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Spectral comparative analysis of acid and salt forms of nucleotides and their components

Chaired by **Dr. Alfredo Berzal-Herranz**
and **Prof. Dr. Maria Emilia Sousa**



pharmaceuticals



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Abstract: It is well known that studies of the luminescence of bases, nucleosides, nucleotides, and especially their acid forms at room temperature are not sufficiently detailed. Therefore, the aim of this study was to analyse and compare their spectral properties. We studied the absorption and emission of monoribonucleotides and their components dissolved in water at room temperature. For these samples, we measured absorption spectra using a Specord 210plus instrument and fluorescence and phosphorescence excitation and emission spectra using Horiba Fluoro Max+ instruments.

We observed a change in the ratio between the peaks of the absorption spectra of acidic and salt forms of nucleotides. The presence of luminescence was observed in nucleotides in both their acid and salt forms, as well as in their nucleosides and bases. In addition, slight changes in the luminescence spectra were observed for acid and salt forms of nucleotides. The luminescence intensity decreases from nucleotides to bases. Therefore, our observations confirm that nucleotides, nucleosides, and nucleic acid bases exhibit luminescence at room temperature, which may be useful information for further research in this direction. In addition, changes in the luminescence spectra between acidic and salt forms of nucleotides may be important diagnostic features in biochemical and medical studies where the identification and separation of different forms of nucleotides is important.

Keywords: Nucleic acid bases, luminescence, nucleosides, nucleotides.



Introduction

The properties of nucleotides, or nucleic acid monomers, play an important role in intracellular processes. This is why the study of bases, nucleosides and nucleotides is so important for understanding the processes involved in maintaining cell vitality. One of the most widely used areas of experimental research is spectroscopy, which can provide a large amount of data about the samples being measured. We have used spectroscopy to study bases, nucleosides and nucleotides in acid and salt forms. The absorption, excitation, emission and phosphorescence spectra of the samples have been measured.



