PHARMACIA

Pharmacia 70(4): 837–845
DOI 10.3897/pharmacia.70.e110353

Review Article

INNs granted with specific storage requirements in Bulgarian pharmacies. Part 2: Antineoplastic and immunomodulating agents

Evgeni Grigorov[1], Maya Radeva-Ilieva[2], Kaloyan D. Georgiev[2]

1 *Department of Organization and Economics of Pharmacy, Faculty of Pharmacy, Medical University, Varna, Bulgaria*
2 *Department of Pharmacology, Toxicology and Pharmacotherapy, Faculty of Pharmacy, Medical University, Varna, Bulgaria*

Corresponding author: Evgeni Grigorov;(evgeni.grigorov@mu-varna.bg)

Received 31 July 2023 ♦ **Accepted** 12 September 2023 ♦ **Published** 20 September 2023

Citation: Grigorov E, Radeva-Ilieva M, Georgiev KD (2023) INNs granted with specific storage requirements in Bulgarian pharmacies. Part 2: Antineoplastic and immunomodulating agents. Pharmacia 70(4): 837–845. https://doi.org/10.3897/pharmacia.70.e110353

Abstract

There are drugs that require special storage in Bulgarian pharmacies as well as extra caution during the dispensing process. This is due to serious adverse reactions that may be even fatal. These medicines are included in Appendix № 9 to Art. 17, para. 1 of Ordinance № 28/9.12.2008, issued by the Minister of Health. The performed study of the anticancer drugs listed in the Appendix showed that a major part of these medicines that have a marketing authorization for use in Bulgaria are not included in the Appendix № 9. In addition, there are antitumor drugs that are listed in the appendix but are not authorized in Bulgaria to date. In conclusion, it is necessary to periodically update the drugs in Appendix № 9 as well as to develop clear and precise criteria for the inclusion of medicines in it.

Keywords

Appendix № 9, drug, special storage, pharmacy, anticancer, immunosuppressants

Introduction

The main normative acts governing healthcare in the Republic of Bulgaria, the processes of prescribing and dispensing of medicines as well as their proper storage were discussed in part 1 of this article. In Appendix № 9 to Art. 17, para. 1 of Ordinance № 28/9.12.2008 are included medicines that must be stored in a separate locked cabinet. Thermolabile medicines included in the Appendix are stored in a refrigerator (Ministry of Health, Ordinance № 28).

About half of the drugs in the Appendix are antineoplastic agents. It is well known that anticancer drugs have various side effects such as nausea, vomiting, alopecia, myelosuppression, mucositis and many others. This is due to the fact chemotherapy is cytotoxic to either cancer and normal cells (Naeem et al. 2022). According to the Anatomical Therapeutic Chemical (ATC) classification system Antineoplastic agents are divided into following groups: Alkylating agents, Antimetabolites, Plant alkaloids and other natural products, Cytotoxic antibiotics and related substances, Protein kinase inhibitors, Monoclonal antibodies and antibody drug conjugates and Other antineoplastic agents (WHO 2023b). The two main strategies for cancer treatment with medicines are chemotherapy and targeted therapy (Debela et al. 2021; Naeem et al. 2022). Chemotherapy drugs exert their toxic effect on cell cycle phases and are divided into cell cycle-specific drugs and cell cycle-nonspecific drugs. Medicines from the class of alkylating agents, antimetabolites,

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plant alkaloids, cytotoxic antibiotics as well as other antineoplastic agents are used in chemotherapy (Dickens and Ahmed 2018; Naeem et al. 2022). The two main drug classes used in targeted cancer therapies are small molecule inhibitors and monoclonal antibodies. Most small molecule inhibitors act as protein kinase inhibitors (Liu et al. 2022). Small molecule inhibitors target specific molecular targets within cancer cells and this is how they differ from chemotherapy. These targets may be genetically modified in cancer cells leading to uncontrolled cell proliferation and survival (Naeem et al. 2022). Monoclonal antibodies selectively target cell surface antigens in tumor cells (Bayer 2019; Debela et al. 2021; Tsao et al. 2021). Targeted therapy has advantages over chemotherapy by inhibiting cancer cell growth while less harmful to healthy cells. However, adverse reactions are also reported (Debela et al. 2021).

In Appendix № 9 are included two immunosuppressive drugs. Immunosuppressants are used in solid organ transplantation to prevent rejection and in the treatment of autoimmune diseases. There are some classes of immunosuppressants that have different pharmacological action but all of them limit inflammation and suppress immune responses to various antigens. Similar to antineoplastic agents, these drugs have characteristic side effects (Meneghini et al. 2021; Neuberger 2021).

