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Редакционна колегия Editorial Board

Българска медицина

Съдържание

Обзори

Оригинални статии

Изисквания към авторите

Българска медицина се реферира в международната база данни Index Copernicus International

Bulgarian medicine

Съдържание

Reviews

Original papers

Author's guidelines

Bulgarian Medicine is included in
Index Copernicus International Journals Master List

Molecular cancer therapy

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Молекулярно лечение на рака

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РЕЗЮМЕ

Злокачествените тумори са едни от най-изследваните заболявания, но прилаганото лечение често е неуспешно. Ракът все още е едно огромно предизвикателство пред медицината и човечеството. Хирургичното лечение, стандартната химиотерапия и лъчевата терапия доказаха своята ефективност при някои видове рак при човека. Голям брой пациенти с рак, за съжаление, не отговарят дори на комбинирано лечение. Необходимостта от създаването и използването на нови подходи в лечението на рака е неизбежна. Проучванията на биологични промени при рака допринесоха за въвеждането на прицелното молекулярно лечение. Избирателното атакуване на молекулярни мишени и възстановяването на увредени противотуморни механизми се налагат като незаменима част от съвременното лечение на рака. Молекулярното лечение на рака засега е свързано със забележителни постижения, но и с много трудности и провали. Молекулярното лечение на рака несъмнено ще помогне в битката с това смъртоносно заболяване.

Ключови думи: рак, цел, молекулярно лечение, проучване

ABSTRACT

Malignant tumors are among the most studied diseases, but the applied treatment is often unsuccessful. Cancer is still a great challenge for the medicine and human race. Surgical treatment, standard chemotherapy and radiotherapy proved their efficacy in some types of human cancers. A large number of cancer patients, unfortunately, do not respond even to combined treatment. The necessity of creation and utilization of new approaches in the cancer treatment is inevitable. The investigations of biological alterations in the cancer contribute for the implementation of the molecular targeted therapy. Selective attack on molecular targets and the restoration of impaired antitumor mechanisms are imposed as irreplaceable part of contemporary cancer therapy. At present, the molecular therapy of cancer is connected with remarkable achievements, but and with a lot of hardships and failures. Molecular cancer therapy undoubtedly will help in the battle with this lethal disease.

Key words: cancer, target, molecular therapy, study

INTRODUCTION

Cancer is a multistage process of malignant transformation of normal cells. Hallmarks of cancer consist of self-sufficiency in growth signals, insensitivity to antigrowth signals, evasion of programmed apoptosis and host immune control, limitless replicative potential, sustained angiogenesis, tissue invasion and metastasis, and disturbed energy metabolism (13). The complex nature of human cancers deters the invention of universal cancer therapy. The action of classical anticancer drugs is based on the interference with the DNA replication machinery resulting in DNA damage, and disruption of cell division by affecting microtubules. Efficacy of chemotherapeutic agents depends basically on increased cell proliferation and impaired ability to repair DNA damage. Chemotherapeutics are supposed to work mostly in cycling cells, although these agents are occasionally effective in therapy of solid tumors with a long doubling time. Unfortunately, chemotherapy is often associated with serious side effects, resistance and unsatisfactory results. Molecular targeted therapy of cancer is an option to overcome the poor prognosis in cancer patients.

MONOCLONAL ANTIBODIES IN CANCER THERAPY

Monoclonal antibodies (mAbs) are among the most exploited agents in targeted cancer therapy that take advantage of the strictly differentiated structure of cell surface (Table 1). Preferably, cancer specific antigens have to be expressed readily and uniformly on cancer cell surface with negligible secretion in order to avoid undesirable binding to circulating antibody. Action of mAbs is based upon connection and blockade of cell receptor/ligand or agonist impact, promotion of immunologic answer, induction of apoptosis, and manipulation of tumor stroma and neoangiogenesis. There are multiple mAbs that inhibit mitogenic pathways directly. Blockade of the extracellular EGFR/HER1 domain with cetuximab disrupts the ligand-independent downstream signaling and reduces proliferation in cancer cells. Cetuximab is applied in the therapy of metastatic colorectal cancer (CRC), head and neck cancer, and in pancreatic cancer in combination with gemcitabine (32). Notably, cetuximab resistance is found in patients with mutant K-Ras. Panitumumab, another EGFR mAb, is used in the treatment of wild-type KRAS refractory metastatic CRC (35). Amplification or overexpression of HER2 receptor is found in approximately 20% of invasive breast cancers. Clinical data confirmed the benefit of HER2 directed mAb trastuzumab, coupled with chemotherapy in patients with HER2-positive advanced breast cancer (12). Unfortunately, despite the benefit of trastuzumab therapy, the disease is almost always progressing. In 2010, FDA approved trastuzumab in combination with cisplatin/fluoropyrimidine for the treatment of HER2-overexpressing metastatic gastric or gastroesophageal junction adenocarcinoma. Promising data ensued from recent phase 2 studies that assessed the efficacy and safety of new drug pertuzumab plus trastuzumab plus docetaxel in HER2-positive breast cancer patients (2). Pertuzumab also inhibits HER2 signaling and induces antibody dependent cell-mediated cytotoxicity (ADCC) like trastuzumab, but the new drug is attached to different epitope of the HER2 extracellular domain and prevents dimerization with other HER receptors. Development of drug resistance via secondary mutations of HER1 or overexpression of hepatocyte growth factor receptor (c-Met) and HER3 is observed during the therapy directed to HER1 and HER2. All these molecular alterations let the targeted downstream signaling pathways to continue uninterrupted.

Bone metastases and cancer related bone pain are extremely unsusceptible to treatment. Denosumab (Xgeva), an agent indicated for cancer patients with metastatic bone lesions, binds the receptor activator of NF-κβ ligand (RANKL) that participates in osteoclast activation. A large randomized study showed that Denosumab (Prolia) could postpone the bone metastasis in

men with prostate cancer (29). Antibodies are also harnessed to inhibit cancer neovascularization. Bevacizumab, directed to all isoforms of angiogenic VEGF-A, breaks the connection between VEGF and its receptor. Bevacizumab, in combination with 5-fluorouracil/capecatibine, is used as a first-line treatment of metastatic CRC (16). Bevacizumab is included also in the therapy of metastatic kidney cancer, advanced non-small cell lung cancer (NSCLC), ovarian cancer, glioblastoma and advanced gastric cancer. Ramucirumab, other promising antiangiogenic mAb against VEGFR-2, is in phase II/III trials for treatment of multitude cancers.

Exposure of phosphatidylserine (PS) on the external leaflet of the plasma cell membrane promotes phagocytosis of apoptotic cells. Bavituximab is a chimeric PS-targeting mAb that induces vascular disruption and promotes antitumor immune response. Therapeutic response has been reached in phase I clinical trial of bavituximab in combination with chemotherapy in patients with advanced solid cancers (9). Three randomized phase II clinical trials with this intriguing antibody are ongoing for NSCLC and pancreatic cancer. Cluster of differentiation (CD) molecules are regular clients of mAbs. Rituximab reacts with human CD20 antigen and attains response in the therapy of non-Hodgkin's lymphoma (NHL). Alemtuzumab, designed to target CD52 antigen found in malignant lymphocytes and monocytes, is utilized in the chronic lymphocytic leukemia (CLL) treatment (10). Action of rituximab and alemtuzumab is based not only on ADCC, but on induction of complement-dependent cytotoxicity and apoptosis too. Use of agonistic mAbs against TNF-related apoptosis-inducing ligand (TRAIL) death receptors DR4 or DR5 also induces impressive apoptotic answer in cancer therapy. Anti-DR4 mAb mapatumumab in combination with hyperthermia showed antitumor activity in colon cancer cells (30). Ongoing study is evaluating the efficacy of mapatumumab in combination with sorafenib in patients with advanced hepatocellular carcinoma (HCC).

Use of conjugated mAbs is enhancing the impact of targeted cancer therapy. Anti-CD30 agent brentuximab, conjugated with toxic antimitotic monomethyl auristatin (vedotin), was approved for the therapy of relapsed or refractory Hodgkin's lymphoma and systemic anaplastic large cell lymphoma (ALCL) by FDA in 2011. Doxorubicin containing liposomes attached to anti-HER2 mAb are introduced in breast cancer therapy to enhance efficacy. Trastuzumab emtansine (T-DM1) is a novel drug used in the therapy of HER2-positive breast cancers. T-DM1 consists of trastuzumab linked to a chemotherapy agent emtansine. Recent data from phase III study showed that T-DM1 increased survival in patients with HER2-positive metastatic breast cancer. Conjugation of mAbs with radioisotopes is also implemented in practice. Ibritumomab is delivering the conjugated radioisotope ^{90}Y in the therapy of B cell NHL, while mAbs against CEA, MUC1, PSMA, EGFR and melanin, labelled with ^{131}I, ^{90}Y, ^{177}Lu, ^{125}I and ^{177}Re, are being evaluated in clinical trials.

Cell adhesion is a unique feature inherent to multicellular organisms. Cell-cell and cell-extracellular matrix (ECM) adhesion is mediated by cell adhesion molecules (CAMs) that are involved in variety of biological processes. Abnormal expression of CAMs is frequently observed in the development of invasive cancers. Epithelial cell-adhesion molecule (EpCAM) is overexpressed in different epithelial cancers and is associated with shorter survival. Catumaxomab is a trifunctional bispecific mAb against EpCAM and CD3 on T cells, whereas Fc region binds to macrophages, dendritic cells and natural killer cells. Catumaxomab produces strong antitumor immune response and therapy with cetuximab was the first approved causal therapy for malignant ascites by the European Medicines Agency (EMA) in 2009. Application of catumaxomab was safe and showed noteworthy clinical responses in the treatment of malignant ascites in ovarian, gastric and other cancers (15). Phase IB study of the adecatumumab, other EpCAM mAb, combined with docetaxel showed safety

Table 1. Approved anticancer drugs with related molecular targets and targeted cancers

Drug	Drug target	Approval	Indication
Monoclonal antibodies			
Cetuximab (Erbitux)	EGFR	EMA and FDA	CRC, HNSCC and pancreatic cancer
Panitumumab (Vectibix)	EGFR	EMA and FDA	CRC
Trastuzumab (Herceptin)	HER2	EMA and FDA	Breast cancer and gastric/ gastroesophageal junction adenocarcinoma
Denosumab (Prolia)	RANKL	EMA and FDA	Prostate cancer (nonmetastatic)
Denosumab (Xgeva)	RANKL	EMA and FDA	Bone metastases from solid tumors
Bevacizumab (Avastin)	VEGF	EMA and FDA	CRC, NSCLC, glioblastoma, kidney cancer, ovarian cancer and gastric cancer
Iplimumab (Yervoy)	CTLA4	EMA and FDA	Melanoma
Rituximab (Mabthera)	CD20	EMA and FDA	B cell NHL and CLL
Alemtuzumab (Campath)	CD52	EMA and FDA	B cell CLL
^{90}Y –labelled ibritumomab tiuxetan (Zevalin)	CD2O	EMA and FDA	Follicular B cell NHL
^{131}I-labelled tositumomab (Bexxar) ^	CD20	FDA	Follicular lymphoma
Brentuximab vedotin (Adcetris)"	CD30	FDA	Hodgkin's lymphoma and ALCL
Catumaxomab (Removab)	EpCAM, CD3	EMA	Malignant ascites
Smal molecule inhibitors			
Erlotinib (Tarceva)	EGFR	EMA and FDA	NSCLC and pancreatic cancer
Gefitinib (Iressa)	EGFR	EMA and FDA	NSCLC
Lapatinib (Tykerb) ¶	EGFR, HER2	EMA and FDA	Breast cancer
Sorafenib (Nexavar)	B-Raf, VEGFR, PDGFR, EGFR, FLT3	EMA and FDA	Liver cancer and renal cancer
Imatinib mesylate (Glivek)	BCR-ABL, KIT, PDGFR	EMA and FDA	CML, GIST, myelodysplastic-myeloproliferative diseases, dermatofibrosarcoma, precursor cell lymphoblastic leukemia-lymphoma and hypereosinophilic syndrome
Sunitinib (Sutent)	KIT, VEGFR, PDGFR, FLT3	EMA and FDA	Renal cancer, GIST and pancreatic neuroendocrine tumors
Crizotinib (Xalkori)#	ALK, c-Met	FDA	NSCLC
Gene delivery			
Recombinant human p53 adenovirus injection (Gendicine)	p53	SFDA	HNSCC
Demethylation agents			
Decitabine (Dacogen)##	DNMT	FDA	MDS
Azacitidine (Vidaza)	DNMT	EMA and FDA	MDS
Histone deacetylase inhibitors			
Vorinostat (Zolinza)	HDAC	EMA and FDA	Cutaneous T cell lymphoma
Romidepsin (Istodax)*	HDAC	FDA	Cutaneous T cell lymphoma
Proteasome inhibitors			
Bortezomib (Velcade)	26S proteasome	EMA and FDA	Multiple myeloma