Results and discussion

Abbreviations

A - adenine

rA - adenosine

AMP - adenosine 5`-monophosphate

AMPNa - adenosine 5`-
monophosphate sodium

C - cytosine

rC - cytidine

CMP - cytidine 5`-monophosphate

CMPNa - cytidine 5`-monophosphate
sodium

G - guanine

rG - guanosine

GMP - guanosine 5`-monophosphate

GMPNa - guanosine 5`-
monophosphate sodium

U - uracil

rU - uridine

UMP - uridine 5`- monophosphate

UMPNa - uridine 5`-monophosphate
sodium

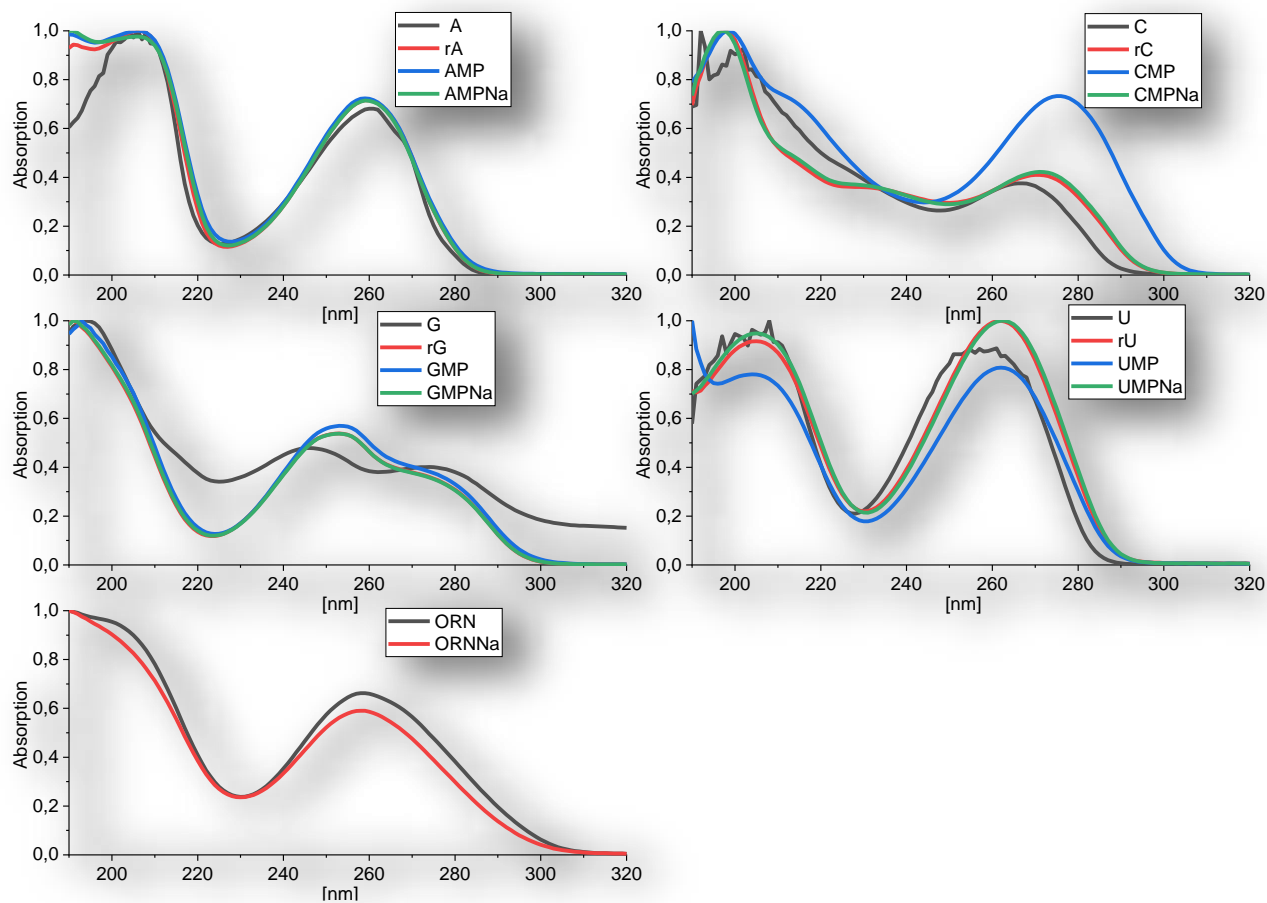
ORN – yeast RNA

ORNa - yeast RNA sodium



Results and discussion

Normalized absorption spectra





Results and discussion

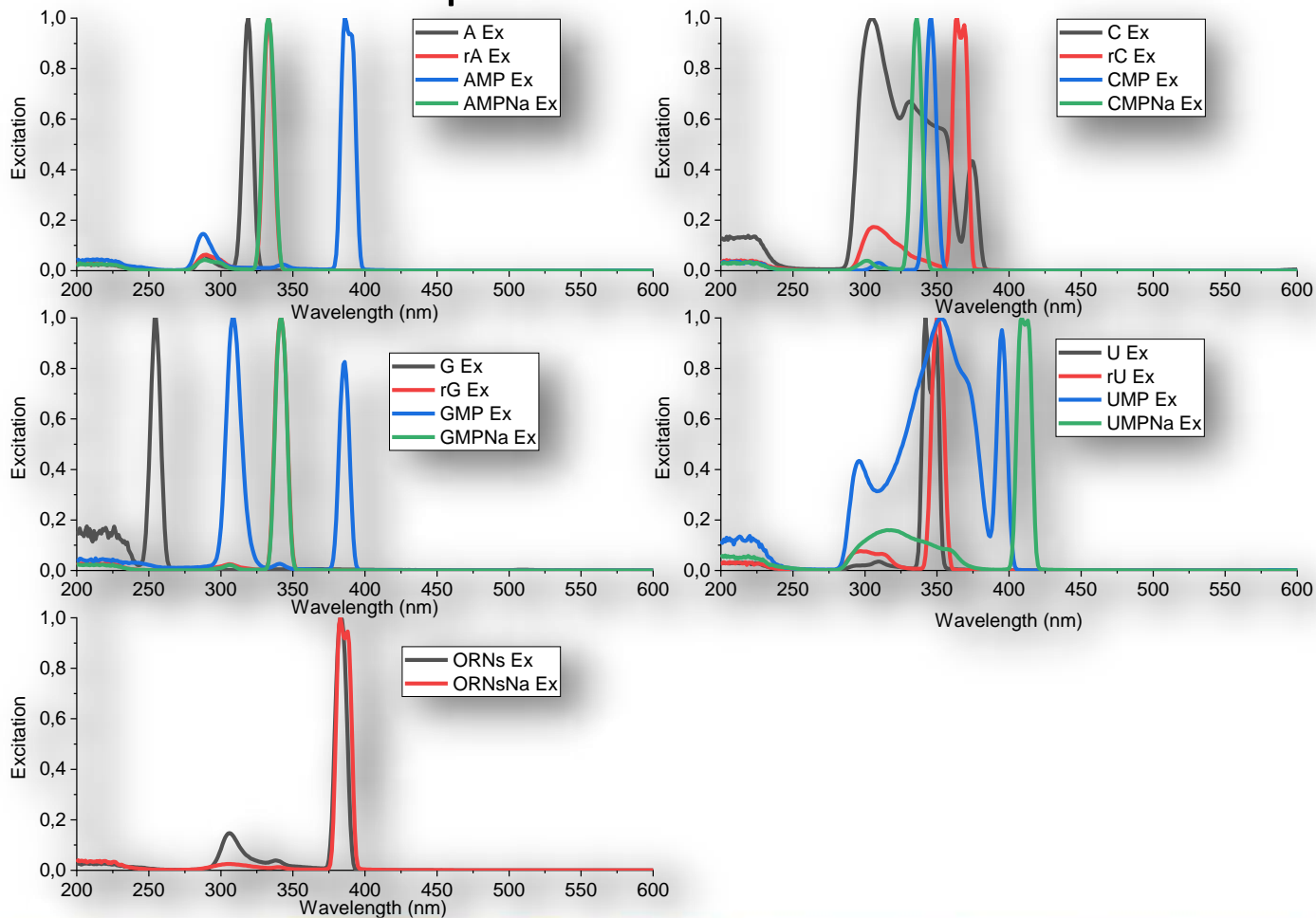
The absorption spectra of adenine and its compounds show two peaks common to all compounds. These peaks are at 206 nm and 259 nm. The absorption spectra of uracil and its compounds show two peaks common to all compounds except uracil, whose second peak is slightly different. These peaks are located at 205 nm and 262 nm (for uracil - 205 nm and 257 nm). In the absorption spectra of cytosine and its compounds, two peaks can be observed for all compounds except cytosine. The peaks of cytosine are at 192 nm, 201 nm and 267 nm. The peaks of cytidine are at 198 nm and 271 nm.