The large number of anticancer drugs listed in Appendix № 9 is probably due to the high toxicity of these medications. In this regard, it is of interest whether all antitumor drugs and immunosuppressants that are authorized in Bulgaria are included in the Appendix. The purpose of this study is to evaluate the specificity and particularity of the antineoplastic agents and immunosuppressants included in Appendix № 9 to Art. 17, para. 1 of Ordinance № 28/9.12.2008, issued by the Minister of Health.

Methods

For the purpose of the present study, it was performed a thorough literature review of the available official documentation as well as scientific databases about the drugs listed in Appendix № 9 of Ordinance № 28/9.12.2008, especially antineoplastic and immunomodulating agents. The data found were analyzed and summarized in order to assess the relevance of the Appendix and to clarify the reasons why specific storage conditions are required for these drugs.

Results and discussion

Appendix № 9 to Art. 17, para. 1 of Ordinance № 28/9.12.2008 includes a total of 70 medicines from different pharmacotherapeutic groups, according to the Anatomical Therapeutic Chemical (ATC) classification (Ministry of Health, Ordinance № 28). As it was mentioned in the first part of this article, medicines were divided into four groups – Drugs acting on cardiovascular system and blood coagulation, Drugs acting on peripheral and central nervous system, Anabolic steroids and Antineoplastic and immunomodulating agents. The first three groups were discussed in part 1 of the article. The medicinal products included in Appendix № 9, are shown in Table 1 by their international nonproprietary name (INN).

Table 1. INNs included in Appendix № 9 (Ministry of Health, Ordinance № 28).

Antineoplastic and immunomodulating agents		Medicines acting on peripheral and central nervous system	Medicines acting on cardiovascular system and blood coagulation	Anabolic steroids
Amsacrine	Idarubicin	Alcuronium	Acenocoumarol	Metandienone
Asparaginase	Ifosfamide	Ambenonium	Acetyldigoxin	Nandrolone
Azathioprine	Irinotecan	Atracurium	beta-Methyldigoxin	Oxymetholone
Bleomycin	Lomustine	Atropine	Digitoxin	
Busulfan	Melphalan	Biperiden	Digoxin	
Carmustine	Mercaptopurine	Butylscopolamine	Ethyl biscoumacetate	
Chlorambucil	Methotrexate	Ergotamine	Lanatoside C	
Ciclosporin	Mitobronitol	Galantamine		
Cisplatin	Mitolactol	Mevacurium chloride		
Cyclophosphamide	Mitomycin	Nalorphine		
Cytarabine	Mitoxantrone	Naloxone		
Dacarbazine	Paclitaxel	Neostigmine		
Daunorubicin	Procarbazine	Pancuronium		
Doxorubicin	Tegafur	Pilocarpine		
Epirubicin	Teniposide	Pipecuronium		
Estramustine	Tioguanine	bromide		
Etoposide	Vinblastine	Pyridostigmine		
Fluorouracil	Vincristine	Rocuronium		
Fotemustine	Vinorelbine	bromide		
Hydroxycarbamide		Scopolamine		
		Suxametonium		
		Tetracaine		
		Tubocurarine		

Antineoplastic agents included in Appendix № 9 to Art. 17, para. 1 of Ordinance № 28/9.12.2008.

Approximately half of the drugs included in Appendix № 9 belong to the Antineoplastic agents (37 drugs). They refer to the following groups of antineoplastic agents: alkylating agents, antimetabolites, plant alkaloids, cytotoxic antibiotics and other antineoplastic agents. These drugs have different mechanism of action and therapeutic indications, but most of them have serious adverse reactions that may be even life-threatening in some patients. This is due to the fact that antineoplastic agents are toxic to cancer cells as well as to the normal cells (Colvin 2003).

In Table 2 are shown alkylating agents that are listed in the Appendix. The mechanism of action of alkylating agents is based on a covalent interaction between an alkyl group of the drug and DNA (usually the N7 position of guanine). This result in cross-linking of double-stranded DNA and inhibition of DNA replication in the tumor cells. However, alkylating agents also lead to DNA damage in the normal cells that divide frequently and this is the reason for their cytotoxic effects to the gastrointestinal tract, bone marrow, testicles, ovaries and other tissues. The nitrogen mustard analogues are the most frequently used alkylating agents. Some of the alkylating agents require activation by a microsomal P450 enzyme or undergo microsomal metabolism. As a result, drug interactions with CYP450 substrates may occur leading to an increased risk of toxic effects (Colvin 2003). Another alkylating agent included in Appendix № 9 is mitolactol (Dibromodulcitol; DBD) but it doesn't have an ATC code. It's a conformational isomer of mitobronitol (Dibromomannitol; DBM) which refers to the group of other alkylating agents. Mitolactol is under clinical investigation for treatment of different types of cancer (Simonetti et al. 2014; Jeney et al. 2017).