and activity in patients with EpCAM positive relapsed or refractory advanced-stage breast cancer (26). Data from a preclinical study conclude that new anti-EpCAM mAb conjugates with α-amanitin (inhibiting DNA transcription toxin) are effective drugs for the therapy of pancreatic carcinomas and other EpCAM-positive cancers (23). Integrins, large transmembrane receptors composed of α and β subunits, influence proliferation and migration of malignant cells. Integrin α5β1 is abundantly expressed on endothelial cells of tumor vessels, as well on tumor cells. Volociximab is a mAb that targets α5β1 integrin and inhibits tumor neoangiogen-

esis by blocking the interaction between α5β1 and fibronectin. Phase II and III trials with volociximab are ongoing now for the therapy of metastatic melanoma, NSCLC and peritoneal cancer. Cytotoxic T lymphocyte-associated antigen 4 (CTLA4) is a prominent member of CAMs, which decreases the activity of T cells. CTLA4 inhibitor ipilimumab is approved to treat patients with metastatic melanoma

Tumor microenvironment plays a significant role in malignant progression, and occasionally the collapse of stroma signaling is the primary cause of cancer. Stromal and ECM molecules are putative targets, because restitution of normal microenvironment could even provoke reversal of neoplastic phenotype. Fibroblast activation protein (FAP) is a cell surface protein expressed by tumor-associated fibroblasts in majority of human carcinomas. Sibrotuzumab, a mAb against FAP-α, however, was not effective in a phase II study of patients with CRC. Collagen and fibrin, overexpressed in stroma of solid tumors, are used as mAbs targets. Application of conjugated with a topoisomerase I inhibitor mAb against collagen 4 is an attractive chance to improve the diffusion of mAbs in rich in stroma solid tumors (38). Tenascin-C, part of ECM, is abundantly found in solid tumors and mAbs against tenascin-C are developed. Eph receptors and ephrin ligands participate in regulation of cell adhesion, morphology and invasion, and aberrant Eph signaling is found in cancer. Agonistic and antagonistic mAbs against Eph are being tested in cancer therapy (4).

^Orphan designation granted in 2003 in European Union. „Pending decision with positive status in European Union. ¶Conditional approval in European Union. #Pending decision with positive status in European Union. ##Pending decision with positive status in European Union with indication acute myeloid leukaemia. *Pending decision with negative status in European Union. ALCL, Anaplastic large cell lymphoma; CLL, chronic lymphocytic leukemia; CML, chronic myelogenous leukemia; CRC, colorectal cancer; CTLA-4 (Cytotoxic T lymphocyte-associated antigen 4), DNMT, DNA methyltransferase; EGFR, epidermal growth factor receptor; EMA, European Medicines Agency; EpCAM, epithelial cell adhesion molecule; FDA, Food and Drug Administration; FLT3, fms-like tyrosine kinase 3; GIST, gastrointestinal stromal tumors; HDAC, histone deacetylase; HNSCC, head and neck squamous cell carcinoma; MDS, myelodysplastic syndrome; NHL, non-Hodgkin's lymphoma; NSCLC, non-small-cell lung carcinoma; PDGFR, platelet-derived growth factor receptor; RANKL, receptor activator of NF-κβ ligand; SFDA, State Food and Drug Administration; VEGFR, vascular endothelial growth factor receptor.

SMALL MOLECULE INHIBITORS IN CANCER THERAPY

Abnormal expression of protein kinases is often found in human cancers and small molecule inhibitors (SMIs) suppress activity of kinases (Table 1). In contrast to large mAbs, SMIs are compounds of less that 500-D molecular weight, except for some natural substances. Unlike mAbs, SMIs are able to pass through plasma membranes and interact with cytoplasmic part of cell-surface receptors and intracellular molecules. SMIs targets kinases directly and an immune response is not commonly involved as in the therapy with mAbs. Most of the SMIs compete with the ATP-binding site in active or inactive conformation of the tyrosine kinases. Allosteric inhibitors act in non-ATP targeted manner, whereas covalent SMIs are irreversible kinase inhibitors.

Activity of the oncogenic BCR-ABL fusion protein in chronic myelogenous leukemia (CML) is inhibited by imatinib that reacts with the inactive form of tyrosine kinase (TK). Despite spectacular advance in the treatment of CML, imatinib is not able to cure definitely CML. BCR-ABL amplification and mutations induce resistance to imatinib, as well to other BCR-ABL targeting agents (7). Unlike mAbs, SMIs are less specific and imatinib is also used to suppress activity of c-kit in gastrointestinal stromal tu-

mors (GIST) and activity of PDGFR in chronic myeloproliferative syndrome. The therapy of GIST with imatinib also changed the natural course of the disease. Nevertheless, almost half of the treated patients do not respond and complete responses are rare. Genetic switch of the malignant cells could compromise the therapy of GIST with imatinib. Mechanism of resistance to c-Kit-directed therapies is based on the receptor TK (RTK) switch as a result of downregulation of c-Kit, coupled with upregulation of RTK c-Met. Sunitinib, a multiple TK inhibitor, is used to treat resistant to imatinib GIST, as well renal cell carcinoma (RCC) and pancreatic neuroendocrine tumors. Salvage therapy with sunitinib in GIST, however, does not work in resistant to imatinib patients with exon 11 mutation, because of secondary mutations (14). Similar to mAbs, SMIs are produced against HER1 and HER2. Erlotinib and gefitinib are SMIs designed to aim the TK domain of HER1. Erlotinib, interacting with active form of kinase, increased survival rates in patients with NSCLC, whereas gefitinib did not attain efficacy. Erlotinib, in combination with gemcitabine, is also approved for therapy of pancreatic cancer. Lapatinib, a reversible dual HER1/HER2 inhibitor is used for the treatment of HER-2-expressing breast cancer resistant to trastuzumab. Neratinib, an HER1 and HER2 irreversible inhibitor demonstrated remarkable benefit in HER2 positive breast cancer patients (5). Allosteric SMIs are used for suppression of abnormally expressed in many cancers proto-oncogen c-Met. An oral c-Met and anaplastic lymphoma kinase (ALK) ATP-site SMI crizotinib showed efficacy and in 2011 FDA approved crizotinib for use in patients with advanced NSCLC with a rearrangement of ALK gene (21). Mutations in the RET protooncogene are tightly associated with the development of hereditary and sporadic medullary thyroid carcinoma (MTC), and RET-relevant SMIs have been investigated. Motesanib, a multitargeted inhibitor against RET, VEGFRs, c-Kit and PDGFRβ, showed partial responses in a phase II study in locally advanced and metastatic MTC (27). Phase 1b study with Motesanib in combination with erlotinib and gemcitabine also showed tolerability in patients with solid tumors (20). Mutations and amplifications in the catalytic subunit p110α of phosphoinositide 3' kinase (PI3K) are often detected in cancers, and SMIs against PI3K are undergoing clinical evaluation (37). B-raf serine kinase, a part of the RAS pathway, is overexpressed in cancers. Originally, oncogenic B-raf kinase seemed to be the only target of SMI sorafenib, but later it was found that sorafienib is also active against VEGFR-2 and 3, FLT-3, c-Kit and PDGFRβ. Sorafenib is approved for advanced HCC and RCC therapy, although it was unsuccessfully used in melanoma treatment. TGF-β participates in different cellular processes and elevated amounts of secreted TGFb are associated with advanced stage and dismal prognosis in various malignancies. Suppression of TGFb1 with SMIs increased the radiosensitivity of breast cancer cells in vitro and restrains tumor growth in vivo (3). Cyclin-dependent kinases (CDKs) participate in the control of cell cycle and possess potential oncogenic properties. Flavopiridol, a synthetic flavone, is a first generation potent CDKs SMI, which is used with contradictory results to prevent cell proliferation in cancer cells (22). Second generation CDKs SMIs are expected to show better selectivity and less side effects. SMIs are harnessed and for inhibition of antiapoptotic molecules. Navitoclax, Bcl-2 SMI, showed efficacy in patients with a small-cell lung cancer in a phase I study (11). Levo-gossypol, a natural SMI that targets Bcl-2, Bcl-Xl, and Mcl-1, is used now in clinical trials for CLL and prostate cancer. A derivative of gossypol, apogossypol, is in preclinical development.

Malignant cells often initiate DNA repair during chemo- or radiotherapy and suppression of DNA repair molecules could counter this undesired answer. Poly(ADP-ribose) polymerase (PARP)1 is active in DNA base excision repair pathway dealing with base lesions and single-strand breaks, whereas breast cancer type 1 susceptibility protein (BRCA)1 and breast

cancer type 2 susceptibility protein (BRCA)2 act in DNA double-strand breaks (DSBs) repair through homologous recombination (HR) repair pathway. Therefore, downregulation of PARP1 with SMIs in tumors lacking BRCA1/2 could lead to unresolved DNA damage and cell demise. Olaparib, a PARP inhibitor, showed efficacy in BRCA-deficient ovarian and breast cancers (1, 34). New approach is to attack an entire DNA repair system in cancer cells. Dbait oligonucleotides inhibit both HR and nonhomologous end joining pathways employed for repair of DNA DSBs. Recent study demonstrated efficacy of Dbait paired with chemotherapy in mouse models of CRC (8).

p53, guardian of the genome, plays a key role in the cell cycle control, apoptosis and senescence, and is mutated in the majority of human cancers. Nutlin-3 and other antagonists of MDM2 (p53 negative regulator) are being assessed in preclinical trials (6). Oncogenic mutant p53, however, could also be detached from MDM2 and overexpressed as a result of MDM2 targeting. Inhibition of MDM2, as well targeting other antiapoptotic molecules, provokes questions and concerns about the unexpected outcomes of molecular cancer therapy. Moreover, enhanced apoptosis could induce high proliferation rate and liver cancer in mice with deleted antiapoptotic protein Mcl-1(36).

SMIs and mAbs are usually well tolerated and possess mild side effects, including fatigue, cutaneous (acneiform rash, dry skin, pruritus and alopecia) and gastrointestinal symptoms (diarrhoea, vomiting and nausea), although serious side effects (liver, heart and lung toxicity) are reported. Hypertension, bleeding and venous thromboembolism could occur during antiangiogenic therapy. Notably, SMIs and mAbs do not affect bone marrow like chemotherapeutic agents.

GENE DELIVERY IN CANCER THERAPY

Most of the targeted cancer therapies depend on the "oncogene addiction", but the delivery of tumor suppressor genes in malignant cells is other exiting option (Table 1). Reintroduction of proapoptotic genes could restore the antitumor ammunition. Adenoviral vector producing wild-type p53 is approved in China for treatment of patients with head and neck cancer (28). An analogous delivering p53 drug, however, was not approved for use in the USA and Europe, because of the unsatisfactory efficacy. Gene transfection of p53 is also tested in phase I/II clinical trials for therapy of esophageal, ovarian cancer and lung cancer (19). In a recent study, the transfer of carrying p16 gene adenovirus vector induced anoikis (anchorage-dependent apoptosis) in HCC cells by reducing the expression Akt/survivin signaling (17). Delivery of the proapoptotic Bax gene caused apoptosis in glioma cell lines (18). Promotion of the immune system with cytokine genes (IL-2, IL-4, and IL-12) delivery produced conflicting results in the cancer therapy.