The peaks of cytidine 5`-monophosphate are at 199 nm and 276 nm. The peaks of cytidine 5`-monophosphate sodium are at 197 nm and 271 nm. In the absorption spectra of guanine and its compounds, guanine peaks are observed at 194 nm, 246 nm and 274 nm. The peaks of guanosine and guanosine 5`-monophosphate are at 191 nm and 253 nm. In the absorption spectra of RNA and RNA sodium we observe a peak at 258 nm for both compounds.



Results and discussion

Normalized excitation spectra





Results and discussion

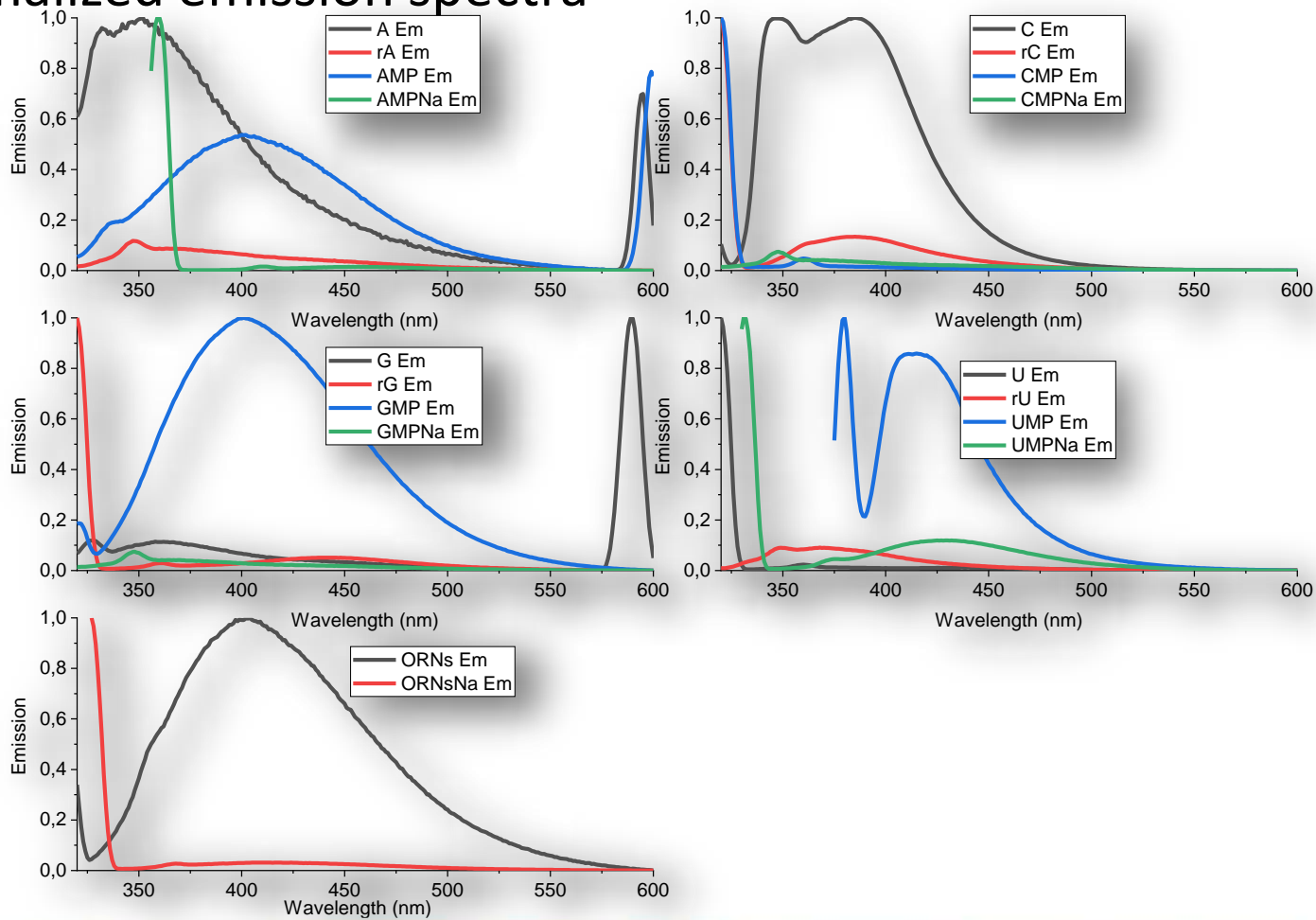
In the excitation spectrum of adenine we observe two peaks at 289 nm and 319 nm. In the excitation spectrum of adenosine we observe peaks at 290 nm and 333 nm. In the excitation spectrum of adenosine 5'-monophosphate we observe distinct peaks at 288 nm, 386 nm and 400 nm. In the excitation spectrum of adenosine 5'-monophosphate sodium we observe peaks at 289 nm and 333 nm. The excitation spectrum of uracil shows peaks at 309 nm, 342 nm and 349 nm. The excitation spectrum of uridine shows peaks at 297 nm and 350 nm. The excitation spectrum of uridine 5'-monophosphate shows peaks at 296 nm, 352 nm and 395 nm.

