phase and blast crisis (AP and BC) of CML may show additional oncogenic aberrations and pronounced anaplasia manifested by an increase in organomegaly and blast count. The abnormal expression of some proto-oncogenes which may accompany or even precede BC of CML warrants their study. *Aim.* The follow-up of oncogene expression during the course of CML. *Methods.* We studied 85 patients (pts.) with the median age opf 50 (range 16-75 years). At the commencement of the study, 29 pts.were in CP, 25 in an AP, and 31 in the BC. The temporal expression (percentage positivity per 1000 analysed cells) of c-kit, c-myc, H-Ras, cyclin A1, p53, bcl-2 and VEGF proto-oncogene proteins over the course of CML was studied using the immunohistochemical tehnique which utilizes relevant monoclonal antibodies. It was correlated with the laboratory (Hb, WBC and platelet counts, and the percentage of blasts) and clinical parameters (organomegaly, duration of CP, AP, and BC) of disease progression. *Results.* The level of c-kit expression differed significantly in time with the largest values observed in the BC (x2, p=0,025). The level of anti-apoptotic protein bcl-2 increased significantly with the progression of CML (x2, p=0,005). Conversely, the expression of c-myc was highest in CP (x2, p=0,033). The expression of VEGF protein was most pronounced in an AP (ANOVA, p=0,033). There was no significant difference in the level of expression of H-Ras, cyclin A1 and p53 over the course of CML. The level of VEGF expression correlated inversely with degree of organomegaly (Pearson, r=-0,400, p=0,011). The c-kit expression correlated directly with the extent of bone marrow fibrosis (Spearman, r=0,407, p=0,000). High expression of VEGF correlated with a longer duration of CP (log rank, p=0,0304) and with a longer overal survival (log rank, p=0,042). *Conclusion.* The significance of changes in oncogene expression, estimated by a histochemical approach over the course of CML, may be of clinical importance in deciding on and timing of therapy. The details of the temporally-related changes in oncoprotein expression in leukemic cells require the study at the molecular level.

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P190 BCR-ABL CHRONIC MYELOID LEUKEMIA PARTLY RESEMBLING CHRONIC MYELOMONOCYTIC LEUKEMIA IN A YOUNG PATIENT TREATED WITH IMATINIB

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Background. In Ph-positive CML, the breakpoint on the BCR gene nearly always occurs within the major breakpoint cluster region (M-bcr), and the BCR-ABL fusion gene encodes a protein of 210 kDa molecular weight (P210). In most Ph-positive ALL, by contrast, the breakpoint on chromosome 22 occurs within the first intron of the BCR gene, or minor bcr (m-bcr). In these cases, the first BCR exon is fused to ABL exon 2 (e1a2 junction) and a Bcr-Abl protein of 190 kDa is formed (P190). This form of CML was reported as having some unusual clinical and haematological features, partly resembling chronic myelomonocytic leukemia (Melo *et al.*, 1994). Here we describe a 24 year-old female patient who presented in July 2005 with leukocytosis, when she volunteered as a blood donor, and was diagnosed as chronic phase CML. *Methods and Results.* She was assymptomatic, with only splenomegaly detected on physical examination. The peripheral blood examination showed a WBC count of 29.7×10^9/L, basophilia (4%), monocytosis (8%) and a platelet count of 713 ×10^9/L. No pseudo-Pelger-Huet hypolobulation or peripheral blood myeloblasts were detected. Bone marrow cytogenetic analysis at diagnosis showed a karyotype 46,XX t(9;22)(q34;q11) in 20 metaphases. Molecular studies detected the presence of an e1a2 transcript. FISH analysis confirmed the m-bcr as the sole type of BCR-ABL rearrangement present in bone marrow cells. She was put on hydroxyurea (HU) 2g daily, with a partial haematological response; 3 weeks later she was started on α-IFN 3 MU/day. Treatment with imatinib was initiated in October at a dose of 400 mg a day and after one month the dose was reduced to 300 mg daily due to moderate toxicity. The patient achieved complete haematological response and remained clinically well. After 3 months of imatinib therapy, the abnormal clone persisted and RT-PCR quantification showed a 50% BCR-ABL/ABL ratio. Despite remaining in complete haematological remission, the abnormal clone persists 4 months after initiation of imatinib therapy. Twenty-one cases of CML with a breakpoint in the m-bcr, resulting in P190 type BCR-ABL have been reported, so far, and only 17 of them in detail. This is to our knowledge, the first P190 CML case reported in a very young patient, in contrast to those previously described whose age ranged from 32 to 83 years (median 53.5) . It remains to be seen whether the long term response to imatinib in this type of CML will compare to that observed in classical P210 cases or will resemble more the poorer response achieved in Ph-positive ALL.