Antimetabolites are anticancer drugs that substitute the actual metabolites in the DNA synthesis and thus inhibit DNA replication, cancer cell growth and survival. Antimetabolite agents are folic acid analogues, purine analogues and pyrimidine analogues. Methotrexate is folic acid analogue that competitively inhibits dihydrofolate reductase, an essential enzyme in the synthesis of tetrahydrofolic acid (THFA). The latter is the active form of folic acid in humans and is needed as a cofactor in the synthesis of thymidylate, purine nucleotides and several amino acids. Therefore, reduced production of THFA by methotrexate leads to an inhibition of DNA/RNA and protein synthesis in cancer cells (Howard et al. 2016). Purine and pyrimidine antimetabolites diffuse into cells and are metabolized respectively into purine and pyrimidine analogues that inhibit enzymes essential for DNA synthesis. Therefore, purine and pyrimidine analogues inhibit DNA replication, resulting in DNA damage and cell death (Parker 2009). Purine analogues have also an immunosuppressive effect and are used in the treatment of autoimmune diseases (LiverTox 2014). Antimetabolites listed in Appendix № 9 are presented in Table 3.

Some plant alkaloids and other natural products have also significant anticancer effect in different types of tumors. Anticancer drugs of plant origin included in Appendix № 9 belong to the class of Vinca alkaloids, Podophyllotoxin derivatives, Taxanes and Topoisomerase 1 (TOP1) inhibitors (Table 4). Vinca alkaloids are derived from *Catharanthus roseus* and other plants of the genus *Vinca*. Natural vinca alkaloids and their analogues are cell cycle–specific cytotoxic drugs. They bind to tubulin dimers and inhibit their polymerization and microtubules formation thus block cancer cell growth and division (Martino et al. 2018; Madsen et al. 2019). Etopo-

Table 2. Alkylating agents included in Appendix № 9 (Simonetti et al. 2014; Jeney et al. 2017; Colvin 2003; WHO 2023a, e, f, g).

INN ATC code	Pharmacotherapeutic group	Therapeutic indications	Undesirable effects
Alkylating agents			
Cyclophosphamide L01AA01	Nitrogen mustard analogues	Malignant lymphomas (Hodgkin's disease, lymphocytic lymphoma, mixed-cell type lymphoma, histiocytic lymphoma, Burkitt's lymphoma); multiple myeloma, leukemias, mycosis fungoides, neuroblastoma, adenocarcinoma of ovary, retinoblastoma, breast carcinoma	Hematopoietic toxicity (myelosuppression, immunosuppression, bone marrow failure, serious or fatal infections); Gastrointestinal toxicity (nausea, vomiting, diarrhea); Gonadal toxicity; Pulmonary Toxicity; CNS toxicity (convulsions, seizures); Alopecia; Teratogenicity; Secondary malignancies; Renal and bladder toxicity (Cyclophosphamide, Ifosfamide, Nitrosoureas); Cardiotoxicity (Cyclophosphamide)
Chlorambucil L01AA02		Chronic lymphocytic leukemia, ovarian carcinoma, malignant lymphomas	
Melphalan L01AA03		Multiple myeloma, ovarian carcinoma, breast cancer, Hodgkin and Non-Hodgkin's lymphoma, Precursor Cell Lymphoblastic Leukemia-Lymphoma, Acute Myeloid Leukemia (AML), Neuroblastoma	
Ifosfamide L01AA06		Testicular tumors and sarcomas	
Busulfan L01AB01	Alkyl sulfonates	Chronic myelocytic leukemia (CML), acute myeloid leukemia	
Carmustine L01AD01	Nitrosoureas	Brain tumors, multiple myeloma, Hodgkin and non-Hodgkin's lymphoma	
Lomustine L01AD02		Brain tumors, Hodgkin's lymphoma	
Fotemustine L01AD05		Metastatic melanoma, Primary brain tumors	
Mitobronitol L01AX01	Other alkylating agents	Chronic myeloid leukemia	
Dacarbazine L01AX04		Hodgkin's disease, Melanoma	

Table 3. Antimetabolites included in Appendix № 9 (Parker 2009; LiverTox 2014; Howard et al. 2016; Wang et al. 2019; WHO 2023d, k, l).

