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Response from S. Zajączek and J. Lubiński

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DOCTORS LOHMANN and Horsthemke summarised published and unpublished results from their laboratory, suggesting that there is no association between the presence of constitutional *Rb-1* mutations and age at tumour diagnosis in patients with sporadic unilateral retinoblastoma (SURB) [1, 2] which is not in agreement with our [3] and Cowell and Cragg's results presented in the *European Journal of Cancer* [4].

The occurrence of *Rb-1* mutations in clinically well-characterised cases of SURB has not been studied intensively. Series of reported consecutive cases of SURB with described age at diagnosis include only 68 cases studied by Lohmann and colleagues and 16 cases studied by our centre [1–3]. Altogether, the number of constitutional mutations in both series is small and include 9 cases diagnosed by Lohmann and colleagues and 3 cases from our studies (1 patient with mutations reported in our paper [3] developed a second eye tumour after 35 months of follow-up (data not shown), thus we excluded him). In such a situation it is obvious that the discussed problem cannot be solved until further investigations are conducted.

Comparing our and Dr Lohmann's studies, we note one very important difference—we identified mutations by complete 'exon by exon' sequencing and Lohmann and colleagues used pre-selection techniques which detect DNA abnormalities with a lower sensitivity. This is of particular importance for the detection of point mutations [5, 6]. In our experience, in cases of SURB the proportion of single base substitutions may be higher.

Thus, further studies on the correlation between the occurrence of *Rb-1* constitutional mutations and the age of tumour diagnosis on larger series studied by full sequencing are needed.

At present, no definitive conclusion on the discussed problem is possible, but our suggestion of the existence of an association between the presence of germline *Rb-1* mutations and early onset of retinoblastomas is supported additionally by the findings of Cowell and Cragg [4] and, what is most important, by the two-hit Knudson hypothesis, which has been verified as appropriate for many other types of hereditary malignancies.

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