The excitation spectrum of uridine 5'-monophosphate sodium shows peaks at 316 nm, 409 nm and 413 nm. The excitation spectrum of cytosine shows peaks at 305 nm, 331 nm and 374 nm. The excitation spectrum of cytidine shows peaks at 306 nm, 364 nm and 369 nm. The excitation spectrum of cytidine 5'-monophosphate shows peaks at 310 nm and 346 nm. The excitation spectrum of cytidine 5'-monophosphate sodium shows peaks at 302 nm and 336 nm. The excitation spectrum of guanine shows a peak at 255 nm. The excitation spectrum of guanosine shows peaks at 306 nm and 342 nm. In the excitation spectrum of guanosine 5'-monophosphate we observe peaks at 309 nm, 341 nm and 386 nm. The excitation spectrum of guanosine 5'-monophosphate sodium shows peaks at 306 nm and 342 nm. The excitation spectrum of RNA shows peaks at 306 nm, 338 nm and 383 nm. The excitation spectrum of RNA sodium shows peaks at 304 nm and 340 nm, 383 nm and 388 nm.



Results and discussion

Normalized emission spectra





Results and discussion

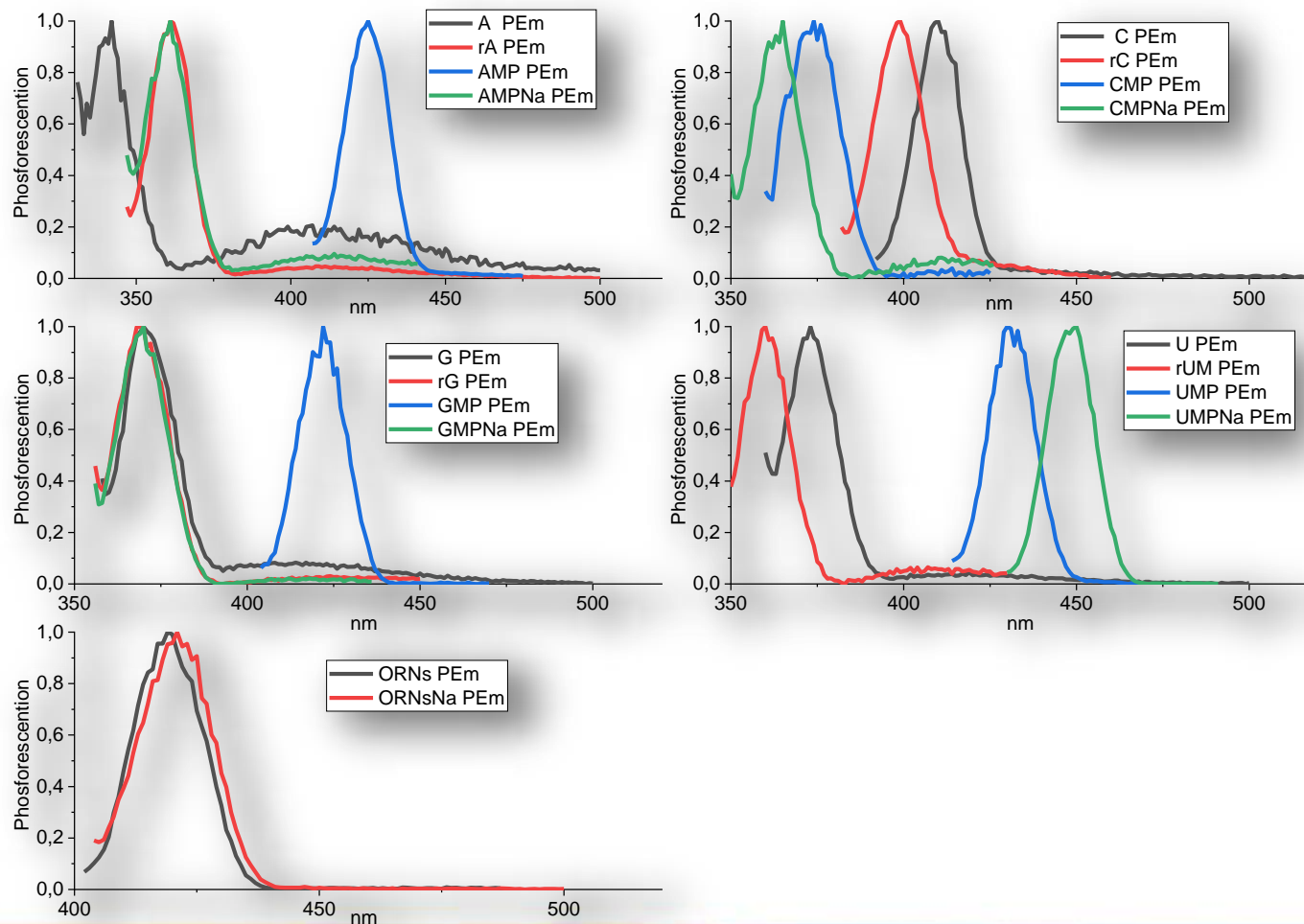
In the emission spectrum of adenine we observe peaks at 351 nm and 595 nm. In the emission spectrum of adenosine we observe a peak at 332 nm. In the emission spectrum of adenosine 5'-monophosphate we observe a peak at 396 nm. The emission spectrum of adenosine 5'-monophosphate sodium shows a peak at 349 nm. The emission spectrum of uracil shows a peak at 360 nm. The emission spectrum of uridine shows peaks at 359 nm and 378 nm. The emission spectrum of uridine 5'-monophosphate shows a peak at 360 nm. The emission spectrum of uridine 5'-monophosphate sodium shows peaks at 366 nm and 417 nm. The emission spectrum of cytosine shows peaks at 348 nm and 385 nm. The emission spectrum of cytidine shows a peak at 380 nm.