Delivering of toxic genes by viral vectors to tumor cells is used in suicide or prodrug activation gene therapy. Inserted genes code enzymes, which turn harmless prodrug into a toxic molecule. Transferring of herpes simplex virus thymidine kinase gene in cancer cells is followed by treatment with the prodrug ganciclovir, which is transformed by thymidine kinase to an apoptosis inducing toxin. Cytochrome P450 converts the prodrug cyclophosphamide to its active form, whereas cytosine deaminase deaminates the nontoxic 5-fluorocytosine to a toxic 5-fluorouracil. Efficacy of suicide gene therapy is still not confirmed, despite encouraging results from some studies. The basic hardships in the gene therapy are the poor potency and the uncertainty of the currently used viral or nonviral vector systems. Safety must be cautiously debated too. Unfortunately, a spectacular clinical breakthrough in the gene cancer therapy is still completely elusive.

RNA-BASED CANCER THERAPY

Gene silencing of oncogenic molecules is an elegant method to fight malignant cells. Antisense oligonucleotides (ASOs) are single-stranded

DNA-like molecules that are complementary to certain mRNA and turn off genes through inhibiting translation of the mRNA into a functional protein. Oblimersen sodium, a Bcl-2 ASO showed efficacy in clinical trial for therapy of CLL (25). Oblimersen application, however, was not approved, because of unsuccessful therapy of melanoma and oblimersen associated side effects. Poor specificity and short half-life are among the reasons for the failure of older ASOs. Encouraging data, however, are reported in ongoing clinical trials with new ASOs directed against antiapoptotic molecules like clusterin, survivin and STAT3. Ribozymes and especially hammerhead (Hh) ribozymes were utilized to suppress the gene expression by cleavage of its mRNA. Angiozyme (RPI.4610), an antiangiogenic ribozyme targeting VEGFR-1 mRNA showed safe profile and clinical activity in solid tumors in preclinical and phase 1 studies. Phase 2, multicenter study, however, did not demonstrate clinical efficacy of angiozyme in patients with metastatic breast cancer (24).

The paradigm of human cancer is changing with the accumulating data for microRNAs (miRNAs). MicroRNAs are small endogenous noncoding RNAs (ncRNAs), which control gene expression by binding specific mRNA for translational repression and/or degradation. MicroRNA pathway is related with RNA interference (RNAi) pathway, a process of post-transcriptional gene silencing in which small duplex RNAs suppress expression of specific genes. Expression of miRNAs is deregulated in cancers and ASOs against miRNAs are undergoing preclinical evaluation. Notably, the expression of most miRNAs in human malignancies is lower than normal. The let-7 miRNA restrains expression of the oncogenes Ras, Myc and HMGA-2, and let-7 is downregulated in many cancers. Encouraging results are achieved in preclinical studies with the delivery of miRNAs let-7 and miR34 in lung tumors (33). Other intriguing posttranscriptional gene silencing molecules are synthetic small-interfering RNAs (siRNAs) that also catalyze the degradation of complementary mRNA targets. Preclinical studies with antioncogenic siRNAs showed appreciable results. Atu-027 is a blunt-ended siRNA directed to protein kinase N-3 that mediates malignant cell growth downstream of activated PI3K pathway. Atu-027 was well tolerated in patients with advanced solid tumors in recently completed phase I clinical study (31). The hurdles for the widespread clinical use of siRNAs are: destruction of siRNAs by serum nucleases, poor tissue penetration, "off-target-effects" and excessive immune response. Interestingly, excessive interferon release induced by siRNAs could enhance anti-tumor immunity. Various chemical modifications are being tested to improve the plasma stability of "naked" siRNAs. Viral (adenovirus, adeno-associated virus and lentivirus) and nonviral (proteins, antibodies, polymers, nanoparticles and lipid molecules) modes for RNA delivery exist, but the poor intracellular delivery of small RNA remains the primary obstacle to RNA-based therapy.

METHYLTRANSFERASE AND HISTONE DEACETYLASE INHIBITORS IN CANCER THERAPY

Promoter CpG island hypermethylation of tumor suppressor genes is an attribute of cancers and DNA methyltransferase (DNMT) inhibitors are used for the reactivation of abnormally methylated tumor suppressor genes. Available data, however, challenge the hypothesis that DNMT inhibitors induce considerable expression of tumor suppressors. A speculation connects the action of these agents with antiproliferative disruption of ribosome biogenesis. Hypomethylating agents like azacitidine and decitabine (nucleoside analogues) are approved for therapy of patients with myelodysplastic syndrome, although in 2012 FDA rejected to add acute myeloid leukemia as indication for use of decitabine (Table 1). The chance for expression of suppressed oncogenes during therapy with hypomethylating drugs is considered low. Histone deacetylases (HDACs) participate in gene transcription control and aberrant expression

of HDACs is observed in cancers. Histone deacetylase inhibitors (HDACi) induce proliferation arrest or apoptosis of tumor cells and comprise hydroxamic acids, cyclic tetrapeptides, short chain fatty acids and benzamides. FDA approved HDACi vorinostat (hydroxamic acid) and romidepsin (cyclic peptide) for therapy of advanced cutaneous T-cell lymphoma, while novel HDACi are currently being evaluated in patients with solid tumors and hematological malignancies (Table 1).

HEAT SHOCK PROTEIN 90 AND PROTEASOME INHIBITORS IN CANCER THERAPY

Heat shock protein 90 (Hsp90), a chaperone engaged in the protein folding, is necessary for activation and stabilization of various signaling proteins. Hsp90 is often upregulated in tumors and Hsp90 inhibitors represent suitable cancer therapeutic tools. Geldanamycin, an Hsp90 inhibitor, showed substantial toxicity in clinical trials. New generation of Hsp90 inhibitors with decreased toxicity and improved potency are under clinical evaluation now. Proteasome, a multicatalytic protease complex that degrades intracellular polyubiquitinated proteins, is used as a target in cancer therapy. Bortezomib is the first proteasome inhibitor approved for therapy of multiple myeloma (Table 1). New proteasome inhibitors are rigorously investigated in early phase clinical studies. Combined inhibition of Hsp90, proteosome, DNA methyl transferase and HADCs is showing promising results in clinical trials.

CONCLUSION

Molecular cancer therapy is relying on multiple approaches. The advancement in the targeted treatment of malignancies is irrefutable, but drugs that can cure the patients of cancer are still wanting. Although with less severe adverse effects, targeted cancer therapy is almost always an additive instrument to the mandatory chemotherapeutic agents or radiotherapy. The future efforts must be concentrated on the invention of novel molecular drugs that are much more potent and safe. These drugs have to attack simultaneously all cancer related molecular culprits, including and dormant cancer stem cells. This dream seems unfeasible, but multiple assaults on cancer network could be the only way to overcome the therapeutic resistance. We must be conscious of the fact that cancer signaling networks are unimaginable sophisticated and ingeniously reactive to alterations in cell milieu. The manipulation of the cell machine is a subtle art with occasionally unpredictable consequences.

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PREVALENCE OF BENZODIAZEPINE MISUSE IN PATIENTS CONSULTED IN AN EMERGENCY DEPARTMENT

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Честота на вредната употреба на бензодиазепини при пациенти, консултирани в спешно отделение

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РЕЗЮМЕ

Проблемната употреба (вредна употреба и зависимост) на бензодиазепини е отдавна известен феномен в медицината, но редица съвременни проучвания показват, че през последните две десетилетия честотата и отновно бележи ръст в някои региони на света. В България има малко актуални епидемиологични данни за разпространението на този проблем. Във връзка с това проведохме малко проучване на честотата на вредна употреба и зависимост към бензодиазепини в извадка от пациенти потърсили лечение в спешно отделение на голяма университетска болница.

Посредством структуриран въпросник за употребата на бензодиазепини бяха изследвани 243 пациента обърнали се за консултация към спешно отделение на УБМАЛ „Д-р Г. Странски“ – Плевен за 8 седмичен период (януари-февруари 2011). Резултатите ни показаха, че 37.5% от пациетите (89 от 243 пациента) употребяват бензодиазепини, като 48 от тях покриват диагностичните критерии на МКБ-10 за вредна упо-

ABSTRACT

Objective: Some recent epidemiological data show that benzodiazepine misuse (harmful use and dependence) may be on the rise globally. Because of the insufficient data regarding this problem in our country we made a small exploratory study of the prevalence of benzodiazepine use and misuse in patients consulted in the emergency department of Pleven University Hospital.

Patients and methods: Data regarding benzodiazepine use along with demographic information were collected for 243 patients admitted to the department for an eight weeks period. Information about medications used and daily dose, duration of use, prescribing physician and reason for prescription were gathered by means of an interview and analyzed.

Results: 89 of the 243 examined patients reported benzodiazepine use, and 48 satisfied ICD – 10 criteria for harmful use or dependence. No difference was found between sexes regarding relative prevalence of BZDs misuse (harmful use and dependence), but female patients as a

треба или зависимост. Употребата на бензодиазепини беше по-честа сред жените в извадката, без разликата да достига статистическа значимост. Подължителността на употреба на бензодиазепини беше най-голяма (над 2 години средно) сред възрастовите групи 40–49 г. и 50–59 г.

В мнозинството от случаите бензодиазепините бяха предписвани от личните лекари поради проблеми със съня и симптоми на тревожност, но също така и поради чисто соматични симптоми погрешно преценени като психологично обусловени. Основният извод, който се налага освен нуждата от допълнителни изследвания в тази област, е че е необходимо да се разработят и въведат надеждни алгоритми свеждащи предписването на бензодиазепини само до пациентите с реални показания за използването и.

whole had longer mean duration of BZDs intake. The majority of patients used BZDs with short elimination half life. Although sleep problems and anxiety symptoms were leading reasons for prescription in most of the cases, a notable number of patients were given BZDs for purely somatic symptoms without sufficient rationale. Nearly 80% of the patients had received their treatment by GP. None of the individuals diagnosed with misuse had informed their prescribing physician about that.

Conclusions: High prevalence of BZDs use and misuse was observed in the sample as a whole with female patients having longer duration of use. One possible inference from such findings is that more precise prescribing guidelines for BZDs are needed in our country.

Key words: benzodiazepines, prevalence, misuse, harmful use, dependence

INTRODUCTION

Developed in the 1960s in the process of research for safer and more effective anxiolytics, benzodiazepines (BZDs) became the most widely prescribed drugs in the United States and Europe between 1968 and 1987 (1). During last two decades they have maintained their popularity and diazepam along with alprazolam, clonazepam, and lorazepam, has appeared among the most commonly used medications worldwide (2).

The risk of abuse and dependence associated with BZDs was recognized as early as 1967, as the first reports in the popular media emerged warning of their illicit and non-medical use. (1; 3). In 1975, the United States Drug Enforcement Agency began regulating valium and several other benzodiazepines as Schedule IV drugs and gradually in the next several years more judicious prescribing practices for BZDs were established in US and elsewhere.

Although BZDs misuse potential is not a new notion, recent epidemiological findings suggest that their abuse may be on the rise. In USA for example, the number of emergency room visits associated with the use of sedative/hypnotics in 2005 was 34% of the total visits involving non-medical use of prescription drugs, thus registering an increase with 19 % compared to 2004 (4). In agreement with this, admissions in substance abuse treatment facilities in the United States, due to "primary tranquilizer" use (including benzodiazepine type drugs) increased with 79% from 1992 to 2002 (4). Data suggesting stable or increasing consumption of BZDs in the last two decades are available also for Italy, Finland, Canada, Brazil, Australia, Thailand and other countries (5; 6; 7). The rise is particularly due to increase in consumption of the short acting drugs at the expense of a proportional decline in use of older compounds with longer half-lives (8).

Within the general population there are specific sub-populations who are at greater risk for inappropriate benzodiazepine taking. These groups include polydrug abusers, patients with histories of alcohol abuse and the elderly (9; 10). In virtually every population studied, women receive about twice as many prescriptions for these drugs as men. (8).

