Pain management is clearly a major focus for nAChR therapy.

Because nAChR modulate the release of dopamine and stimulate locomotor behavior, Parkinson's disease is another therapeutic target for nicotinic drugs. David McClure (SIBIA) described SIB1508Y, a 5′-alkyne derivative of nicotine. This agonist appears to be selective for α4β2 nAChR and is in Phase II clinical trials for Parkinson's disease. When given to rats at the same time as they received a 6-hydroxydopamine lesion of the nigrostriatal tract, there was some sparing of the tyrosine-hydroxylase positive neurones. In an MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) monkey model of Parkinson's disease, SIB1508Y reputedly potentiated the effects of low and subthreshold doses of L-DOPA (3,4-dihydroxy-L-phenylalanine).

Future prospects

Through the evaluation of nAChR diversity, the development of agonists demonstrating improved subtype selectivity, and the demonstration of encouraging physiological and behavioural results, nAChR seems to be firmly established as an appropriate target for the treatment of a spectrum of conditions. Improved understanding of the specific phamacophore for each of the nAChR subtypes should lead to more selective agonists. Furthermore, greater knowledge of the subunit composition and distribution of nAChR subtypes expressed in the human brain, their functional properties and physiological significance will facilitate a more rational approach to ameliorating dysfunction. As most of the applications of nicotinic agents are likely to require long-term administration, questions of tolerance development and consequent changes in receptor numbers and function need to be addressed. So far, most research has been directed towards the development of agonists. Given the complex balance between receptor activation, desensitization, inactivation and regulation, evaluation of the effects of partial agonists and antagonists might be an alternative focus, while the allosteric modulation of nAChR is an attractive potential therapeutic approach. Steroids, peptides, local anaesthetics and antidepressants can modify receptor activity and further research of allosteric mechanisms is clearly warranted.

This conference provided a useful forum for dialogue between researchers in drug discovery programmes in industry and basic scientists in academia. While the former depends on breakthroughs in the fundamental understanding of nAChR structure and function that come, predominantly, from the endeavours of the academic community, the latter will benefit from the desperately needed new and selective research tools that are arising through the development of new drugs. This symbiosis will stimulate basic nAChR research and nicotinic therapeutics in tandem, and great advances can be anticipated for the next IBC conference on neuronal nAChR!

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Book review

Antifolate Drugs in Cancer Therapy

by Ann L. Jackman, Humana Press, 1999. $145.00 (456 pages, hardback), ISBN 0-89603-596-4

Antifolates are compounds that interfere with various stages of folate metabolism. There is an absolute requirement for folate coenzymes in cell division and tissue growth in mammals. Hence, antifolates are useful in regulating tissue growth and proliferation as exemplified by various neoplastic diseases. In addition, they can be used in the treatment of microbial infections, inflammatory disorders and autoimmune diseases. Four decades have elapsed since the introduction of methotrexate (MTX) into the clinic for the treatment of cancer, but its major therapeutically relevant metabolites were only discovered in 1973 [Baugh, C.M., Krumdieck, C.L. and Nair, M.G. (1973) *Biochem. Biophys. Res. Commun.* 52, 27–31; Nair, M.G. and Baugh, C.M. (1973) *Biochemistry* 12, 3923–3928]. This important and interesting discovery dramatically rekindled research efforts in this field [Cover legend (1992) *Cancer Res.* April 15]. The proceedings of the first two symposia pertaining to the role of polyglutamylation in antifolate cytotoxicity are still the most comprehensive reports of the early work and led to the development of the antifolates currently used in the clinic [Goldman, I.D., Chabner, B.A. and Bertino, J.R., eds (1983) in *Folyl and Antifolyl Polyglutamates*, Plenum Press; Goldman, I.D., ed. (1985) in *Proceedings of the Second Workshop on Folyl and Antifolyl Polyglutamates*, Preager Scientific].