The emission spectrum of cytidine 5'-monophosphate shows a peak at 355 nm. The emission spectrum of cytidine 5'-monophosphate sodium shows a peak at 353 nm. The emission spectrum of guanine shows peaks at 327 nm and 360 nm. The emission spectrum of guanosine shows peaks at 327 nm and 360 nm. The emission spectrum of guanosine 5'-monophosphate shows a peak at 376 nm. The emission spectrum of guanosine 5'-monophosphate sodium shows a peak at 334 nm. The emission spectrum of RNA shows a peak at 403 nm. The spectrum of RNA sodium shows peaks at 356 nm and 396 nm.



Results and discussion

Normalized phosphorescence spectra





Results and discussion

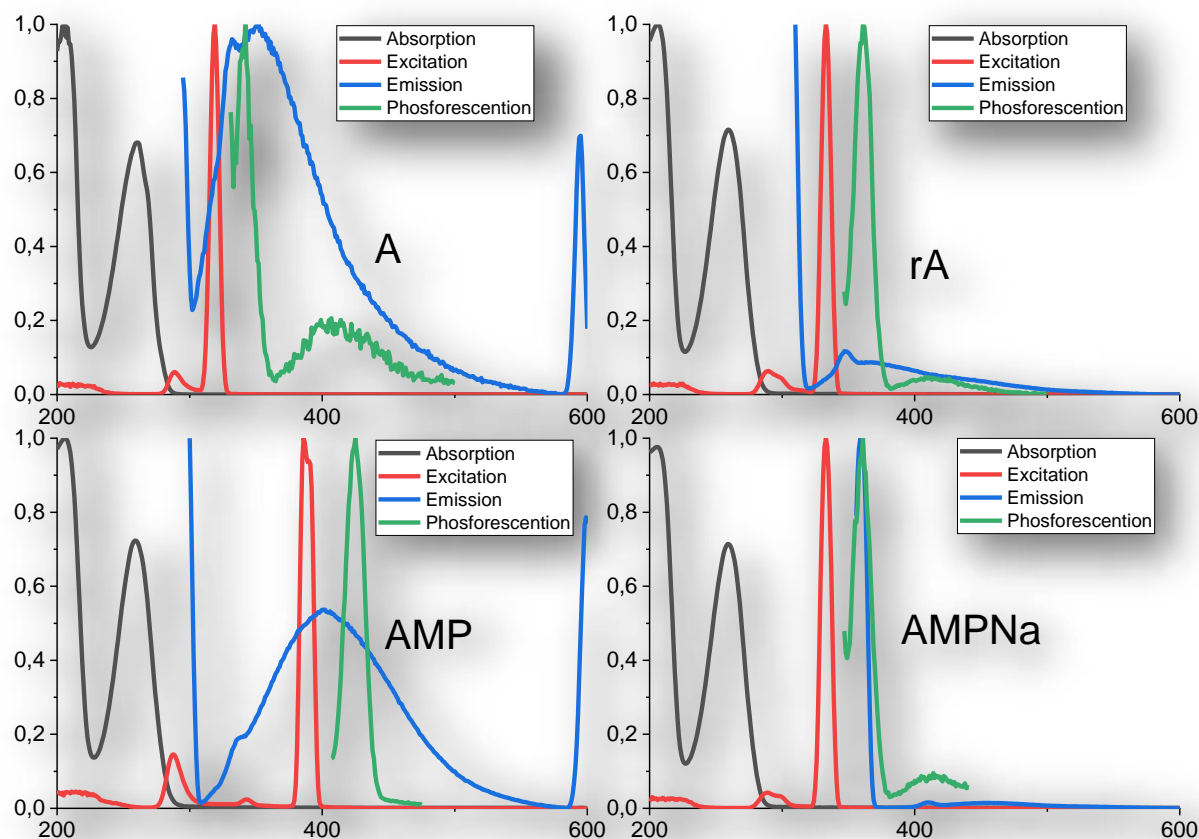
In the phosphorescence spectrum of adenine we observe a peak at 342 nm. In the phosphorescence spectra of adenosine and adenosine 5'-monophosphate we observe a peak at 345 nm. In the phosphorescence spectrum of adenosine 5'-monophosphate sodium we observe a peak at 349 nm. The phosphorescence spectra of uracil and uridine show a peak at 373 nm. The phosphorescence spectrum of uridine 5'-monophosphate shows a peak at 377 nm. The phosphorescence spectrum of uridine 5'-monophosphate sodium shows a peak at 379 nm. The phosphorescence spectrum of cytosine shows a peak at 410 nm. The phosphorescence spectrum of cytidine shows a peak at 409 nm.

