained generally confirmed the assumption made. It has been shown that each organism has two sets of codons bias. The first group is the most numerous. It concentrates approximately 64 to 95 percent of these codons with either an A or T(U) base in the second position. Thus, the priority of the contribution of the "most reliable" bases with initially low structural polymorphism of complementary H-pairing is implemented. This achieves a consistently clear spatial fixation of the central link of the "lock-key" recognition system (codon-anticodon) in the functional complexes of DNA-mRNA, mRNA-tRNA, tRNA-rRNA.

In the second, very small group, the remaining codon usage bias with a central G or C nitrous base gathered.

In general, the resulting split of codon bias into two such groups is quite significant, can reach a ratio of 95:5 (%), and depends on the composition of a particular genome.

S1.73. Pacemaker currents are involved in the regulation of 3-week-old rat's atrial myocardial contractility

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Hyperpolarization-activated cyclic nucleotide-regulated channels (HCN4) are key membrane proteins involved in the initiation and regulation of heartbeat. The pacemaker cells in the sinoatrial node generate the electrical impulse that underlies the contraction of atrial and ventricular cardiomyocytes. HCN4 channels contribute to the control of resting membrane potential and rhythmic activity of excitable cells expressing these channels. In addition to abundant expression in the pacing and conduction system, the membrane of adult rat working myocardium cells is characterized by low levels of HCN channels. However, in the early embryonic stages, HCN4 channels is abundantly transcribed throughout the heart and makes an important role in ventricular myocyte automatism triggered by If. By birth, HCN4 channels transcription is suppressed in working-type cardiomyocytes and remains at low levels in the adult body, which prevents pathological remodeling of the heart.

The aim is to study the effect of pacing currents blockade, in the regulation of atrial myocardial contractility in 3-week-old rats.

An object of the study were chosen 3 weeks old rats, which are at the initial stage of heart sympathetic innervation.

Myocardial contractile activity was studied in an in vivo experiment on right atrial myocardial strips on a Power Lab setup (AD Instruments, Australia).

Registration of spontaneously generated action potentials was performed on a microelectrode unit. The blocker of currents activated by

hyperpolarization, ZD7288, at a concentration of 10-6 M, was used as pharmacological agents.
The initial contraction force of the isolated right atrial myocardium was 0.26±0.13 g. After adding the blocker ZD7288 (10-6 M) to the working solution, there was a gradual decrease of contraction force during the 21st minute of the experiment. During the 1st minute of If blockade, the contraction force of isolated atrial myocardial strips decreased to 0.25±0.13 g ($p<0.01$). By the 14th minute of the experiment, contraction force decreased to 0.23±0.13 g ($p<0.05$). By the final minute of the experiment, contraction force decreased to 0.22±0.12 g ($p<0.01$). The decrease of contraction force of isolated atrial myocardial strips was 15% of the initial value.
When ZD7288 at a concentration of 10-6 M was injected into the perfused solution, the action potential duration at the 50% level increased from 11.86±1.21 ms to 19.14±1.77 ms ($p\leq0.01$). At the 7th minute of the study, the greatest increase in action potential duration at the 90% level was recorded from 22.43±3.6 ms to 30.71±2.69 ms ($p\leq0.01$), the parameter of total action potential cycle length from 160.28±7.85 to 171±9.14 ms ($p\leq0.01$). The baseline value of the action potential generation frequency parameter was 375.04±16.84 units/min. At the 7th and 15th minutes of the experiment, the values of this parameter decreased to 351.71±18.12 units/min ($p\leq0.01$) and 354.58±17.23 units/min ($p\leq0.01$), respectively.
Thus, the results showed that If blockade has a significant effect on the working atrial myocardium of 3-week-old rats.
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S1.74. Phase separation of SARS-CoV-2 N-protein and viral RNA: possible mechanism and regulation by nucleoside derivatives