In Bulgaria benzodiazepines are included in a list of controlled medications and their

use is regulated through a special regime of prescription which enables medical authorities to monitor and regulate prescription rates. Despite that, to our knowledge there are no recent epidemiological studies exploring trends in benzodiazepine prescription and use in our country. According to the latest nationwide epidemiological survey EPIBUL (11) the 12 month prevalence for substance related disorders is estimated to 1.2 % of the adult population but most of these cases have alcohol abuse or dependence and no data concerning BZDs use are specified.

Assuming the possibility that inappropriate benzodiazepine consumption may be an underestimated phenomenon with potential public health significance, we decided to conduct a small study of the prevalence of BZDs misuse (abuse and dependence) among patients presenting at the emergency department of our hospital.

PATIENTS AND METHODS

Pleven University Multipfofile Hospital is one of the leading hospitals in Bulgaria with annual admission rate of over 40 000 patients. Because of the large number of emergency cases (nearly 2000 patients per month) and the limitations of our resources, we included in our sample only the patients consulted at the emergency department during fourteen 12 hour shifts spanning over a 8 week period (January – February 2011). Thus, a total of 243 patients were examined which is approximately 10% of all the patients admitted to the department for the same period. All data were obtained by means of a short structured interview specially developed for the purposes of the study and conducted by the same investigator for all the patients. They were asked to report all BZDs used, duration of intake, daily dosage taken, reason for prescription and the position of prescribing physician. In addition, data on demographics were collected. Diagnoses of BZDs misuse (harmful use – F10.1 or dependence – F10.2) were based on the 10-th revision of the International Classification of Diseases (ICD-10).

Data were analyzed using the principles of thematic analysis and incorporating the data-driven inductive approach as the study was primarily observational by nature and did not seek to prove a preconceived theory. Quantitative variables were compared using Fischer's t test and ANOVA with Bonferroni test being used for multiple quantitative comparisons. For comparison of proportions, we used Pearson $\chi 2$ test with Yates correction being applied for small numbers. Statistical analysis was conducted electronically with "Statistical Package for the Social Services" (SPSS) ver. 16.0.

This study was approved by the Independent Ethics Committee of Pleven University Hospital.

RESULTS

Of the total of 243 patients admitted to the emergency department during the study period 89 reported BZDs use (51 women and 38 men). Upon administration of the structured interview, 48 of them satisfied the ICD-10 criteria for harmful use of benzodiazepines or benzodiazepine dependence. In all these cases duration of BZDs use was substantially longer than the recommended in the short product characteristics of the particular drug (8–12 weeks for bromazepam, alprazolam, clonazepam, diazepam and clorazepate and 2 weeks for triazolam and midazolam) and symptoms of withdrawal or other symptoms associated with BZDs use were identified to be primary reason for consultation.

Sociodemographic data of the patients are presented on table 1. Although women as a whole were diagnosed with harmful use more often than men, no statistical difference could be found between sexes in terms of prevalence of harmful use and dependence even after controlling for number differences ($p\chi 2=0.26$).

Tabl. 2 shows all BZD preparations taken. None of the patients used more than one BZD

and only 10 of the patients were prescribed a medication with long elimination half-life (elimination half-life of the BZD or metabolite > 24 h, for example diazepam or clorazepate). The mean age of those patients taking BZDs with short half life (46.3 years) did not differ significantly from those receiving long acting BZDs (45.5 years) (p=0.43).

Tabl. 1:
Sociodemogpaphic characteristics of benzodiazepine users

Characteristics	Total sample	Harmful use	Dependence
Gender (Males/ females)	26/22	2/5	24/17
Mean age (with SD) – Males – Females	48.27 ± 6.78 55.32 ± 9.02	40.50 ± 4.95 47.00 ± 9.14	48.92 ± 6.57 57.76 ± 7.60
Residence – Principal town – Village/ Non-principal town	31 17	4 2	23 12
Occupational status – Employed – Unemployed	37 11	5 1	27 8
Marital status – Married – Single	39 9	2 4	21 14

Tabl. 2:
Benzodiazepine preparations used

Benzodiazepine	Number of patients
Short elimination Half-life (< 24 h) – Clonazepam – Bromzepam – Alprazolam – Triazolam – Midazolam	 34 14 23 5 3
Long elimination Half-life (>24 h) – Diazepam – Clorazepate	 7 3

The mean duration of benzodiazepine intake was 32.40 months for the group of female patients with harmful use and dependence (tabl. 3). For males, mean duration of intake was 23.50 months. Duration of intake was not calculated for patients considered not to have benzodiazepine misuse. When we analyzed in a multiple comparison test the difference in mean duration of DZDs use (in months) between the separate age groups (30–39 years; 40–49 years; 50–59 years; 60–69 years), we found statistically significant difference between groups 40–49 years and 50–59 years respectively (F=3.595; p=0.021) (tabl. 3).

According to prescription indices, no excessive dose was taken by any patient.

Tabl. 3
Mean duration of benzodiazepines intake in patients with harmful use and dependence distributed by sex and age group i

Characteristics	Mean duration of benzodiazepines intake (months)	Ststistical significance
Sex – Males (N=26) – Females (N=22)	 23.50 ± 4.02 32.36 ±5.60	
Age group – 30–39 years (N=5) – 40–49 years (N=19) – 50–59 years (N=16) – 60–69 years (N=8)	 26.2 ± 3.27l 24.35 ± 4.91 30.25 ± 6.63 30.62 ± 7.98	 F = 3.595 (p=0.021)

Most of the patients had complaints of insomnia and anxiety spectrum symptoms. Primary complaints that have lead to BZDs prescription and type of prescribing physician are summarized on tabl. 4 and 5 respectively. Although no cases of self-medication were found, it is worth noting that none of the patients diagnosed with harmful use or dependence had informed his or her prescribing physician about the problems associated with BZDs use.

Tabl. 4:
Distribution of patients according to their primary complaint

Symptom/ complaint	Use (Total; males/ females)	Harmful use (Total; males/ females)	Dependence (Total; males/ females)
Insomnia	20 (15/5)	4 (1/3)	12 (4/8)
Palpitations/chest discomfort	4 (1/3)	0 (0/0)	5 (1/4)
Inner tension/Anxiety	11 (5/6)	1 (0/1)	10 (3/7)
Headache	6 (2/4)	1 (0/1)	5 (1/4)
Hypertension	0 (0/0)	0 (0/0)	1 (0/1)
Other somatic symptom/discomfort	7 (3/4)	0 (0/0)	3 (1/2)

Tabl. 5
Position of prescribing physician

Medical practitioner	Use	Harmful Use	Dependence
General practitioner/ Family physician	36	5	29
Psychiatrist	10	–	4
Other medical specialist	2	1	3

DISCUSSION

As far as our knowledge goes, this is the first study in our country to evaluate the use of BZDs in a sample of patients consisting of visitors of an emergency department. Our data showed that 35.7 % of the examined patients (89 out of 249 cases) were currently prescribed BZDs. More than half of them (48 patients) satisfied ICD-10 criteria for harmful use/dependence and their primary reason for consultation were symptoms/complaints induced by BZDs use. Because of the methodological approach with questionnaire, and because psychotropic agents are among the most frequently underreported drugs used (12) these figures may underestimate the true prevalence of BZDs use.

Regarding the mean duration of BZDs use, a significant difference was found between the age groups of 40–49 years and 50–59 years with mean duration of intake of 24.35 months and 30.25 months respectively. These results are in line with other reports in literature exploring BDZs use in particular age groups (13).

Although no statistical difference was found between sexes in terms of prevalence of harmful use and dependence, the overall proportion of BZD users was higher among women, a finding that is consistent with many other studies (14; 15). One reason for this might be the greater prevalence of sleep disorders in the female population. Nevertheless, alternative explanations are also possible like for example higher levels of emotional stress, respectively higher incidence of stress induced somatic complaints, increased frequency of physician contacts and as a result greater use of prescription medications (14). Another finding deserving attention is the longer duration of BZDs intake in women. It is well known that long administration of BZDs provokes psychological dependency and abstinence reactions on withdrawal, both supporting chronic drug misuse (16). This mechanism may at least in part explain the higher number of female patients with harmful use and dependence that we found.

Although as expected most of the patients in the sample were prescribed BZDs because of sleep problems and symptoms of anxiety, it is important to note that in non-ignorable number of cases BZDs have been given for purely somatic complaints that have been misjudged to have "neurotic" basis. Moreover, in the majority of these cases (and especially in all patients with palpitations/chest discomfort) BZDs were combined with other symptomatically oriented drugs. Even in patients that were prescribed BZDs because of psychiatric symptoms, this was mainly done by GPs who often base their drug prescriptions on the most evident phenomenology not having the time and resources to obtain sufficient information, establish a correct diagnosis and implement proper pharmaceutical therapy.

CONCLUSIONS

In summary, our results showed high prevalence of BZDs use and misuse in the examined sample as a whole with female patients in particular having greater BZDs prescription rates and longer duration of BZDs use. One of the possible implications of such finding is that more judicious prescribing guidelines for BZDs should be developed and implemented especially for outpatient healthcare providers. Nevertheless, more studies with similar populations are in need to elucidate the prevalence of patients with unreported BZDs misuse and its causes.

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Cognitive impairments in brain tumors – does localization really matter?

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Когнитивни нарушения при мозъчни тумори – има ли значение локализацията?

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РЕЗЮМЕ

Степента на нарушенията на функционалната цялост в рамките на фронталните невронни мрежи, предизвикани от патологичния процес, намира отражение в резултатите от прилагането на чувствителни невропсихологични тестове.

ЦЕЛ

Да се извърши проспективно проучване на специфични неврокогнитивни нарушения зависещи от локализацията на мозъчните тумори.

МАТЕРИАЛИ И МЕТОДИ

В проучването са включени 38 пациента със супратенториални мозъчни тумори, които са разпределени в две подгрупи в зависимост от лока-

ABSTRACT

The extent of impairment of functional integrity within the frontal lobe network, caused by the pathological process, can be assessed by means of sensitive neuropsychological tests.

AIM

To prospectively investigate specific cognitive impairments depending on the brain tumor localization.

MATERIALS AND METHODS

Thirty-eight adult patients with supratentorial brain tumors were divided into frontal (N=19) and non-frontal (N=19) tumor subgroups for comparison based on their localization. They underwent

лизацията им: фронтални (N=19) и не-фронтални (N=19). Проведено е трикратно изследване за период от 7 месеца чрез прилагането на обширен набор от стандартизирани невропсихологични тестове.

РЕЗУЛТАТИ

Подгрупата на фронталните мозъчни тумори показва значително по-слаби резултати при изходното изследване по отношение на семантичната памет ($p<0.05$), селективното внимание, способността за екзекутивен контрол ($p<0.01$) и екзекутивните функции ($p<0.05$) в сравнение с пациентите от подгрупата на не-фронталните мозъчни тумори. Локализацията е фактор, който влияе върху резултатите получени от теста за категорийна ($F=4.88$; $p=0.03$) и фонемна флуентност ($F=5.82$; $p=0.02$), и от теста на Струп ($F=11.20$; $p=0.002$) в началото на периода на проследяване.

ЗАКЛЮЧЕНИЕ

Челните дялове са тясно свързани с определени аспекти на вниманието и екзекутивните функции. Няколко сравнително кратки когнитивни теста (теста на Струп, тестовете за категорийна и фонемна флуентност) могат да се прилагат като чувствителни невропсихологични средства за регистрация на когнитивни нарушения при пациенти, страдащи от тумори на челните дялове.
Ключови думи: мозъчен тумор, когниция, челни дялове

three point assessment for a period of 7 months using an extensive battery of standardized neuropsychological tests.

RESULTS

The frontal tumor subgroup showed significantly worse baseline results in semantic memory ($p<0.05$), selective attention, executive control ($p<0.01$) and executive functions ($p<0.05$) compared to the non-frontal tumor subgroup. ANOVA showed that the localization is a factor which influences the results from the categoric ($F=4.88$; $p=0.03$), phonemic ($F=5.82$; $p=0.02$) fluency tests and the Stroop test ($F=11.20$; $p=0.002$) at baseline.