INN ATC code	Pharmacotherapeutic group	Therapeutic indications	Undesirable effects
Antimetabolites			
Methotrexate 1. L01BA01	1. Folic acid analogues	1. Breast cancer, leukemias, lymphomas, lung cancer, osteosarcoma, head and neck tumors	Nausea, vomiting, diarrhea, mucositis, nephrotoxicity (including acute kidney injury), hepatotoxicity, neurotoxicity, myelosuppression
2. L04AX03	2. Other immunosuppressants	2. Autoimmune diseases (i.e. rheumatoid arthritis, psoriasis, lupus, Crohn's disease)	
Mercaptopurine L01BB02	Purine analogues	Acute lymphoblastic leukaemia	Nausea, vomiting, diarrhea, loss of apetite, hepatotoxicity, myelosuppression (leukopenia), acute pancreatitis
Tioguanine L01BB03		Acute lymphoblastic leukaemia; Acute myelogenous leukemia	
Cytarabine L01BC01	Pyrimidine analogues	Acute lymphoblastic leukaemia; Acute myeloid leukemia; Chronic myeloid leukemia; Prophylaxis and treatment of meningeal leukemia	Nausea, vomiting, diarrhea, mucositis, leukopenia, thrombocytopenia, hair loss, neurotoxicity and pericarditis
Fluorouracil L01BC02 L01BC52		Colorectal, breast, stomach, and pancreatic cancer, head and neck cancer, brain cancer, liver cancer	Severe gastrointestinal toxicity (nausea, vomiting, diarrhea, stomatitis, gastritis); myelosuppresion; cardiac toxicity; neurotoxicity; alopecia
Tegafur L01BC03 L01BC53			

Table 4. Plant alkaloids and other natural products included in Appendix № 9 (de Man et al. 2018; Martino et al. 2018; Madsen et al. 2019; LiverTox 2020; Farrar and Jacobs 2023; WHO 2023j, m, n, o).

INN ATC code	Pharmacotherapeutic group	Therapeutic indications	Undesirable effects
Plant alkaloids and other natural products			
Vinblastine L01CA01	Vinca alkaloids and analogues	Hematological and lymphatic neoplasms, as well as of solid tumors (i.e. breast cancer, non- small cell lung cancer	Chemotherapy-induced peripheral sensory and/or motor neuropathy (numbness, paresthesia, impaired balance, altered gait, constipation, paralytic ileus, urinary retention, orthostatic hypotension); myelosuppresion; hair loss
Vincristine L01CA02			
Vinorelbine L01CA04			
Etoposide L01CB01	Podophyllotoxin derivatives	Solid tumors (testicular cancer, small cell lung cancer, ovarian cancer), leukemia and lymphoma	Myelosuppresion (neutropenia, anemia, thrombocytopenia); nausea, vomiting, diarrhea; alopecia
Teniposide L01CB02			
Paclitaxel L01CD01	Taxanes	Ovarian cancer, breast cancer, lung cancer, pancreatic cancer, Kaposi's sarcoma	Bone marrow suppression (neutropenia); neuropathy; alopecia; nausea, vomiting, diarrhea; cardiotoxicity; hypersensitivity reactions
Irinotecan L01CE02	Topoisomerase 1 (TOP1) inhibitors	Colrectal cancer, small cell lung cancer, pancreatic cancer	Severe gastrointestinal toxicity (nausea, vomiting, diarrhea); neutropenia; asthenia

side and teniposide are semisynthetic derivatives of podophyllotoxin, the active ingredient of the resin Podophyllin, derived from the rhizomes of *Podophyllum peltatum*. They inhibit topoisomerase II enzyme that is essential for DNA replication, resulting in single- and double-strand breaks in DNA and promotes apoptosis (LiverTox 2020). Topoisomerase I inhibitors are also used as anticancer drugs. Member of this group is irinotecan that is a semisynthetic derivative of camptothecin, a cytotoxic alkaloid extracted from the plant *Camptotheca acuminata*. Irinotecan binds to the Topoisomerase I – DNA complex, leading to DNA damage, inhibition of DNA replication and cell death (de Man et al. 2018; LiverTox 2020). Taxanes are natural antineoplastic drugs extracted from the bark of *Taxus brevifolia* or their semisynthetic derivatives. They bind to tubulin, stabilize the microtubules and block their disassembly. In result, taxanes stop the cell cycle and induce apoptosis (Farrar and Jacobs 2023).