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The nucleocapsid protein (N) of severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2) tends to form biomacromolecular condensates with RNA in aqueous media via the liquid-liquid phase separation (LLPS) mechanism. N-RNA LLPS in the cytoplasm of the host cell supposedly enables viral immune evasion by blocking an intracellular signaling pathway and may contribute to viral replication. Within the framework of the "scaffold-client" model, which is typically used to describe multicomponent condensates, N protein can be regarded as the main driver ("scaffold") of LLPS, and its "clients" include the components of the replication/transcription machinery. Their concentration and redistribution on SARS-CoV-2 genomic RNA are needed for the onset of replication-transcription and the switch from classical to discontinuous transcription, respectively, resulting in the production of genomic and subgenomic RNA.
Previously, small-molecule LLPS modulators that reduce condensate density, have been shown to enhance the activity of viral polymerase inhibitors by facilitating their access to the target. Such modulators and inhibitors of N-RNA LLPS may find application in the development of combination strategies for antiviral therapy, and further investigation of N-RNA phase transitions may contribute to the rational design of new antivirals. Condensate-stabilizing compounds, in turn, are theoretically capable of disrupting the viral life cycle by the unpacking of the nucleocapsid right after infection. Thus, they may also prove to be therapeutically relevant. Despite the growing interest to this matter, studies of N-RNA LLPS modulators are at an early stage. One limitation is the lack of a simple and adequate in vitro model of the condensates.
The aim of this work was to obtain a model of N-RNA condensates under physiological conditions and to evaluate the effects of known nucleoside/nucleotide-based antivirals on these condensates. The key difference between the proposed model and those described previously is the choice of RNA and N-RNA ratio. We used fragments of the SARS-CoV-2 genome that contain primary and secondary structure elements recognized by N protein, while in previous studies random-sequence oligoribonucleotides were typically used. We also used an N:RNA ratio comparable to that expected in SARS-CoV-2 virions. Nucleoside/nucleotide analogs were considered as possible modulators of N-RNA LLPS based on the reports of condensate sensitivity to ATP, which is arguable the key endogenous modulator of biopolymer phase transitions.
The condensates were obtained using a fluorescently labeled recombinant N protein and an RNA sequence from the 5'-untranslated region of the viral genome, which forms a branched hairpin and contains sequences recognized by N protein. Phase separation was confirmed using fluorescence microscopy and turbidimetry. The model of the condensates was validated by comparing observed and predicted effects of pH, temperature, solution ionic strength, and known modulators. To assess the effects of nucleoside/nucleotide modulators, changes in N partitioning coefficient and the total area of the condensates per surface unit were calculated based on fluorescence microscopy analyses of N-RNA mixtures in the presence/absence of the modulators. Finally, the effects of nucleoside/nucleotide analogs on the condensates were correlated with antiviral activity in cells.
Analysis of condensate sensitivity to external factors supported the recently proposed hypothesis explaining the relationship between specific electrostatic N-RNA interactions and nonspecific hydrophobic interactions within unstructured N regions. According to that hypothesis, the mechanism of the RNA-dependent phase transition includes the following steps: 1) recognition of sequence motifs characteristic of the 5'-untranslated region of the viral genome by the RNA-binding domain of N increases promotes contacts between the backbone of the adjacent RNA region and the positively charged N fragment near the dimerization domain; 2) RNA binding alters the conformation of the dimerization domain, causing its partial denaturation; 3) partial protein denaturation and exposure of hydrophobic regions initiate the phase transition.
Analysis of the effects of nucleoside/nucleotide analogs revealed weak correlation between LLPS modulation and antiviral activity in the series of 5'-norcarbocyclic nucleoside derivatives and fleximer-containing nucleoside analogs [1]. Non-nucleoside modulators, such as perylene-based antivirals, showed no such correlation. In the nucleoside analog series, top effects were obtained for 5'-norcarbocyclic derivatives, which caused up to 15-fold increase in the efficiency of N separation. These compounds might hold promise for the development of therapeutic agents. However, to take into account possible side effects, their effects on host cell condensates (membraneless organelles) should be tested.
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S1.75. Photodimerization of thymine chromophores in poly-T aqueous solutions at room temperature

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Our interest in the study of photophysical and photochemical processes in aqueous solutions of poly-T was associated both with the fact that similar processes occur in living organisms and with an attempt to approach the implementation of photochemical recording of information on thymine or its derivatives [1, 2].