In the phosphorescence spectrum of cytidine 5'-monophosphate we observe a peak at 406 nm. In the phosphorescence spectrum of cytidine 5'-monophosphate sodium we observe a peak at 407 nm. The phosphorescence spectrum of guanine shows a peak at 370 nm. The phosphorescence spectrum of guanosine shows a peak at 371 nm. The phosphorescence spectrum of guanosine 5'-monophosphate shows a peak at 376 nm. The phosphorescence spectrum of guanosine 5'-monophosphate sodium shows a peak at 372 nm. The phosphorescence spectra of RNA and RNA sodium show a peak at 419 nm.



Results and discussion

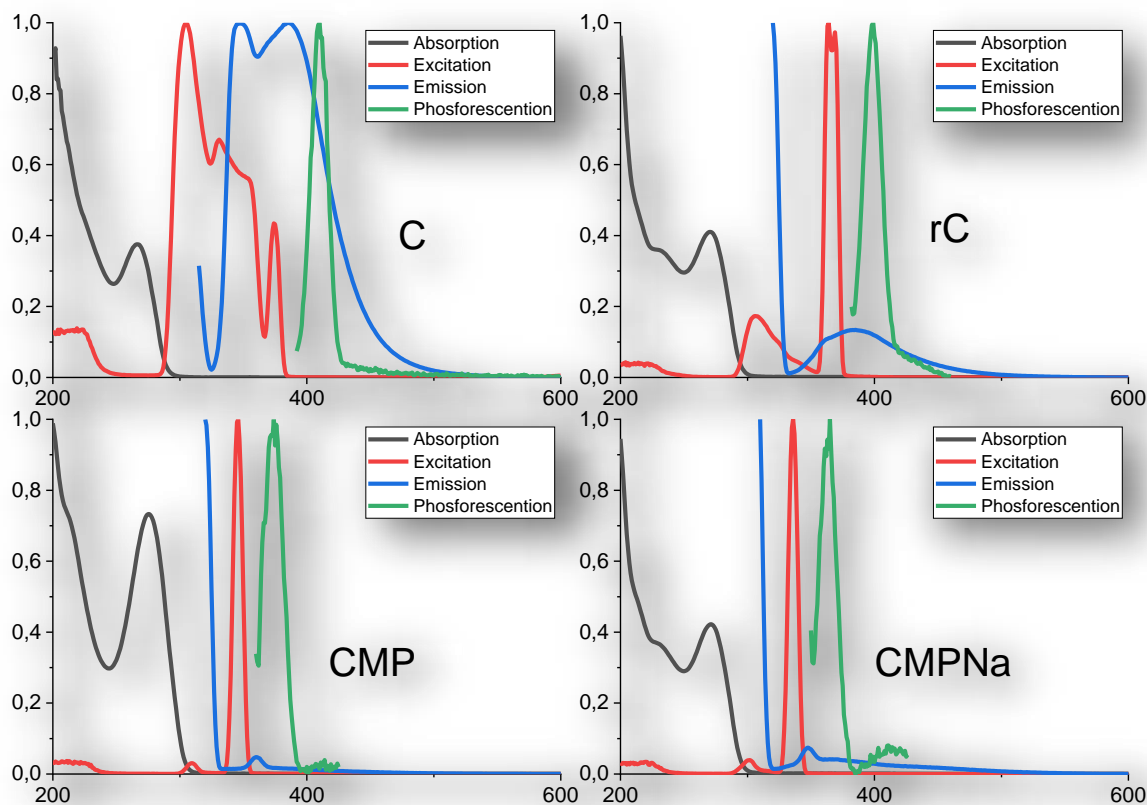
Normalized absorption, excitation, emission and phosphorescence spectrum of adenine, adenosine, adenosine 5'-monophosphate and adenosine 5'-monophosphate sodium