CONCLUSION

Frontal lobes are closely connected with certain aspects of attention and executive functions. Several relatively brief cognitive tests (Stroop Color Word Test and verbal fluency) could be applied as sensitive tools for registration of cognitive dysfunction in patients with frontal lobe tumors.
Key words: *brain tumor, cognition, frontal lobes*

INTRODUCTION

Cognitive functions are regarded to be determined by either more localized or distributed substrate in the central nervous system [16,17]. The neuropsychological approach examines cognitive processes as a result of elaborately functioning neuronal networks. This idea was shared a few decades ago in the Luria's theoretical model of the higher psychic functions which are regarded as compound hierarchical self-regulating functional systems. Nowadays, this conception has been confirmed by means of functional neuroimaging investigations used to examine the anatomical substrate responsible for the execution of the versatile cognitive tasks. The execution of the different cognitive functions is based on the collaboration of multiple brain structures acting at different levels as well as a large number of afferent and efferent cortico-subcortical circuits. Meanwhile, the neurons from a certain association area take part in functional networks which link remote neuronal populations. The higher cortical functions cannot be strictly correlated to a certain anatomical substrate due to their complex structural and functional organization. It is not always possible to link a definite cognitive function to a specific brain region. The established

neuronal correlations are related to highly differentiated components of a given cognitive function [14,18].

The approach of executive functions began with the early description of behavioral disorders induced by frontal damage. The development of neuropsychology has led to the description of a large variety of cognitive impairments specific to frontal lobe pathology [8].

The aim of this study was to prospectively investigate specific cognitive impairments depending on the brain tumor localization.

MATERIALS AND METHODS

Materials: Thirty-eight adult patients (16 male and 22 female) with newly diagnosed supratentorial brain tumors enrolled in the study who were admitted for operative treatment to the Clinic of neurosurgery at the St George University Hospital of Plovdiv between 2010 and 2012. Patients were selected after meeting the predetermined inclusion and exclusion criteria and signing an informed consent. Mean age was 55.08±1.67 (SD 10.27); mean years of education were 10.82±0.41 (SD 2.51). In order to achieve the purpose of this study the patients were divided into frontal (N=19) and non-frontal (N=19) tumor subgroups for comparison according to tumor localization. The exact localization of the lesions was determined preoperatively by neuroimaging investigations – computed tomography scanning and/or magnetic resonance tomography with contrast enhancement. The frontal tumor subgroup included 7 cases of left frontal lobe tumors, 8 cases of right frontal lobe tumors and 4 cases of bifrontal tumors. The non-frontal tumor subgroup included as follows: 1 case of left temporo-medial and parasellar tumor; 1 case of the left temporo-parietal tumor; 1 case of the left temporal tumor; 1 case of the left parieto-occipital tumor; 1 case of the left parietal tumor; 5 cases of right parietal tumors; 2 cases of right parieto-occipital tumors; 6 cases of right temporal tumors; 1 case of right temporo-parietal tumor. According to their histology based on the Classification of Central Nervous System Tumors of the WHO from 2007, the study included 12 cases of Meningioma Gr.I and 3 cases of Meningioma Gr. II&III; 1 case of epidermoid tumor (cholesteatoma); 4 cases of low-grade glioma (Gr. II); 13 cases of high-grade glioma (Gr. III&IV); and 5 cases of metastatic brain tumors.

Methods: Patients were followed up for a period of 7 months. They underwent a three point assessment of their cognitive status: baseline (before surgery), at 1st and 7th postoperative month. Cognitive functions were evaluated by means of extensive battery of widely used and standardized neuropsychological tests which included: The Mini Mental State Examination as a screening tool assessing global cognitive functioning; The Bulgarian version of the Stroop Color Word Test measuring selective attention and the capability of executive control [15]; Trail-making test part A and B – visual scanning, information processing speed, motor planning and attention/executive functions (divided attention and shifting); Go/no-go test – decision making and executive motor control; Verbal (categoric and phonemic) fluency – semantic memory, executive functions; Digit Span test forward – short-term memory, and backward – working memory; Digit-Symbol test – visual scanning, mental flexibility, sustained attention, psychomotor and information processing speed; Clock drawing test – planning within the executive functions.

Statistical analyses: Descriptive, parametric, nonparametric, correlation and ANOVA statistical analyses were made by SPSS software (version 17.0). Graphic analyses of data were performed using MS Office Excel 2003. Scores were expressed as mean (±SE). Kolmogorov-Smirnov Test was used to test for normality of distribution. Most of the scores were non-normally distributed and were, therefore, compared by nonparametric methods. The numerical comparisons between consecutive measurements (dependent groups) were assessed by Friedman within the whole group comparisons

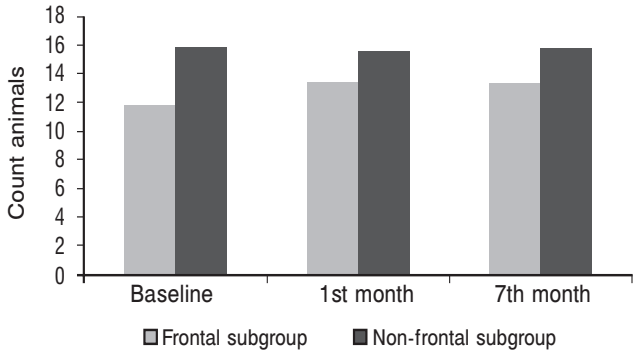
and by Wilcoxon test in pairwise comparisons. The pairwise comparisons between two independent groups were made by Mann–Whitney U test. A $p<0.05$ was considered to be statistically significant.

RESULTS

The frontal tumor subgroup demonstrated significantly worse semantic memory assessed by the categoric fluency test ($p<0.05$), poorer selective attention and executive control evaluated by the third part of the Stroop Color and Word Test ($p<0.01$) and executive functions examined by the phonemic fluency test ($p<0.05$) at baseline. ANOVA showed that the localization is a factor which influences the results from the categoric ($F=4.88$; $p=0.03$) and phonemic ($F=5.82$; $p=0.02$) fluency tests, and the Stroop test ($F=11.20$; $p=0.002$) at baseline.

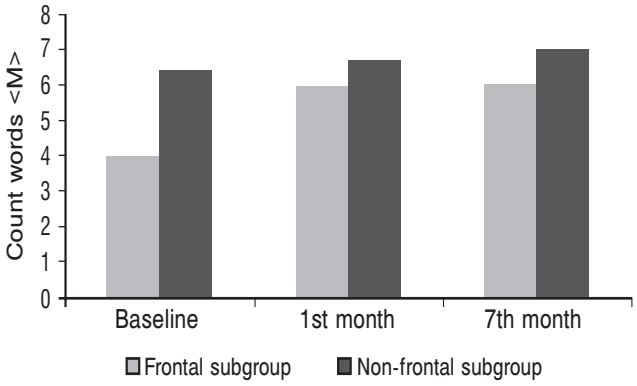
The frontal tumor subgroup experienced non-significant overall improvement of semantic memory assessed by the categoric fluency test of 15% compared to only 3% in the non-frontal subgroup for the entire period of longitudinal assessments. Nevertheless, patients with frontal tumors scored lower results at each assessment (Fig.1).

Fig.1. Dynamics of semantic memory assessed by the categoric fluency test.



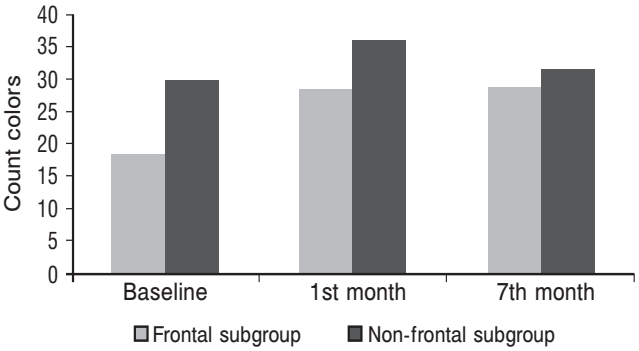
The patients with frontal tumors demonstrated significant ($p<0.05$) alleviation of their executive functions evaluated by the phonemic fluency test with nearly 62% as opposed to only 24% in the non-frontal subgroup. These results are confirmed by the presence of a moderate correlation between the affiliation of the cases to the frontal and non-frontal subgroups and the dynamics of the results from the phonemic fluency test ($r_s=-0.38$; $p<0.05$). Here as well, patients with frontal tumors scored lower results at each assessment (Fig.2).

Fig.2. Dynamics of executive functions assessed by the phonemic fluency test.



As to the Stroop Test, the frontal subgroup showed significant and permanent improvement of selective attention and executive control for the entire period with 83% ($p<0.01$), whereas the non-frontal subgroup experienced non-significant overall improvement of only 6%. These results are confirmed by the presence of a moderate correlation between the affiliation of the cases to the frontal and non-frontal subgroups and the dynamics of the results from the third part of the Stroop Test ($r_s=-0.45$; $p<0.05$). Again, patients with frontal tumors scored lower results at each assessment (Fig.3).

Fig.3. Dynamics of selective attention and executive control assessed by the Stroop Test.



The comparison of the results from the other neuropsychological tests used in this battery did not show statistically significant differences between the frontal and non-frontal subgroups.

DISCUSSION

The frontal lobes primarily support higher-level cognitive processes, comprising executive skills and working memory [6]. Executive functions include vital cognitive activities including decision-making, planning, executive control, sustained attention, awareness and insight. Not surprisingly, processing deficiencies can have far ranging effects that impact on educational attainment, employment and social functioning. For instance, executive skills deficits have been associated with poor outcomes on cognitive rehabilitation programmes [9]. The frontal lobe function integrates reasoning, learning, and creative abilities in the service of executive control and decision-making [2]. Furthermore, frontopolar regions are engaged in exploratory behavior long-term memory cued retrieval, and in the early phase of learning new behaviors [3,5].

Our study demonstrates that frontal tumor patients suffer from more severe deficits in semantic memory, selective attention, executive control and executive functions than non-frontal cases but, in the same time, surgical removal of the frontal tumors creates favorable conditions for recovery of those deficits. These impairments can be successfully detected and followed up by sensitive neuropsychological tests such as the verbal (categoric and phonemic) fluency test and the Stroop Test.

J. Alvarez и E. Emory (2006) point out that verbal fluency does not depend only on the semantic memory and vocabulary of the individual but also on his/her executive functions because memory would be impaired if the individual cannot focus and sustain attention to the task and develop an effective strategy for retrieval of words from the available linguistic vocabulary as well as to exert self-control on the execution of the task such as to avoid repetitions [1]. Therefore, we consider that the semantic memory domain in our frontal subgroup is secondarily affected as a result of the impairment of attention and executive functions which reflects the inability to develop an effective strategy for word retrieval and generation.

We confirm the findings from other studies which indicate that the verbal (categoric and phonemic) fluency tasks and the Stroop Test are sensitive neuropsychological tools for assessment of frontal lobe dysfunction [1,7,11–13].

However, neuroimaging studies demonstrate that the cognitive function cannot be linked to one and only anatomical region. Moreover, the frontal lobe regions have multiple connections with various other cortical, subcortical, and brain stem sites and, thus, the frontal lobes should "be conceived as one aspect of an executive system involving many structures of the central nervous system" [4]. For example, Alvarez and Emory (2006) share that the phonemic fluency performance is associated with increased activation not only of certain frontal regions such as the left dorsolateral prefrontal cortex, anterior cingulate and left inferior frontal gyrus but it also activates a number of non-frontal brain areas, including the thalamus, parietal lobes and temporal lobes [1]. In addition, Heflin et al. (2011) noticed that apart from dorsolateral prefrontal cortical atrophy, the bilateral parietal cortical atrophy was also associated with poorer scores on the Stroop Test [10].