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ANGIOGENIC ACTIVATORS IN PATIENTS WITH CHRONIC MYELOID LEUKEMIA: EFFECT OF TREATMENT WITH IMATINIB MESYLATE

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Background. Angiogenesis is nowadays considered an important factor in biology of various hematological malignancies including chronic myeloid leukemia (CML). Several studies have recently reported elevated levels of angiogenic activators such as vascular endothelial growth factor (VEGF) and basic fibroblast growth factor (bFGF) in CML patients. However, there have been only few data on the influence of imatinib mesylate (IM) treatment on the levels of angiogenic cytokines in CML. *Aims.* To analyze peripheral blood levels of angiogenic activators in patients with newly diagnosed CML and during imatinib treatment. *Methods.* We measured plasma concentrations of VEGF, bFGF and soluble endoglin (sCD105) using sandwich enzyme-linked immunosorbent assay (ELISA) in 16 patients with chronic-phase CML and 80 healthy blood donors; furthemore, repeated samples during the therapy with (IM) were analyzed. *Results.* We found a statistically significant increase in VEGF (mean ± SD [standard deviation], 491.0 ± 365.3 vs. 64.2 ± 69.5 pg/ml, 95% CI [confidence interval] of mean, 296.4-685.7 vs. 51.0-77.5 pg/ml, p<0.0001) and sCD105 (mean ± SD, 7.0 ± 1.95 vs. 4.57 ± 1.51 ng/mL, 95% CI of mean, 5.83-8.18 vs. 4.20-4.93 ng/mL, p<0.0001) but not bFGF (p=0.606) in comparison to the control group. VEGF levels significantly decreased in 7 patients who achieved hematological remission (6 complete remissions, 1 partial remission) during therapy with IM (mean ± SD, 679.6 ± 431.5 vs. 132.7 ± 63.3 pg/ml, 95% CI, 280.6-1078.6 vs. 74.1-191.3 pg/mL, p=0.015). There was no significant change in bFGF or sCD105 (p= 0.938 and 0.125, respectively). *Conclusions.* We found significantly elevated VEGF and sCD105 levels in CML patients. In addition, successful treatment with IM resulted in significant decrease of VEGF. These data lend further support to the importance of angiogenesis in patophysiology of CML. Further studies incorporating larger number of patients are needed to confirm our findings. Supported by research project MZO 00179906 from Ministry of Health of Czech Republic

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THE E19A2 BCR-ABL BREAKPOINT: MORE FREQUENT THAN OTHER ATYPICAL BCR-ABL VARIANTS IN CHRONIC MYELOGENOUS LEUKEMIA?

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In the vast majority of patients diagnosed as having chronic myelogenous leukemia (CML) and t(9;22), the breakpoint on chromosome 22 occurs in the major region of the BCR gene (M-BCR); this translocation usually results in a hybrid BCR-ABL mRNA with a b2a2 and/or b3a2 junction, which encodes a p210 fusion protein proved to be involved in the mechanism that underlines the chronic phase of CML. Here, we report 7 newly diagnosed chronic phase CML patients with an unusual e19a2 BCR-ABL transcript. The BCR breakpoint in this type of rearrangement occurs downstream from M-BCR, in the µ-BCR region, between exons e19 (c3) and e20 (c4). This novel translocation, previously reported by our group in only few patients, results in the transcription of e19a2 type BCR-ABL fusion mRNA, which is translated into a p230-kD BCR-ABL protein. We observed that in some patients e19a2 was associated with neutrophilic leukemia while in the other patients the rare rearrangement was associated with a classical CML in chronic phase. In particular, in a 45-year-old male hemoglobin was 14.7 g/L, white blood cell count 71.8×10^9/L, neutrophils 64%, lymphocytes 8%, monocytes 2%, eosinophils 3%, basophils 5%, metamyelocytes 7%, myelocytes 9%, promyelocytes 2% and platetet count 277×10^9/L. In a 30-year-old female hemoglobin was 9.4 g/L, white blood cell count 108 ×10^9/L, neutrophils 30%, lymphocytes 4.8%, monocytes 4.3%, eosinophils 0.9%, basophils 4.2%, metamyelocytes 30%, myelocytes 20% and platetet count 9.9 ×10^9/L. In all 7 patients cytogenetic analysis of 20 bone marrow