The most widely used cytotoxic antibiotics are anthracyclines and their derivatives. Cytotoxic antibiotics included in the Appendix are shown on Table 5. Anthracyclines are natural compounds, produced by different strains of *Streptomyces bacterium*. The mechanism of action of anthracyclines is not fully understood. However, two main mechanisms of anthracyclines cytotoxic activity have been proposed. These are formation and accumulation of reactive oxygen species that cause oxidative stress and DNA intercalation and inhibition of Topoisomerase II isoenzymes, leading to double-strand breaks in DNA. In result, anthracyclines cause DNA damage, inhibit DNA and RNA synthesis and initiate programmed cell death (McGowan et al. 2017; Radeva-Ilieva et al. 2020). Bleomycin and mitomycin are the other cytotoxic antibiotics, included in Appendix № 9. Bleomycin is a member of the family of natural glycopeptide antibiotics (bleomycins), produced by the Gram-positive bacteria *Streptomyces verticillus* while mitomycin is a member of mitomycins, aziridine-containing natural antibiotics isolated from *Streptomyces caespitosus*. The mode of action of bleomycin is associated mostly with signle- stranded DNA breaks and inhibition of DNA replication. The anticancer activity of mitomycin is due to induction of DNA cross-linking and blockade of DNA synthesis (LiverTox 2017; Sinawe and Casadesus 2022).

Other antineoplastic agents include platinum compounds, methylhydrazines and drugs with different structure (Table 6). The platinum anticancer drugs contain a platinum ion (Pt) linked to different ligands (-Cl, -NH3 or other). Their mechanism of action is similar to that of alkylating agents. They form covalent cross-links in DNA that prevent DNA replication and cause cell death. Procarbazine is a member of Methylhydrazines that acts like the alkylating agents (Colvin 2003). Amsacrine is an inhibitor of Topoisomerase II enzyme, that cause double-strand breaks in DNA, similar to anthracyclines (Cassileth and Gale 1986). Asparaginase metabolizes the amino acid asparagine that plays an important role in proteins biosynthesis. Unlike normal cells, some tumor cells have a limited ability to synthesize asparagine, which is why it diffuses from the extracellular fluid. Asparaginase treatment leads to reduced asparagine levels in serum, resulting in inhibition of protein synthesis in cancer cells and induction of apoptosis (Lopes et al. 2017). Hydroxycarbamide, also known as hydroxyurea, blocks ribonucleotide reductase, an enzyme involved in deoxyribonucleotides production. Thus, it inhibits DNA synthesis in cancer cells (Jinna and Khandhar 2022). Estramustine is used as estramustine phosphate which is dephosphorylated in the gastrointestinal tract. Estramustine is a normustine ester of estradiol that acts as an agonist of the estrogen receptors and leads to suppression of androgens production, such as testosterone. In addition, estramustine phosphate has a direct cytotoxic effect due to tubulin and microtubule-associated proteins binding and induction of microtubules depolymerization. The result is inhibition of mitosis and initiation of apoptosis in tumor cells (Qin et al. 2016).

In Tables 7, 8 is summarized information about the abovementioned anticancer drugs that are authorized in Bulgaria and their brand names. Defined daily doses (DDDs) have not been established in this group because of highly individualized use and wide dosage ranges.

Table 5. Cytotoxic antibiotics and related substances included in Appendix № 9 (Evison et al. 2016; LiverTox 2017; McGowan et al. 2017; Radeva-Ilieva et al. 2020; Sinawe and Casadesus 2022).

INN ATC code	Pharmacotherapeutic group	Therapeutic indications	Undesirable effects
Cytotoxic antibiotics and related substances			
Doxorubicin L01DB01	Anthracyclines and related substances	Leukemias (i.e. acute lymphoblastic leukaemia), lymphomas (i.e. Hodgkin and non-Hodgkin's lymphoma), breast, stomach, uterine, ovarian, bladder cancer, and lung cancers; prostate cancer (Mitoxantrone); multiple sclerosis (Mitoxantrone)	Nausea, vomiting, cardiotoxicity (congestive heart failure); neutropenia; alopecia; secondary hematologic malignancy; nephrotoxicity
Daunorubicin L01DB02			
Epirubicin L01DB03			
Idarubicin L01DB06			
Mitoxantrone L01DB07			
Bleomycin L01DC01	Other cytotoxic antibiotics	Head and neck cancer (i.e. mouth, tongue, nasopharynx, larynx); cervical cancer; Hodgkin and non-Hodgkin's lymphoma; testicular cancer	Pulmonary toxicity (pneumonitis, pulmonary fibrosis); severe idiosyncratic reaction (hypotension, mental confusion, fever, chills); renal and hepatic toxicity, rash, hyperpigmentation, alopecia
Mitomycin L01DC03		Stomach and pancreatic cancer, breast cancer, bladder cancer; Low- grade upper tract urothelial cancer (UTUC)	Bone marrow suppression, nausea, vomiting, diarrhea, Hemolytic Uremic Syndrome (HUS) (hemolytic anemia, thrombocytopenia, renal failure); liver and pulmonary toxicity

Table 6. Other antineoplastic agents included in Appendix № 9 (Cassileth and Gale 1986; Colvin 2003; Qin et al. 2016; Lopes et al. 2017; Jinna and Khandhar 2022; WHO 2023h).