CONCLUSION

The current study suggests that the verbal fluency tests and the Stroop Test are sensitive tools for assessment of frontal lobe attention and executive dysfunction. In addition, the specificity of these tests should be addressed in future longitudinal neuropsychological and functional neuroimaging studies assessing executive skills and memory before and after brain tumor resections which will hold promise in elucidating the nature and mechanisms underlying frontal lobe dysfunction.

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The impact of tumor malignancy on cognitive functioning and quality of life in adult patients with supratentorial brain tumors

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Влияние на малигнеността върху когнитивното функциониране и качеството на живот при пациенти със супратенториални мозъчни тумори

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РЕЗЮМЕ

Значимостта на когнитивното функциониране и качеството на живот при пациентите страдащи от мозъчни тумори е оценена едва през последните години.

ЦЕЛ

Да се извърши проспективно проследяване на влиянието на малигнеността на мозъчните тумори върху когнитивния статус и качеството на

ABSTRACT

The importance of cognitive functioning and quality of life in brain tumor patients has only recently been recognized.

AIM

To prospectively study the impact of tumor malignancy on cognitive status and quality of life in patients with supratentorial brain tumors.

живот при пациентите със супратенториални мозъчни неоплазми.

МАТЕРИАЛИ И МЕТОДИ

В проучването са включени общо 38 пациента със супратенториални мозъчни тумори, които са разпределени в две подгрупи в зависимост от хистологията им: злокачествени (N=25) и доброкачествени (N=13). Проведено е трикратно изследване за период от 7 месеца чрез прилагането на набор от широко използвани и стандартизирани невропсихологични тестове и скали за оценка на качеството на живот {Скала на Карнофски (KPS) и въпросник за оценка на качеството на живот версия 3.0 на Европейската Организация за Проучване и Лечение на Рака (EORTC QLQ-C30)}.

РЕЗУЛТАТИ

Подгрупата на малигнените мозъчни тумори показва значително по-слаба краткосрочна памет на седми следоперативен месец ($p<0.05$), по-лош функционален статус ($p<0.05$) в началото и края на проследяването, както и по-лоша самооценка на общото качество на живот ($p<0.01$) и ролевата дееспособност ($p<0.05$) измерени чрез QLQ-C30 в края на проследяването.

ЗАКЛЮЧЕНИЕ

Малигнените мозъчни тумори са свързани със значимо по-голямо нарушение на краткосрочната памет и качеството на живот спрямо тези с доброкачествени тумори. Комбинацията от невропсихологичното изследване и измерването на качеството на живот е важно средство за оценка на изхода от проведеното лечение при пациентите с мозъчни неоплазми.
Ключови думи: *мозъчен тумор, качество на живот,когнитивен дефицит, малигненост*

MATERIALS AND METHODS

A total of 38 patients with supratentorial brain tumors were divided into malignant (N=25) and benign (N=13) tumor subgroups for comparison according to their histology. They underwent threefold assessment for a period of 7 months using a battery of widely used and standardized neuropsychological tests and quality of life scales {Karnofsky Performance Status Scale (KPS) and Quality of Life Questionnaire-C30, version 3.0 of the European Organization for Research and Treatment of Cancer (EORTC QLQ-C30)}.

RESULTS

The malignant tumor subgroup showed significantly worse short-term memory at the 7th postoperative month ($p<0.05$), lower KPS scores at baseline and 7th postoperative month ($p<0.05$) and demonstrated worse global QOL ($p<0.01$) and role functioning ($p<0.05$) measured by QLQ-C30 at the end of the study compared to the benign tumor subgroup.

CONCLUSION

Malignant brain tumors are associated with greater impairment of short-term memory and quality of life than benign tumors. The combination of neuropsychological and QOL assessment is an important outcome measure in the treatment of brain tumor patients.
Key words: Brain tumor, quality of life, cognitive deficit, malignancy

INTRODUCTION

The incidence of brain tumors varies between 5% and 10% of all neoplasms [17]. It is well recognized that impairment of neurocognitive functioning, resulting in behavioral, emotional, and intellectual difficulties, occurs in nearly all patients with brain tumors and eventually compromises their independence. This impairment is related to a combination of various factors, including the tumor itself, tumor-related treatment, and patient-related factors [8]. Neurocognitive function is an important determinant of quality of life (QOL) [9]. One of the most important and widely used tools for assessment of QOL in brain tumor patients are the KPS and QLQ-C30 of the EORTC [1,3].

The purpose of this study was to prospectively study the effect of tumor malignancy on cognitive status and QOL in adult patients with supratentorial brain tumors.

MATERIALS AND METHODS

Materials: The study included a total of 38 adult patients (16 male and 22 female) with newly diagnosed supratentorial brain tumors who were admitted for operative treatment to the Clinic of neurosurgery at the St George University Hospital of Plovdiv between 2010 and 2012. Patients were selected after meeting the predetermined inclusion and exclusion criteria and signing an informed consent. Mean age was 55.08±1.67 (SD 10.27); mean years of education were 10.82±0.41 (SD 2.51). In order to achieve the final goal of this study the patients were divided into malignant (N=25) and benign (N=13) tumor subgroups for comparison according to their histology based on the Classification of Central Nervous System Tumors of the WHO from 2007. The benign tumor subgroup included 12 cases of Meningioma Gr.I and 1 case of epidermoid tumor (cholesteatoma). The malignant tumor subgroup included 4 cases of low-grade glioma (Gr. II), 13 cases of high-grade glioma (Gr. III&IV), 3 cases of Meningioma Gr. II&III and 5 cases of metastatic brain tumors. All of the patients from the malignant tumor subgroup were subject to adjunctive therapy (radiotherapy and/or chemotherapy) in addition to the surgical resection of the lesion.

Methods: Patients were followed up for a period of 7 months. They underwent a threefold assessment of their cognitive status and QOL: baseline (before surgery), at 1st and 7th postoperative month. Cognitive functions were evaluated by means of an extensive battery of widely used and standardized neuropsychological tests which included: The Mini Mental State Examination as a screening tool assessing global cognitive functioning; The Bulgarian version of the Stroop Color Word Test measuring selective attention and the capability of executive control [18]; Trail-making test part A and B – visual scanning, information processing speed, motor planning and attention/executive functions (divided attention and shifting); Go/no-go test – decision making and executive motor control; Verbal (categoric and phonemic) fluency – semantic memory, executive functions; Digit Span test forward – short-term memory, and backward – working memory; Digit-Symbol test – visual scanning, mental flexibility, sustained attention, psychomotor and information processing speed; Clock drawing test – planning within executive functions.

Patients' functional status was assessed by the KPS score (varying 0–100). Health-related QOL was evaluated by means of the QLQ-C30 version 3.0 of the EORTC. The latter is a 30-item questionnaire composed of multi-item scales and single items that reflect the multidimensionality of the QOL construct. It includes five functional scales (physical, role, cognitive, emotional, and social), three symptom scales (fatigue, pain, and nausea/vomiting), and a global health status/QOL scale. The remaining single items assess additional symptoms commonly reported by cancer patients (dyspnea, appetite loss, sleep disturbance, constipation, and diarrhea), as well as the perceived financial impact of the disease and treatment.

Statistical analyses: Descriptive, parametric, nonparametric, correlation and ANOVA sta-

tistical analyses were made by SPSS software (version 17.0). Graphic analyses of data were performed using MS Office Excel 2003. Scores were expressed as mean (±SE). Kolmogorov-Smirnov Test was used to test for normality of distribution. Most of the scores were non-normally distributed and were, therefore, compared by nonparametric methods. The numerical comparisons between consecutive measurements (dependent groups) were assessed by Friedman within the whole group comparisons and by Wilcoxon test in pairwise comparisons. The pairwise comparisons between two independent groups were made by Mann–Whitney U test. A $p<0.05$ was considered to be statistically significant.

RESULTS

From all cognitive domains studied, the only one which was statistically different between the two subgroups was the short-term memory domain assessed by the Digit Span test forward. The patients from the malignant brain tumor subgroup showed significantly worse results on short-term memory test at the 7th postoperative month ($p<0.05$) compared to the patients suffering from benign tumors. The malignant tumor subgroup exhibited a slight postoperative improvement of this domain at the first postoperative month which was only temporary and followed by an overall drop for the entire period of 4%. By contrast, the benign tumor subgroup had a temporary drop of short-term memory at the 1st month after surgery but they recovered and experienced insignificant overall improvement of 6% at 7th postoperative month compared to baseline results (Fig. 1).

The malignant tumor subgroup also demonstrated significantly lower functional status (KPS) at baseline and 7th postoperative month ($p<0.05$), poorer global QOL ($p<0.01$) and role functioning ($p<0.05$) at 7th postoperative month. Some symptoms such as pain, nausea and vomiting were also more prominent in the malignant tumor subgroup at the 1st postoperative month ($p<0.05$).

Both subgroups demonstrated significant postoperative improvement in functional status and global QOL that were stable for the entire follow-up period but it was more prominent in the benign tumor subgroup – overall improvement of KPS was 15% and that of global QOL – 78%, whereas in the malignant tumor subgroup these per cents were 10% and 25% respectively (Fig.2 and 3).

The benign tumor subgroup demonstrated significant and stable postoperative improvement of role functioning for the entire period reaching 44% ($p<0.05$), whereas in the malignant tumor subgroup the improvement was only temporary at the 1st postoperative month ($p<0.05$) without reaching statistical significance for the entire period (18%) compared to baseline results.

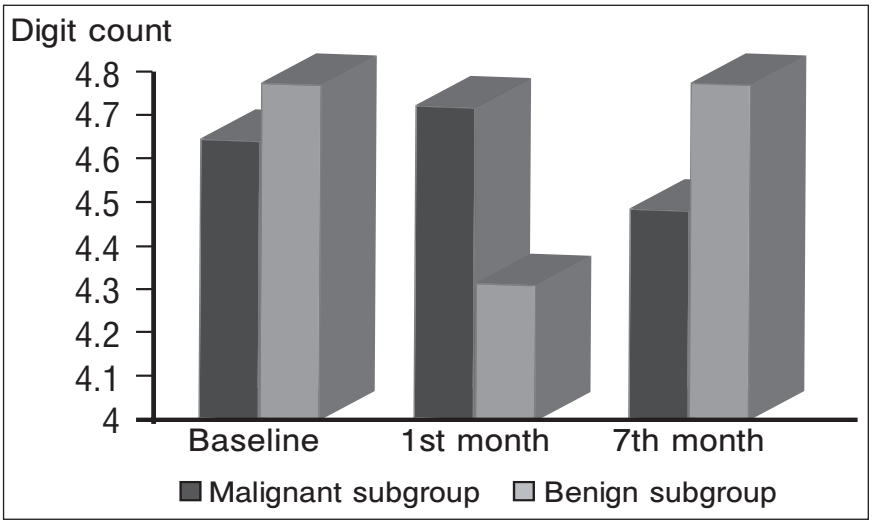

Fig.1.
Dynamics of the short-term memory domain assessed by the Digit Span Forward Test.

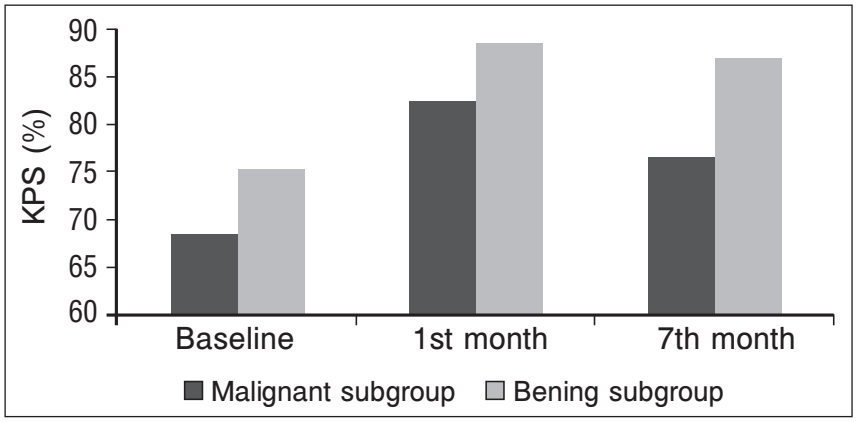

Fig.2. Dynamics of the functional status assessed by KPS.