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The effect of polyglutamylation

Polyglutamylation is a general phenomenon of glutamate-bearing classical folate-analogue inhibitors of folate-dependent enzymes. These compounds include aminopterin, 10-deazaaminopterins [10-DAM; Nair, M.G. *et al.* (1988) *J. Med. Chem.* 31, 181–186], CB3717, tomudex, lometrexol and multi-targeted antifolate (MTA). Polyglutamylation potentiates the inhibitory capacity of thymidylate synthase (TS), glycinamide ribonucleotide formyltransferase (GARFT) and, to a lesser extent, dihydrofolate reductase (DHFR) inhibitors. However, polyglutamylation of DHFR inhibitors (such as MTX and 10-DAM) results in significant potentiation of TS and aminoimidazole carboxamide ribonucleotide formyltransferase (AICARFT) inhibition [Allegra, C.J. *et al.* (1985) *J. Biol. Chem.* 260, 9720–9726]. After the antifolate polyglutamates are formed in the cells, they are not readily effluxed, being retained for prolonged periods. Antifolate polyglutamylation potentiates the inhibition of both their target enzymes and other folate-dependent enzymes, as well as facilitating their intracellular retention, and this then partly contributes to their undesirable host toxicity.

Currently, there is no convincing evidence to demonstrate selective antifolate polyglutamylation or differential polyglutamate degradation in normal versus tumor tissues in humans. There is also no conclusive evidence to show that antifolate polyglutamylation is a crucial and desirable metabolic process that produces an improved therapeutic index for antifolate drugs. In fact, several emerging drugs such as dideazatetrahydrofolate (DDATHF), MTA and tomudex that are highly efficiently polyglutamylated exhibit severe toxicity. The only nonpolyglutamylatable classical folate-analogue inhibitor of DHFR that is currently in the clinic is 4′-methylene-10-deazaaminopterin (MDAM). Although MDAM is completely inert to polyglutamylation, it exhibits superior antitumor activity and lower toxicity in preclinical studies relative to the polyglutamylatable MTX [Cao, S. *et al.* (1996) *Clin. Cancer Res.* 2, 707–712]. This book, *Antifolate Drugs in Cancer Therapy*, has emerged at the appropriate time and focuses on the development of both polyglutamylatable and nonpolyglutamylatable antifolates for the treatment of cancer.

Another important aspect of antifolate efficacy is their metabolic transformation to entities that are inactive or toxic to normal tissues. It is well known that, in addition to polyglutamylation, MTX undergoes hydroxylation to 7-hydroxy-MTX, conversion to 4-amino-4-deoxy-10-methylpteroic acid (AMTA) and cleavage at the 9–10 position. All these products are poor inhibitors of DHFR and do not contribute to antitumor activity. Hydroxylation of antifolates possessing a pteridine ring is mediated by aldehyde oxidase. The biochemical pharmacology of MTX is further complicated by the fact that 7-hydroxy-MTX competes with MTX for polyglutamylation and transport via the RFC [Abraham, A. *et al.* (1996) *Cell. Pharmacol.* 3, 29–34]. The polyglutamates of 7-hydroxy-MTX could be directly toxic to kidney and liver cells [Smeland, E. *et al.* (1994) *Proc. Am. Assoc. Cancer Res.* 35, 302]. Competition of 7-hydroxy-MTX with MTX for RFC effectively lowers the concentration of the active drug in tumor cells. Therefore, blocking polyglutamylation and 7-hydroxylation of DHFR inhibitors could lead to better pharmacological control of these drugs in clinical situations.

In chapter 4, excellent arguments in support of developing nonpolyglutamylatable inhibitors of DHFR have been presented. These arguments have also been advanced as rationale for the development of nonpolyglutamylatable antifolates such as GW1843, AG337 and ZD9331. However, unlike MTX, the inability of these compounds (including MTA) to undergo oxidative deactivation could contribute to further enhancement of activity, and this has not been adequately addressed in these chapters.

Importance of polyglutamylation

The work described in this book on the development of nonpolyglutamylatable antifolates emphasizes the current rationale advanced by a few investigators, this being to modulate polyglutamylation to reduce host toxicity and achieve better pharmacological control. Furthermore, the data presented addresses the key issue of whether polyglutamylation of DHFR, TS and GARFT inhibitors is an absolute requirement for eliciting clinically exploitable antitumor activity. It is tempting to conclude that, if therapeutically effective concentrations of an antifolate drug can be reached and retained in target tissues for a sufficiently long time, and be cleared by simple adjustment of the dosage or schedule (to optimize therapeutic effectiveness and to reduce drug-induced host toxicity), then polyglutamylation becomes a less significant and even unnecessary determinant of therapeutic success. In this context, the influx and efflux characteristics of an antifolate become more important as therapeutic determinants than polyglutamylation. These aspects are presented in sufficient detail by Jansen in chapter 14, and makes not only delightful reading but is also extremely useful for medicinal chemists, pharmacologists and clinicians alike. Elucidation of the structures and identities of the various carriers and receptors that mediate folate and antifolate influx and efflux, as well as understanding their regulation in tumor versus normal tissues, will undoubtedly contribute to the rational design and development of more tumor-selective antifolate drugs.