INN ATC code	Pharmacotherapeutic group	Therapeutic indications	Undesirable effects
Other antineoplastic agents			
Cisplatin L01XA01	Platinum compounds	Lymphoma, squamous cell carcinoma of the head and neck, ovarian cancer, bladder cancer, testicular cancer, cervical cancer	Gastrointestinal toxicity (severe nausea and vomiting), renal toxicity, neurotoxicity (paresthesia, weakness, tremor, seizures), ototoxicity, anemia
Procarbazine L01XB01	Methylhydrazines	Hodgkin's disease, Primary brain tumors	Myelosuppression, nausea, vomiting, diarrhea, gonadal toxicity, pulmonary toxicity, CNS toxicity, CNS depression, hypertension, alopecia, secondary malignances, immunosuppression
Amsacrine L01XX01	Other antineoplastic agents	Acute lymphoblastic leukemia	Myelopuppresion, nausea, vomiting, mucositis, hepatotoxicity, arrhythmias
Asparaginase L01XX02		Acute lymphoblastic leukemia	Severe anaphylactic reactions, hepatotoxicity, acute pancreatitis, coagulation disorders, hyperglycemia, myelosuppresion
Hydroxycarbamide L01XX05		Sickle cell disease, chronic myeloid leukemia, cervical cancer	Bone marrow suppression (leukopenia), secondary malignancies (skin cancer), cutaneous vasculitic toxicities, skin toxicity, gastrointestinal toxicity, gout, neurotoxicity, interstitial lung disease
Estramustine L01XX11		Prostate cancer	Gastrointestinal toxicity (nausea, vomiting, diarrhea), gynecomastia, erectile dysfunction, impotence, allergic reactions, angioedema, cardiotoxicity, thrombosis, leukocytosis

Table 7. Access to alkylating agents and antimetabolites included in Appendix № 9 in Bulgaria (BDA, Register of pharmaceutical products).

INN	Prescription drugs	Marketing authorization for use in Bulgaria	Brand name, dose, dosage form
Alkylating agents			
Cyclophosphamide	✓	✓	Endoxan 200 mg, 500 mg or 1 g powder for sol. for inj.; 50 mg coat. tabl.
Chlorambucil	✓	✓	Leukeran 2 mg film-coat. tabl.
Melphalan	✓	✓	Phelinun 50 mg or 200 mg powder and solvent for concentrate for sol. for inf.
Ifosfamide	✓	✓	Holoxan 500 mg, 1 g or 2 g powder for sol. for inf.
Busulfan	✓	✓	Busilvex 6 mg/ml concentrate for sol. for inf.
Carmustine	✓	✓	Carmustine Obvius 100 mg powder and solvent for concentrate for sol. for inf.
Lomustine	✓		
Fotemustine	✓	✓	Mustophoran 208 mg powder and solvent for sol. for inf.
Mitobronitol	✓		
Dacarbazine	✓		
Antimetabolites			
Methotrexate	✓	✓	Methotrexate Ebewe, Namaxir 2,5mg, 5 mg or 10 mg tabl.
			Methotrexate Ebewe, Methotrexate Accord 100 mg/ml concentrate for sol. for inf.;
			Ebetrexat 10 or 20 mg/ml sol. for inj.; Injexate 50 mg/ml sol. for inj.
Mercaptopurine	✓	✓	Puri – Nethol 50 mg tabl. Xaluprine 20 mg/ml oral suspension
Tioguanine	✓		
Cytarabine	✓	✓	Alexan 50 mg/ml sol. for inj.
			Cytarabin Accord 100 mg/ml sol. for inj. or inf.
Fluorouracil	✓	✓	5-Fluorouracil Ebewe, Fluorouracil Accord 50 mg/ml sol. for inj. and inf.;
Tegafur	✓		

Table 8. Access to plant alkaloids, cytotoxic antibiotics and other antineoplastic agents included in Appendix № 9 (BDA, Register of pharmaceutical products).