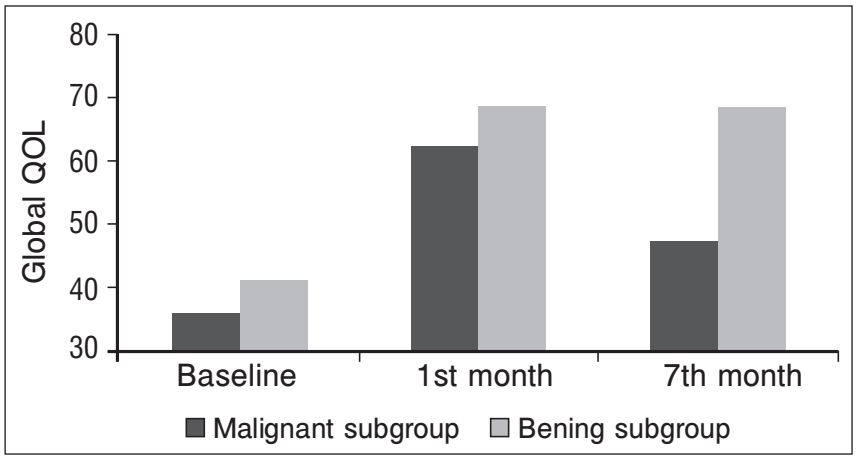

Fig.3.
Dynamics of the global QOL assessed by QLQ-C30.

ANOVA analysis showed that tumor malignancy is a factor which influences the functional status (KPS) at baseline (F=8.08;p=0.007) and at 7th postoperative month (F=5.70;p=0.02), and global QOL assessed by QLQ-C30 at the end of the follow-up period (F=11.34;p=0.002).

DISCUSSION

Tucha et al. (2000) share that memory is one of the most commonly affected cognitive domains which presents in as much as 60% of patients suffering from brain neoplasms [15]. The impact of tumor histology on cognitive functioning has been a matter of debate. Some publications point out that tumor malignancy is an important factor playing a role in the development of cognitive impairments, while others disagree and share that tumor lateralization really matters [10,11]. Nonetheless, Hahn et al. (2003) emphasize that cognitive impairment is more severe in rapidly-growing malignant tumors as opposed to slow-growing ones [7]. The current study shows that short-term memory is more prominently and permanently impaired in patients with malignant brain tumors despite of the fact that surgical treatment can lead to temporary postoperative improvement in this subgroup. One of the reasonable explanations of this phenomenon may be the influence of adjuvant therapies such as radiotherapy and/or chemotherapy which are usually started in virtually all cases of malignant brain tumors after surgery. Some authors share that radiation-induced cognitive impairment may affect 50%-90% of patients 6 months post-irradiation and one of the most prominent deficits is found in the memory domain [6]. In addition, Warrington et al. (2012) point out that memory impairment, considered to be irreversible, can alleviate over time in long-term survivals (more than 3 years) with malignant brain tumors [16].

The benign tumor subgroup demonstrated a temporary postoperative drop of short-term

memory at 1st postoperative month which was followed by a period of recovery and showed overall improvement of 6% at the 7th postoperative month compared to baseline results. The temporary worsening 1 month after operative treatment may be due to surgical side-effects. Our observation is confirmed by other publications which share, that due to brain plasticity, patients can experience long-term improvements of cognitive deficits usually after transient postoperative decline [5,13]. Cerebral plasticity is considered to be a continuous process allowing short-term, middle-term and long-term remodeling of neuron-synaptic maps, to optimize the functioning of brain networks [4,12].

Health-related quality of life (HRQOL) has become an important outcome measure in brain tumor clinical trials [14]. Our study demonstrates that surgery and adjuvant therapy leads to permanent improvement of functional status (KPS) and global QOL in both the benign and malignant subgroups. Nevertheless, the malignant brain tumor subgroup showed significantly poorer levels of functional status, global QOL and role functioning compared to the patients with benign lesions. Baseline results were worse for both groups as opposed to postoperative ones. This observation was shared by Budrukkar et al. (2009) who implicate the idea that brain tumor itself and the resulting cognitive deficit are the major factors influencing the patients' QOL [2]. Some authors share the opinion that surgery may initially, at least temporarily, improve QOL dramatically in a significant proportion of patients with severe symptoms related to increased intracranial pressure. Conversely, radiotherapy may decrease QOL in some patients from adverse effects such as hair loss, fatigue, somnolence, or cognitive problems [8].

Actually, our findings confirm the abovementioned statement because the malignant tumor subgroup showed considerable postoperative improvement at 1st month due to surgery followed by a period of worsening between the 1st and 7th postoperative month which can be explained by the impact of the adjuvant therapies side-effects on patients' QOL. A piece of evidence confirming that is the fact that the benign tumor subgroup, which was not exposed do adjunctive therapies, demonstrated significantly better and more stable improvement in terms of their functional status, global QOL and role functioning.

CONCLUSION

The current study demonstrates that tumor malignancy have certain impact on neurocognitive functioning and QOL of adult patients suffering from supratentorial brain tumors. Patients with malignant brain tumors are prone to exhibit a more pronounced impairment of short-term memory. They also have poorer functional status and QOL which tends to worsen over time probably owing to the devastating nature of their disease and the side-effects of the adjuvant therapies such as radiotherapy and/or chemotherapy that they received.

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Current investigation approaches for quality of life assessment in patients with rare diseases

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Съвременни подходи за изследване качеството на живот при пациенти с редки болести

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РЕЗЮМЕ

Изследванията на Качеството на живот, свързано със здравето биха могли да се използват за определяне на състоянието на пациентите, тяхното благополучие и възприемането на собственото им здраве във всяка една от следните 3 области: физическа, психическа и социална. Целта на статията е да се направи обзор и анализ на използването на въпросници за оценка на качеството на живот на пациенти с редки болести, обсъждайки приложенията на стандартизирани общи и специфични въпросници. Дискутира се необходимостта и значението от разработването на специфични въпросници за оценка на качеството на живот и ролята им за стимулиране броя и качеството на изследванията на пациентите с реки болести, в часност Таласемия и значението на инструмента за клиничната практика. Чрез обединените усилия на медицински специалисти, психолози, пациентски организации и хора ангажирани с проблема

ABSTRACT

Quality of life (QoL) assessment, related to health issues, could be useful when determining a patient's condition, well-being and self-perception of own health in the context of physical, mental an social functioning. Aim of this article is to review and analyze current practices in the implementation of QoL instruments for the successful management of patients with rare diseases. Different standardized generic and disease-specific QoL instruments are described. Discussion evolves on the unmet need for developing and validating disease-specific instruments for QoL assessment in thalassemia and how this would improve research on patient-reported outcomes in rare diseases. We propose a new validated disease-specific instrument and its potential use for QoL assessment in thalassemia.

Key words: Quality of life, rare diseases, thalassemia, generic and disease-specific questionnaires.

ние предлагаме валидизиран въпросник за изследване на качеството на живот на пациенти с таласемия.

Ключови думи: качество на живот, редки болести, таласемия, общи и специфични въпросници

Quality of life (QoL) can be defined as an integral reflection of a patient's wellbeing in the context of an illness, social functioning, daily activities and family life based on the patient's overall perception. It is an important measure for purposefulness, substantiveness, value and fulfillness of life. This concept is strongly applicable for patients with chronic diseases. Defining a robust methodology for the analysis of QoL assessment and improving outcomes is of great importance for current medical sciences. QoL questionnaires impose increasingly as an indispensable measure for evaluating patient's satisfaction and the quality of healthcare services [6].

Aim of this paper is to review and analyze information on the use of generic and specific QoL questionnaires in patients with rare diseases, as well as to present a new thalassemia-specific QoL questionnaire.

1. QOL ASSESSMENT WITH GENERIC AND DISEASE-SPECIFIC INSTRUMENTS

1.1 Generic instruments

For QoL assessment two types of instruments can be used – "generic" and "disease-specific". QOL instruments applied into practice need to demonstrate good psychometric properties. More than 250 types of instruments have been developed. Pre-determined health concepts are measured by rating on scales answers to each question.

Several QoL questionnaires and rating scales are known such as SF-36 (Health Survey), World Health Organization QoL instrument (WHOQOL), EQ-5D, (HQ) Harvard Pilgrim Health Care. An appropriate example for a generic questionnaire, applicable worldwide both for chronically ill patients and healthy individuals is the SF-36 Health Survey and its short version SF-12, developed at the RAND Corporation in the USA.

Good instruments yield results not only for general health indices but also for profiles which assess different components of QoL. For example the instrument SF-36 Health Survey measures eight different health concepts of QoL: physical functioning, role limitations due to physical health problems, bodily pain, general pain, vitality, social functioning, role limitation due to emotional problems, mental health [29]. Main disadvantage of generic instruments is their inability to yield the benefits from already implemented health programs thus limiting the identification of possible measures for improving patient's satisfaction.

1.2 Disease-specific instruments

Implementation of disease-specific instruments in certain patient populations has proven to be beneficial in terms of effective patient management. Therefore it is of crucial importance to widely use these types of instruments in medical practice. Aim of this article is to present the composition and implementation of thalassemia-specific QoL instrument.

"Rare diseases" is an umbrella term, including many nosological entities. However, patients with a certain diagnosis are few. This restricts gaining awareness on QoL in patients with rare diseases and dampens scientific research on the topic as well. Disease-specific instruments can evaluate treatment-related QoL changes in

a patient's life on the basis of repeated measures [26]. This type of instruments enables all involved specialists in the decision-making process (attending physicians, social workers, psychologists) to take relevant actions on improving the patient's well-being. The most widely used disease-specific QOL instruments in medical practice are presented in Tables 1 and 2:

Table 1.
Most commonly used disease-specific QoL instruments in general medical practice.

Title	Abbreviation
Child Health Questionnaire [10]	(CHQ)
Child Health and Illness Profile-Adolescent Edition [10]	(CHIP-AE)
Osteoporosis Quality of Life Questionnaire[17]	(Qualeffo-41)
Kidney Disease Quality of Life [18]	(KDQOL)
Duke Health Profile [3]	(DUKE)
London Handicap Scale [1]	(LHS)
Quality of Well-Being Scale [21]	(QWB)
Primary Care Assessment Survey [33]	(PCAS)
Adult Asthma Quality of Life Questionnaire [12]	(AQLQ)
Pediatric Asthma Quality of Life Questionnaires [7]	(PAQLQ)
MOS-HIV Health Survey [14]	(MOS-HIV)
Migraine Specific Quality of Life [8]	(MSQOL)
Migraine Specific Quality of Life Questionnaire [9]	(MSQ v.2.1)
24-Hour Migraine Quality of Life Questionnaire [13]	(24-Hr-MQOLQ)
St. George's Respiratory Questionnaire – UK Parent Version [20]	(SGRQ-UK)
Seattle Angina Questionnaire [24]	(SAQ)
Urinary Incontinence-Specific Quality of Life Questionnaire [32]	(I-QOL-US)

Table 2.
QoL instruments specific for rare diseases

Title	Abbreviation
Idiopathic thrombocytopenic purpura Quality of Life Questionnaire for children [36]	(ITP-QoL)
Haemophilia Quality of life questionnaire [5,4]	(Haemo-QoL)
Acromegaly Quality of Life questionnaire [37]	(AcroQoL)
Atopic Dermatitis Quality of Life questionnaire [39]	(QoLIAD)
Cystic fibrosis Quality of Life questionnaire [30]	(CFQ)
Hypercortisolism Quality of Life questionnaire [38]	(Cushing QoL)

Table 1 lists several of the most commonly used disease-specific QOL instruments. They have been developed for a particular disease condition. Furthermore, they must be translated and adapted for use in particular country.

Table 2 includes disease-specific QoL instruments for patients with rare diseases. Due to the small number of patients and their irregular distribution worldwide only few QoL instruments have been developed for small part of the rare diseases.