Results and discussion

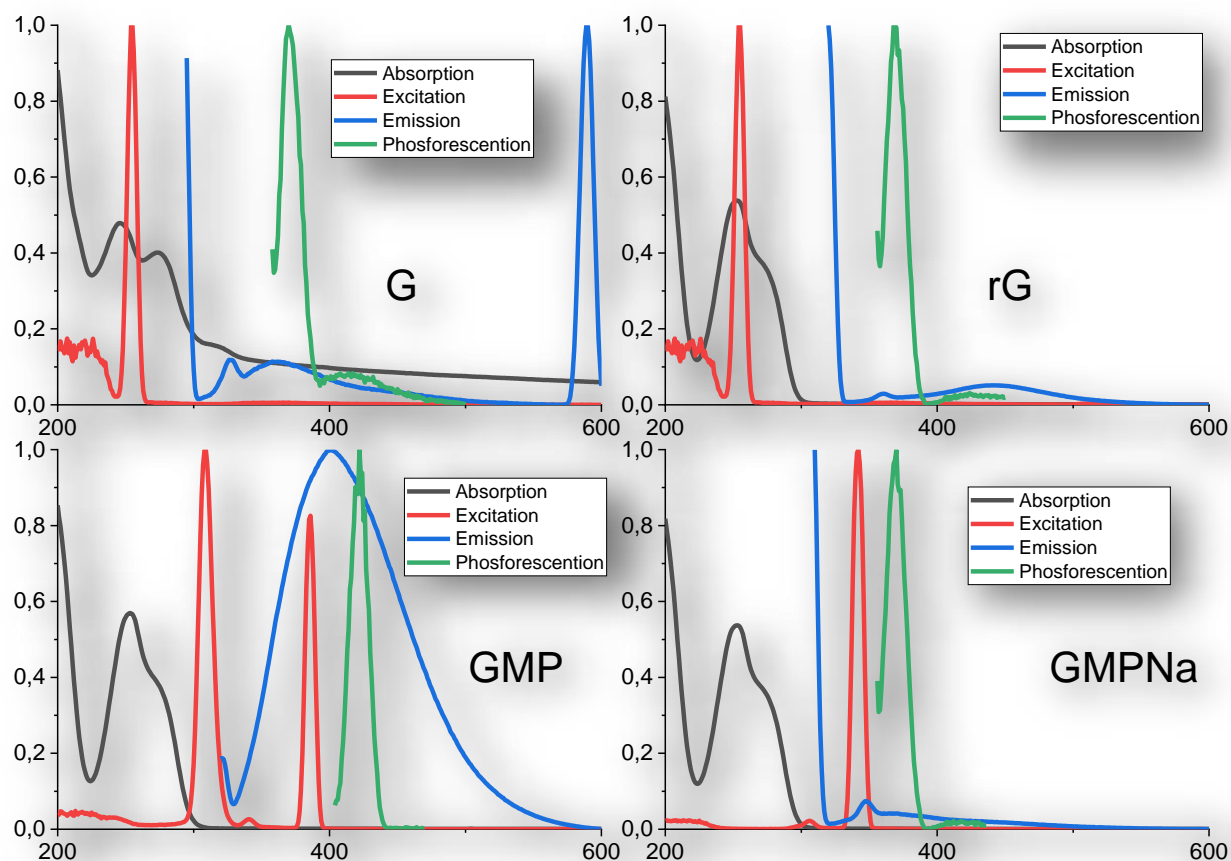
Normalized absorption, excitation, emission and phosphorescence spectrum of cytosine, cytidine, cytidine 5`-monophosphate and cytidine 5`-monophosphate sodium





Results and discussion

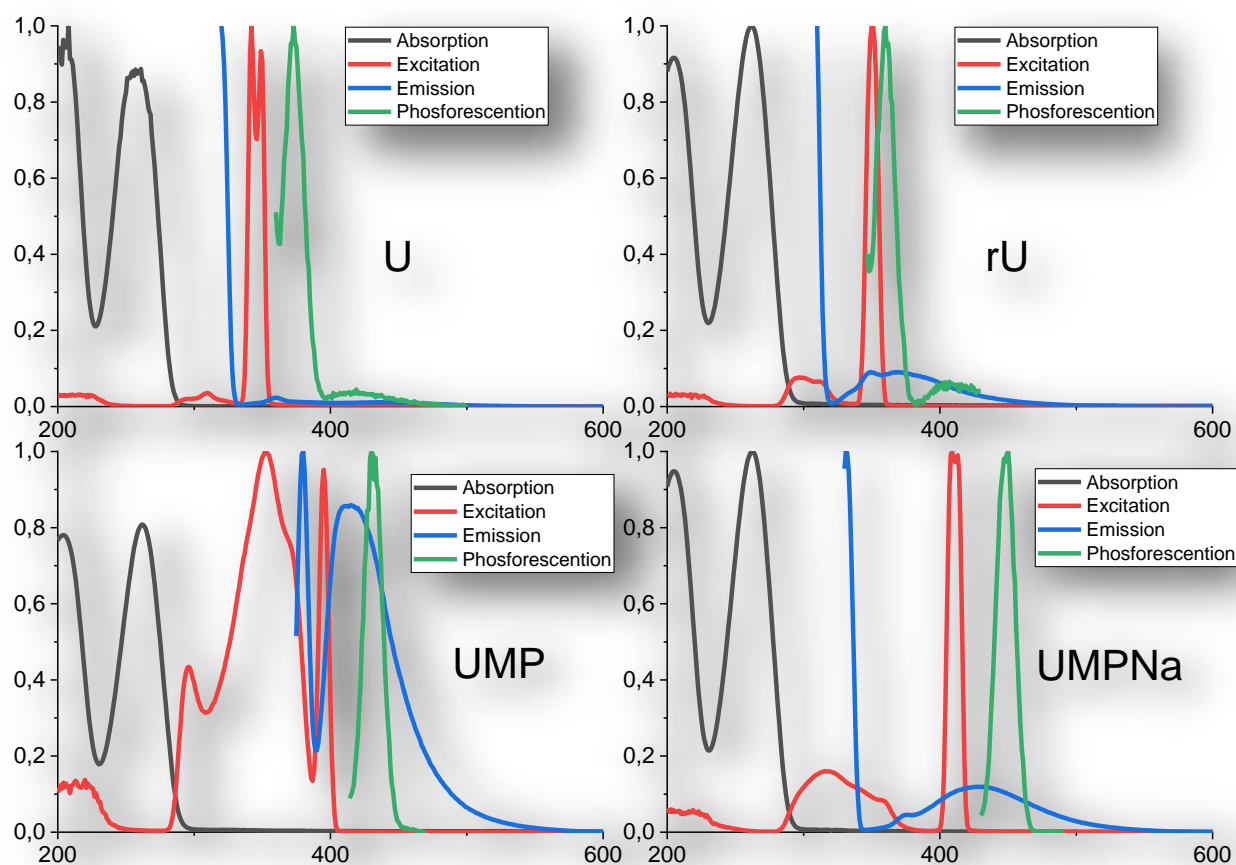
Normalized absorption, excitation, emission and phosphorescence spectrum of guanine, guanosine, guanosine 5`-monophosphate and guanosine 5`-monophosphate sodium