INN	Prescription drugs	Marketing authorization for use in Bulgaria	Brand name, dose, dosage form
Plant alkaloids and other natural products			
Vinblastine	✓		
Vincristine	✓	✓	Cytocristin 1mg/ml sol. for inj.
Vinorelbine	✓	✓	Vinorelbin Ebewe 10 mg/ml concentrate for sol. for inf.
Etoposide	✓	✓	Etoposide Accord/ Etoposide Ebewe / Etosid 20 mg/ ml concentrate for sol. for inf.
Teniposide	✓		
Paclitaxel	✓	✓	Paclitaxel Ebewe/ Paclitaxel Accord/ Paclitaxel Bulgermed/ Genexol 6 mg/ ml concentrate for sol. for inf.
Irinotecan	✓	✓	Irinotecan Accord/ Irinotecan Actavis/ Irinotecan Bulgermed/ Irinotecan Novamed/ Neotecan 20 mg/ ml concentrate for sol. for inf.
Cytotoxic antibiotics and related substances			
Doxorubicin	✓	✓	Doxorubicin Ebewe, Doxorubicin Accord 2 mg/ml conc. for sol. for inf.; Doxorubicin Stada 2 mg/ml sol. for. inj.
Daunorubicin	✓		
Epirubicin	✓	✓	Epirubicin Ebewe, Episindan, Doxorubicin Ebewe Farmorubicin PFS 2 mg/ml conc. for sol. for inj./ inf.;
Idarubicin	✓	✓	Zavedos 5 mg or 10 mg capsules, hard; Zavedos 5 mg or 10 mg powd. for sol. for inf.
Mitoxantrone	✓	✓	Mitoxantron Ebewe 2 mg/ml conc. for sol. for inf.
Bleomycin	✓		
Mitomycin	✓	✓	Mytomycin Accord 2 mg, 10 mg or 20 mg powder for sol. for. inj./inf.
Other antineoplastic agents			
Cisplatin	✓	✓	Cisplatin Ebewe 0,5 or 1 mg/ml conc. for sol. for inf.; Cisplatin Accord 1 mg/ml conc. for sol. for inf.
Procarbazine	✓		
Amsacrine	✓		
Asparaginase	✓	✓	Kidrolase 10 000 IU powder for sol. for. inj./inf.
Hydroxycarbamide	✓	✓	Hydrea 500 mg capsules, hard
Estramustine	✓		

Immunomodulating agents included in Appendix № 9 to Art. 17, para. 1 of Ordinance № 28/9.12.2008

Immunosuppressants included in Appendix № 9 are ciclosporin and azathioprine. Ciclosporin is a calcineurin inhibitor (L04AD01, DDD = 0.25 g O/P; S01XA18), while azathioprine (L04AX01, DDD = 0.15 g O/P) refers to the group of other immunosuppressants (WHO, Calcineurin inhibitors; WHO, Other immunosuppressants). Ciclosporin blocks the activity of calcineurin, a protein which activates the T cells of the immune system and stimulates

the production of cytokines. Azathioprine is a purine analog and inhibits purine synthesis, leading to production of less DNA and RNA for the synthesis of white blood cells. In result, both drugs lead to immunosuppression and are used in transplanted patients to prevent rejection and to treat autoimmune diseases (rheumatoid arthritis, granulomatosis, Crohn's disease, ulcerative colitis and systemic lupus erythematosus). Ciclosporin is also authorized for use in the European Union to treat severe vernal keratoconjunctivitis (VKC) and severe keratitis in adult patients with dry eye disease. In these cases, it is applied locally in the eye. The most serious side effects of these immunosuppressants are bone marrow suppression, anemia, an increased risk of infection and lymphoma, kidney toxicity is characteristic for ciclosporin while azathioprine can lead to severe hepatic impairment (Mohammadi and Kassim 2023; Tapia et al. 2023). Ciclosporin is metabolized extensively in the liver mainly by CYP3A4 enzyme and inhibits the activity of CYP3A4 and P- glycoprotein. Thus, there is an increased risk of drug interactions if taken together with other drugs especially CYP3A4 substrates (Tapia et al. 2023). In Bulgaria ciclosporin is registered under the brand name Sandimmun Neoral (soft capsules, 25, 50 and 100 mg and oral solution, 100 mg/ml) and azathioprine is sold under the brand name Imuran 50 mg film-coated tablets (BDA, Register of pharmaceutical products).

In the present study, we analyzed and summarized the available information about the anticancer drugs and immunosuppressants included in Appendix № 9 to Art. 17, para. 1 of Ordinance № 28/9.12.2008, issued by the Minister of Health. In result, we established that some of them are not registered for use by the Bulgarian Drug Agency (BDA) to date and are not used in clinical practice in Bulgaria. Furthermore, there is a number of antineoplastic agents that have a marketing authorization granted by the BDA or are authorized for use under a centralized procedure under Regulation (EC) №726 / 2004 of the European Parliament and of the Council of 31 March 2004 but are not included in Appendix № 9. An example is the anticancer agent capecitabine, a prodrug of 5-fluorouracil (5-FU), that is widely used to treat colorectal carcinoma. Capecitabine is a pyrimidine analogue, such as fluorouracil but unlike it capecitabine is not included in Appendix № 9 although it is metabolized in the body to 5-FU (Stoeva et al. 2020). More examples are given in Table 9.