An emerging and exciting area in cancer therapy is the proper exploitation and extension of the Kisliuk Effect (the pronounced folic acid enhancement of synergism observed with lipophilic DHFR inhibitors when mixed with polyglutamylatable inhibitors of TS, GARFT

and AICARFT) observed from *in vitro* to clinical situations *in vivo*. Preliminary efforts in this area have already begun in several laboratories and these are mentioned in chapter 13, which might stimulate additional reading on this subject. The book has placed emphasis on the design and development of therapeutic entities that act against the folate-dependent enzymes, TS and GARFT. Although these antifolates represent the newest drugs under development, the book fails to address the investigations pertaining to the development of inhibitors of other folate-dependent enzymes such as AICARFT, folylpolyglutamate synthetase (FPGS) and folylpolyglutamate hydrolase (FPGH). Furthermore, the chapter on fluoropyrimidines might have been more appropriate in a book dealing with pyrimidine antimetabolites rather than antifolates.

The chapters on thymineless death and genetic determinants demonstrate the importance of how antifolate-induced DNA damage triggers molecular events that initiate cell death through apoptosis, and highlights the important involvement of p53 tumor suppressor gene in this process within the current framework of available knowledge and data. These chapters also provide extremely useful reading when attempting to understand the underlying molecular mechanisms of cytostatic versus cytotoxic effects of antifolate antimetabolites.

Conclusion

Despite the existence of several excellent reviews, a textbook that addresses the theory, rationale, and biochemical and clinical pharmacology of newer antifolates has not yet been available. The present title fulfills this gap at the appropriate time by providing a base for stimulating, innovative and utilitarian research in antifolates. This book is highly recommended and is essential reading for advanced students, medicinal chemists, pharmacologists and oncologists.

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Enhancing information sharing

It currently takes 10–12 years for a new drug to reach the market. However, once the patent has expired on the new drug entity, the company loses most of the revenue on that particular drug. It has been estimated that for every extra day taken in the development stage, the company loses an average of $1 million [*The Delphi Report on Knowledge Management* (1997) Delphi Consulting Group]. One important aspect of getting a drug to the market quickly is teamwork. In the pharmaceutical and biotechnology industries, this teamwork involves several different groups of workers, often between many different global sites. Xerox (Stamford, CT, USA) have devised a number of packages designed to save time during the drug discovery and development processes, registration and packaging [Lawrence, R.N. (1999) *Pharm. Sci. Technol. Today* 2, 390–391]. The company say these packages enable access to current knowledge (AskOnce™), distributing and sharing it between the different groups (DocuShare™) and monitoring the information (Eureka™).

Accessing knowledge

Throughout the drug discovery process, it is essential to remain aware of any similar research being carried out by other companies so that an early decision can be made whether to proceed with the research. However, such information is often indexed, catalogued and stored in numerous databases, both externally on the Internet and internally, and can only be accessed by using a combination of different methods. AskOnce is a query package that searches several Web search engines (e.g. AltaVista, Excite), Web databases (e.g. the US Patent Office), legacy databases (e.g. Oracle), document repositories (e.g. Documentum, DocuShare) and groupware (e.g. Lotus Notes, Microsoft Exchange) simultaneously in one query. This therefore reduces the number of different searches necessary and eliminates the need to combine the results of several different searches. New links to databases and search engines can be created, queries can search non-indexed information to view the most up-to-date information and it can be set to run continually so that any emerging relevant information is immediately highlighted.

A query can also be translated into the language of the database that it is programmed to search. Furthermore, the package has been designed to reduce the number of hits returned, being reported to compress the output of these queries by up to ten-fold. The results of the queries are expressed in a common format, and to simplify examination of the output, instant summaries of documents can be produced (5–6 sentences in the required language)

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