Results and discussion

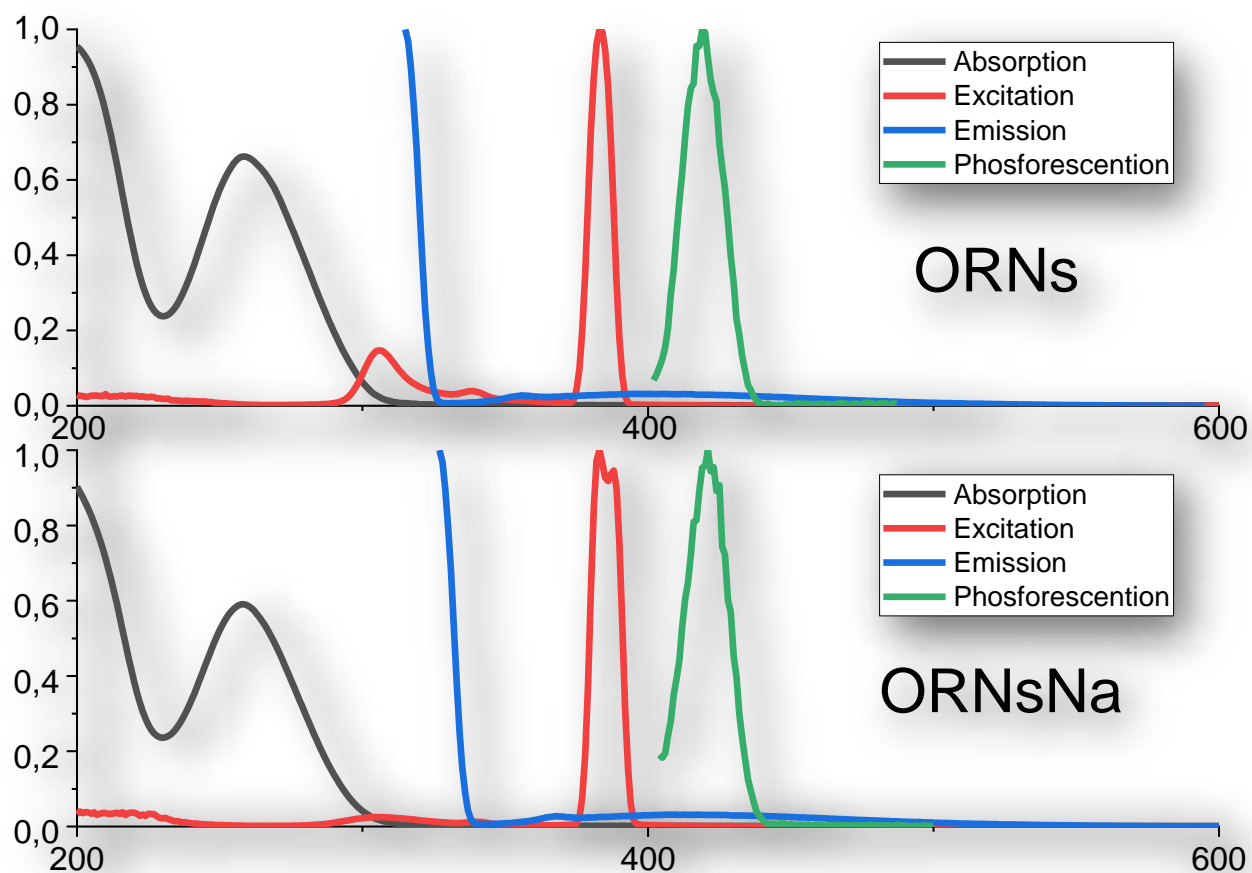
Absorbtion, excitation, emission and phosphorescence spectrum of uracil, uridine, uridine 5`- monophosphate and uridine 5`-monophosphate sodium





Results and discussion

Absorption, excitation, emission and phosphorescence spectrum of uracil, uridine, uridine 5`- monophosphate and uridine 5`-monophosphate sodium





Conclusions

- We studied the absorption and emission of monoribonucleotides and their components dissolved in water at room temperature.
- We observed a change in the ratio between the peaks of the absorption spectra of acidic and salt forms of nucleotides.
- Our observations confirm that nucleotides, nucleosides, and nucleic acid bases exhibit luminescence at room temperature, which may be useful information for further research in this direction.
- The most significant changes in the spectra occur when fluorescence excitation and phosphorescence emission are examined, with acidic forms showing the most pronounced shifts.