It remains unclear why these anticancer drugs and immunosuppressants shown in table 8 are not included in the Appendix. Moreover, in Appendix № 9 is not included any representative of Protein kinase inhibitors (ATC code: L01E), Monoclonal antibodies and antibody drug conjugates (ATC code: L01F) as well as other antineoplastic agents (ATC code: L01X) that are the main drugs in the targeted cancer therapy and are widely used in recent years. In addition, many protein kinase inhibitors as well as monoclonal antibodies have marketing authorization

Table 9. Medicines that are authorized in Bulgaria but are not included in Appendix № 9 (Ministry of Health, Ordinance № 28; BDA, Register of pharmaceutical products).

Pharmacotherapeutic group	INNs		
	Included in Appendix № 9 with marketing authorization in Bulgaria*	**Included in Appendix № 9 without marketing authorization in Bulgaria***	**Not included in Appendix № 9, but have marketing authorization in Bulgaria***
Alkylating agents, Nitrogen mustard analogues	Cyclophosphamide, Chlorambucil, Melphalan, Ifosfamide	–	Bendamustin, Chlormethine hydrochloride
Alkylating agents, Alkyl sulfonates	Busulfan	–	Treosulfan
Alkylating agents, Ethylene imines	–	–	Thiotepa
Alkylating agents, Other alkylating agents	–	Mitobronitol, Dacarbazine	Temozolomide
Antimetabolites, Folic acid analogues	Methotrexate	–	Pemetrexed
Antimetabolites, Purine analogues	Mercaptopurine	Tioguanine	Fludarabine, Clofarabine, Cladribine, Nelarabine
Antimetabolites, Pyrimidine analogues	Fluorouracil, Tegafur, Cytarabine	–	Capecitabine, Gemcitabine, Azacitidine, Decitabine, Trifluridine
Plant alkaloids and other natural products, Vinca alkaloids and analogues	Vincristine, Vinorelbine	Vinblastine	Vinflunine
Plant alkaloids and other natural products, Podophyllotoxin derivatives	Etoposide, Teniposide	–	–
Plant alkaloids and other natural products, Taxanes	Paclitaxel	–	Docetaxel, Cabazitaxel
Plant alkaloids and other natural products, Topoisomerase 1 (TOP1) inhibitors	Irinotecan	–	Topotecan
Plant alkaloids and other natural products, other plant alkaloids and other natural products	–	–	Trabectedine
Cytotoxic antibiotics and related substances, Anthracyclines and related substances	Doxorubicin, Epirubicin, Idarubicin, Mitoxantrone, Daunorubicin in combination	–	Pixantrone
Cytotoxic antibiotics and related substances, other cytotoxic antibiotics	Mitomycin	Bleomycin	–
Other antineoplastic agents, Platinum compounds	Cisplatin	–	Oxaliplatin, Carboplatin

Pharmacotherapeutic group	INNs		
	Included in Appendix № 9 with marketing authorization in Bulgaria*	Included in Appendix № 9 without marketing authorization in Bulgaria*	Not included in Appendix № 9, but have marketing authorization in Bulgaria*
Other antineoplastic agents, other antineoplastic agents	Asparaginase, Hydroxycarbamide	Procarbazine, Amsacrine, Estramustine	Mitotane, Pegaspargase, Arsenic trioxide, Anagrelide, Aflibercept, Talimogene laherparepvec, Venetoclax, Tagraxofusp, Axicabtagene ciloleucel, Tisagenlecleucel, Eribulin, Selinexor
Immunosuppressants, Calcineurin inhibitors	Ciclosporin	–	Tacrolimus, Voclosporin,
Immunosuppressants, Other immunosuppressants	Azathioprine, Methotrexate	–	Lenalidomide, Pirfenidone, Pomalidomide, Dimethyl fumarate, Darvadstrocel, Thalidomide, Diroximel fumarate

* by the BDA or under a centralized procedure.

in Bulgaria via the Centralized procedure pursue to Regulation (EC) No 726/2004 of the European Parliament and of the Council of 31 March 2004 (BDA, Register of pharmaceutical products).

Conclusion

In conclusion, there are some INNs that are listed in Appendix № 9 but are currently not available on the Bulgarian market. At the same time, there are many antitumor and immunosuppressive medicines that have marketing authorization for use in Bulgaria but are not included in the Appendix. Overall, it seems that Appendix № 9 is incomplete and is not updated recently. In this regard, it remains unclear whether the correct and safe storage of medicines is ensured. Thus, it is necessary to develop clear and precise criteria for the inclusion and exclusion of drugs in Appendix № 9.

Acknowledgments

The authors have no support to report.

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