1.3 QoL ASSESSMENT IN PATIENTS WITH RARE DISEASES

Most of the research on QoL in patients with rare diseases has been conducted using generic instruments or in combination with disease-specific ones, which are listed in Table 3.

Table 3.
Generic and disease-specific instruments used for QOL assessment in patients with rare diseases

Disease/Condition	Applied instrument
Takayasu's arteritis (*Pulseless disease*) [11]	**SF-36** generic instrument
Hereditary hemorrhagic telangiectasia (HHT) (Rendu-Osler-Weber disease) [28]	**SF-36** generic instrument
Sickle Cell Anemia [25]	**SF-36** generic instrument
Fabry-Anderson disease [27]	**EQ-5D** generic instrument
Neurofibromatosis type 1 [23]	**SF-36** generic instrument **Skindex-29** disease-specific instrument
Duchenne muscular dystrophy [19,22,2]	**WHOQOL-100** generic instrument **BDI** disease-specific instrument **TACQoL** disease-specific instrument (chronic illnesses) (8–15 years) **TACQoL-CF** (15–17 years) **TAAQoL** (adults)
Hypercortisolism (*Cushing's syndrome*)	**SF-36** generic instrument **Cushing QoL** disease-specific instrument
Acromegaly	**SF-36** generic instrument **AcroQoL** disease-specific instrument
Thalassemia [35,31,34]	**SF-36** generic instrument **EQ-5D** generic instrument **GHQ-12** generic instrument **WHOQOL-100** generic instrument
Atopic Dermatitis	**QoLIAD** disease-specific instrument
Idiopathic thrombocytopenic purpura	**ITP-QoL** disease-specific instrument
Haemophilia	**HAEMO-QoL** disease-specific instrument
Cystic fibrosis	**CFQ** disease-specific instrument

For example, studies of QOL in patients with hemophilia have increased in number only after the disease-specific instrument HAEMO-QoL has been developed. All studies conducted in different countries yielded similar results. Disease-specific instruments possess greater potential to capture not only the current condition but also changes related to lifestyle.

2. QOL INSTRUMENT IN PATIENTS WITH THALASSEMIA

The development of a new instrument is time-consuming, usually requires the time-span of a PhD research. Most researchers use already existing instruments and adapt them accordingly to the particular study. In that case they most probably would have to face the question: "How to choose the right instrument among all those, which seem to measure the same concept?" "Good instrument" can be constructed on the basis of a detailed literature review of the different instruments, known so far. Reliable results can be yielded only after the requirement is met for validation, translation and adaptation of the instrument for use in a particular country.

Taking into consideration the above-mentioned, we faced the challenge to develop own QoL instrument for patients with thalassemia. The presented instrument is disease-specific and the process of its development involved an interdisciplinary team of specialists. Our preliminary research indicates that up to this date no specific QoL instrument for thalassemia exists and even though being a rare disease, thalassemia is not uncommon in Bulgaria. Because of treatment specificity and its importance for the prospective disease evolution patients are extremely dependent on how their quality of life is rated. In other words, patients, who undergo treatment and are motivated to adhere to it, experience more favorable outcomes than those who are uninformed and unmotivated [16].

The Rationale behind developing the new instrument is based on:

1. Its ability to evaluate QoL components, which cannot be assessed with current instruments.
2. The new instrument allows improvement in research (it is simple, easily applicable and adaptable, sensitive to QoL changes in patient's condition) [15].

THE INSTRUMENT WAS DEVELOPED AT THREE STAGES:

First stage: Primary pilot study including 24 patients (10 males/14 females).

The instrument consisted of 22 questions in following domains: physical health, mental health and social wellbeing. Numeric scale was used to rate the patient's satisfaction.

RESULTS YIELDED FOLLOWING CONCLUSIONS:

(+)

- Good patient compliance;
- High correlation coefficient between test-retest (r_{xy}>0.90), i.e. good external validation;
- Good statistical values for internal validation (Cronbach coefficient > 0.7).

(–)

- Questions on physical health had to be reconstructed (due to different perception from females and males);
- Rating scale was be changed from numeric to ordinal;
- Second pilot study evaluating the corrected instrument.

Second stage: The second pilot study comprised of 20 patients (8 males/12 females). The instrument included 20 questions in the domains of physical health, mental health and social wellbeing. Patient satisfaction was rated on a five-item Likert scale:

YES	rather YES	neither YES, nor NO	rather NO	NO

In order to guarantee that patients have clearly understood the questions and thus maximizing the accuracy of our results, we have reformulated some questions after the first study.

RESULTS FROM THE SECOND STUDY YIELDED FOLLOWING CONCLUSIONS:

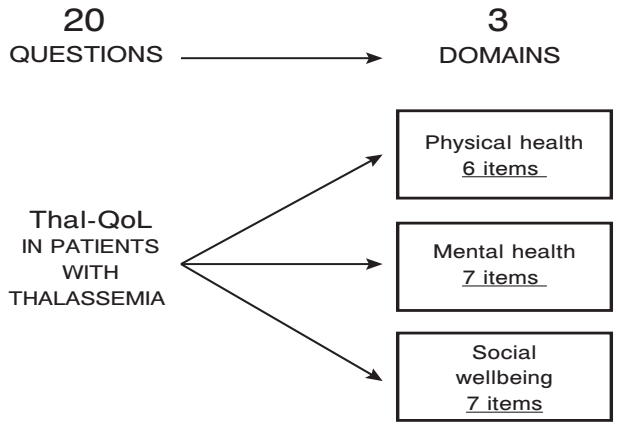
- High correlation coefficient between test-retest;
- Good internal validation;
- Significant QoL improvement, if patients strongly adhere to treatment;
- Good utility measure when this type of instruments is used to evaluate treatment and social care efficacy.

Third stage: The final version was developed after taking into consideration results from both pilot studies and the input from medical professionals, public health professionals, psychologists and patients from the Bulgarian Organization of Patients with Thalassemia.

In the process of developing our instrument we relied on the experience from already existing QoL instruments for rare diseases, chronic illnesses and conditions with negative impact on physical, emotional and social well-being. The instrument was fairly well accepted by patients. Information on their physical and mental condition was useful for evaluating treatment efficacy. Our team hopes for an increase in studies focusing on QoL assessment in thalassemia which could aid the policy making process of improving the QoL of the affected individuals.

Figure 1.
Questionnaire (instrument) for quality of life in patients with thalassemia – structure

Фигура 1. Въпросник (инструмент) за качество на живот при пациенти с талласемия



The instrument is anonymous. However, monitoring and subsequent follow up of patients are possible due to the integrated unique patient identification number. Our questionnaire comprises of 20 questions, relevant to three domains- physical health, mental health and social well-being, which gather information on:

The health condition of a patient – pain, fatigue and different types of discomfort

- Emotional well-being – subjective feelings, personal issues, acceptance of administered therapy.
- Social functioning – daily activities, abilities to study, employment, leisure time and social acquaintances.

This instrument is specific for Thalassemia and gives answers to questions that are particularly related to the disease and the resulting problems and conditions.

In conclusion, we can point out that the instrument (Thal-QoL) is developed following established procedures for creating parameters for QoL assessment – such that have been used for already existing instruments. Interest in assessing QoL among patients with rare diseases is increasing. Even more authors take into consideration the way patients experience their own health, psycho-emotional functioning and every aspect of the received treatment. Balancing between personal perception on one hand and external evaluation on the other, regarding as many as possible health concepts enables reliable measurement and objective analysis of QoL of these patients. QoL research studies are most successful in the context of international cooperation which is necessary for achieving meaningful and sustainable impact worldwide.

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В него се отпечатват оригинални научни статии, казуистични съобщения, обзори, рецензии и съобщения за проведени или предстоящи научни конгреси, симпозиуми и други материали в областа на клиничната и фундаменталната медицина. Списанието излиза на английски език с подробни резюмета на български и английски. Изключения се правят за обзорни статии по особено значими теми. Заглавията, авторските колективи, а също надписите и означенията на илюстрациите и в таблиците се отпечатват и на двата езика.

Материалите трябва да се предоставят в два еднакви екземпляра, напечатани на пишеща машина или на компютър, на хартия формат А4 (21 х 30 см), 60 знака на 30 реда при двоен интервал между редовете (стандартна машинописна страница). Освен това могат да бъдат изпратени като прикачени файлове по електронната поща на адресите, посочени по-долу.

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exceed one standard typewritten page of 200 words.

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Title page

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Examples:

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1. McLachan, S. , M. F. Prumel, B. Rapoport. Cell Mediated or Humoral Immunity in Graves' Ophthalmopathy? J. Clin. Endocrinol. Metab., 78, 1994, 5, 1070–1074.

Reference to a book chapter:

2. Delange, F. Endemic Cretenism. In: The Thyroid (Eds. L. Braveman and R. Utiger). Lippincott Co, Philadelphia, 1991, 942–955.

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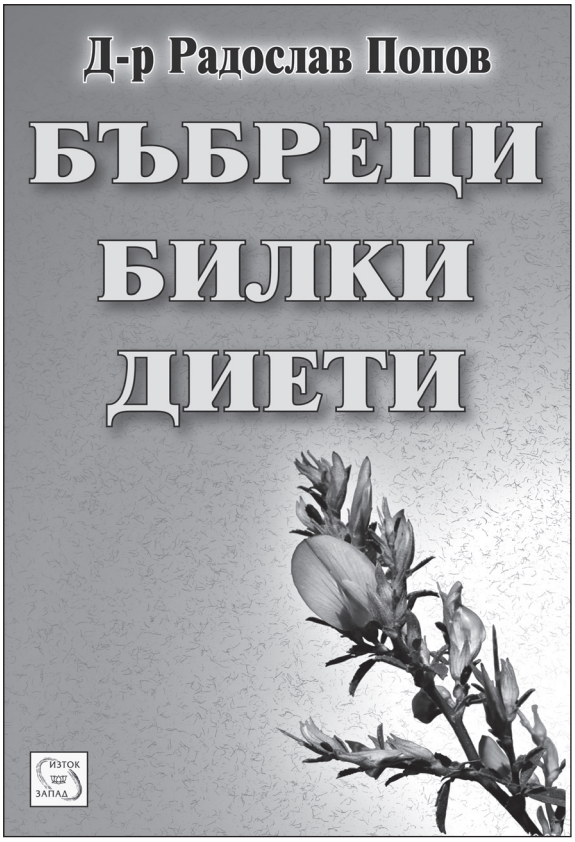
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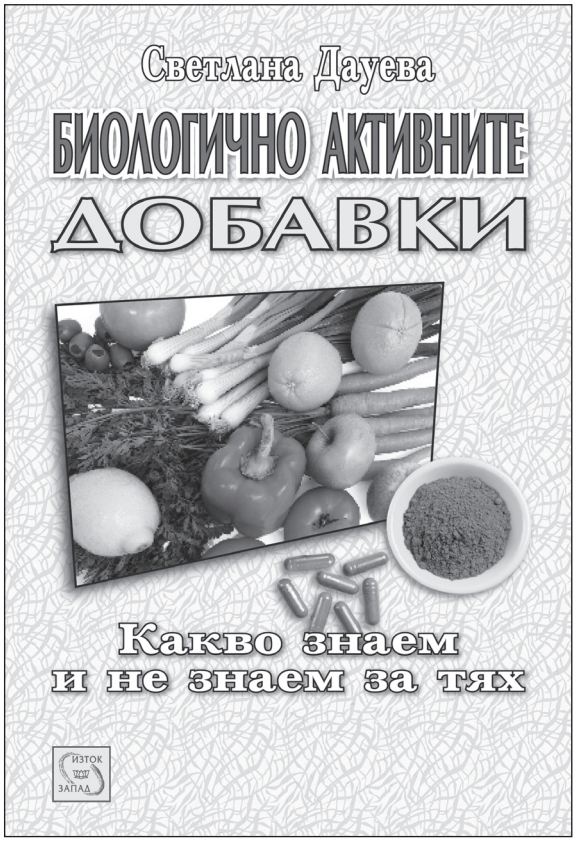
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