**Introduction:** Retinoblastoma is the most common primary ocular malignancy in pediatric age. Knudson proposed his *two-hit* model, allowing the distinction of retinoblastoma in two major classes: heritable and non-heritable. Retinoblastoma was first considered to arise from a well known mutation in the RB1 tumor-suppressor gene (chromosome 13q14). Currently, evidence supports that biallelic inactivation of RB1 gene is the initiating event, but not sufficient for fully malignant progression [1]. The hypothesis of altered expression of p14ARF-MDM2-p53 surveillance pathway components was proposed as an attempt to explain fully retinoblastoma development [2]. Previous studies proposed that p14ARF protein expression was undetectable, in contrast with Mdm2 protein overexpression in retinoblastoma [3].

**Objectives:** The aim of this study was to evaluate the immunohistochemical expression of p53 pathway components (p14ARF, Mdm2 and p53) in order to a better understanding of the molecular pathogenesis and differentiation of retinoblastoma. Additionally, it was attempted to correlate the expression of these proteins with retinoblastoma’s heritable pattern, Reese-Ellsworth staging and vital prognosis.

**Methods:** A cohort of 24 retinoblastoma tissue samples from 22 enucleated cases was obtained from the registry of HUC’s Ophthalmic Pathology Laboratory. Clinical records were consulted to collect information including gender, age, heritable pattern, Reese-Ellsworth stage and prognosis. Immunohistochemistry was performed on formalin-fixed, paraffin-embedded retinoblastoma tissue samples using primary antibodies against p53, p14ARF and Mdm2.

**Results:** Positive p53, p14ARF and Mdm2 expression was obtained in 87.5% (21/24), 87.5% (21/24) and 95.8% (23/24) of the 24 samples, respectively. Overall, p53
protein expression was not positively correlated neither with p14$^\text{ARF}$ (p=0.343) nor Mdm2 expression (p=1.000). In addition, p14$^\text{ARF}$ expression was mainly found in tissue samples that were positive for both p53 and Mdm2. Moreover, we did not obtain a positive relationship between p53, p14$^\text{ARF}$ and Mdm2 expression and the analyzed clinical parameters (heritable pattern, vital prognosis and Reese-Ellsworth staging).

**Conclusions:** In our study, we obtained 87.4% of positive p14$^\text{ARF}$ nuclear and nucleolar expression and we even documented the presence of p14$^\text{ARF}$ overexpression in half of the cases, in opposition to previous reports [3]. According to our results, there was a Mdm2 overexpression in 79.2% of retinoblastoma samples, which supports the hypothesis that MDM2 overexpression may be an important element in retinoblastoma molecular pathogenesis [2,4].

The small cohort of patients involved in this study compromised the final results, which did not show any statistical significance. Further studies need to be performed in order to establish the true prognostic value of these histological markers, using a larger retinoblastoma patient’s population.

**Key-words:** Retinoblastoma, pRB, p53, p14$^\text{ARF}$, Mdm2, immunohistochemistry

**References:**