Amino Acids (2004) 26: 307–309
DOI 10.1007/s00726-004-0076-6

Amino Acids
Printed in Austria

Polyamines and cancer: Minireview article

U. Bachrach

Department of Molecular Biology, Hebrew University-Hadassah Medical School, Jerusalem, Israel

Received December 12, 2003
Accepted January 15, 2004
Published online June 22, 2004;

Summary. The naturally occurring polyamines, spermine, spermidine and the diamine putrescine are widespread in nature. They have been implicated in growth and differentiation processes. Polyamines accumulate in cancerous tissues and their concentration is elevated in body fluids of cancer patients. Assays of urinary and blood polyamines have been used to detect cancer and to determine the success of therapy. Drugs which inhibit the synthesis of polyamines can prevent cancer and may also be used for therapeutic purposes. Ornithine decarboxylase, which catalyzes the rate limiting step in polyamine synthesis, can serve as a marker of proliferation. Recently, a new *in vitro* chemosensitivity test, based on the disappearance of ornithine decarboxylase in drug-treated cancer cells has been developed. The increasing interest in polyamines and their physiological functions may lead to a more extensive application of these compounds or their derivatives in cancer diagnosis and treatment.

Keywords: Cancer – Polyamines – Spermine – Spermidine – Putrescine – Ornithine decarboxylase – Cancer therapy – Diagnosis and prevention

Introduction

Numerous early studies indicated that polyamines accumulate in tissues undergoing rapid growth (Caldarera et al., 1965; Raina and Jänne, 1968). Bacterial growth was also stimulated by polyamines (Bachrach and Weinstein, 1970). It is therefore not surprising that scientists have been looking for polyamines in neoplastic tissues.

Hämälainen (1941), used primitive gravimetric methods to show that bone marrow of leukemic patients was rich in polyamines. Subsequently, Rosenthal and Tabor (1956) reported that blood of lymphoma bearing mice as well as carcinoma, hepatoma and lymphoma tissues of rodents were rich in polyamines. Bachrach et al. (1967), used reliable enzymatic methods to estimate polyamines in Ehrlich ascites cells. Williams-Ashman et al. (1972), demonstrated the correlation between putrescine levels and the growth rate of Morris rat hepatomas. These findings were confirmed by Russell and Durie (1978), who demonstrated that putrescine accumulated in fast growing hepatomas, and that the spermidine/spermine ratios depended on the growth rate.

Polyamines and ornithine decarboxylase in chemically and virus-induced cancers

Tumor promoting agents, which induce skin tumors in mice, increased ornithine decarboxylase activity and caused polyamine accumulation (O'Brien et al., 1975). Tissue cultures were also used to demonstrate the involvement of polyamines in malignant transformation. Tumor viruses such as Rous sarcoma virus (Don et al., 1975), polyoma virus (Goldstein et al., 1976) and mouse sarcoma virus (Gazdar et al., 1976), transformed cultured cells, and increased polyamine levels and their synthesis. Oncogenes like *ras*, (Pakala et al., 1988) *jun* and *fos* (Tabib and Bachrach, 1994) induced transformation which was related to polyamine biosynthesis.

Polyamines in urine of cancer patients

Russell (1971) reported that patients with various types of cancer, excrete polyamines in their urine. It has been suggested (Russell et al., 1971) that measuring urinary polyamines, excreted as conjugates, may be useful in the diagnosis of cancer and predicting the efficacy of chemotherapy or surgery. Acetyl spermidine was identified as the urinary spermidine conjugate and was detected in the urine of patients with chronic leukemia, reticulum cell sarcoma and other tumors (Tsuji et al., 1975) and in cultured cells transformed by Rous sarcoma virus (Bachrach and Seiler, 1981).

This increased interest in polyamines, in relation to cancer, culminated in a Symposium on Polyamines in Normal and Neoplastic Growth, organized by the National Cancer Institute (Baltimore, U.S.A., 1973), in which the possible clinical application of polyamines was discussed.

Polyamines as a diagnostic tool

The studies of Russell (1971) were confirmed by many investigators who found that polyamines were present in the urine of 70–90% cancer patients (Bachrach, 1989; Matsumoto and Suzuki, 1989). The fact that only 80% of cancer patients have high urinary polyamines was explained by Chayen et al. (1985), who speculated that amine and/or diamine oxidases could be responsible for the reduction of urinary polyamine. As polyamines are not elevated in the urine of all cancer patients, this method cannot be used for cancer screening, giving false negative results in approximately 20% of cancer patients. The other proposed use of polyamines in cancer therapy, was the assessment of remission/relapse status. In this case, serial analyses have to be carried out and only those individuals who have elevated polyamines in their body fluids can be studied (Russell and Durie, 1978). Successful treatment causes a significant increase in polyamines within 72 hours, thereafter, normal base values are obtained. In many cases a relapse is associated with an increase in polyamine levels (Russell and Durie, 1978).

Polyamines can also be detected in *blood* and *cerebrospinal fluids.* Takami et al. (1979) concluded that erythrocyte polyamines can be used as tumor markers. As more than 90% of total blood polyamines are associated with erythrocytes, plasma or serum polyamines are rarely used as a tumor marker. Moulinoux and his associates carried out extensive studies on polyamines in human erythrocytes (Moulinoux et al., 1991) and concluded that erythrocyte spermine and spermidine levels could serve as an index for tumor progression. This method was also used for follow-up studies of patients with operated malignant glioma and glioblastoma (Moulinoux et al., 1991). In 40% of recurring cancer patients, increase in polyamine levels was observed 1–6 months prior to the appearance of the first clinical and radiological signs of the recurrence of the brain tumor (Moulinoux et al., 1991). Polyamine assays of cerebrospinal fluids had a sensitivity of 90% for medulloblastoma and glioma (Marton et al., 1979).

Repeated studies suggested that polyamines cannot be used for the early detection of cancer and for screening of population, as ''normal'' urinary and blood polyamine may vary significantly (Yodfat et al., 1988). On the other hand, repeated analyses of an individual, and the increase in polyamines could suggest the beginning of a malignant process.

As expected, polyamine levels are elevated in the urine of pregnant women. Acetyl putrescine reached highest values at the end of pregnancy, while diacetyl spermine peaked at the end of pregnancy (van den Berg et al., 1988). If pregnancy and liver diseases are excluded, false positive results are minimal.

The enthusiasm concerning the clinical use of polyamines, which peaked around 1975, subsequently declined. According to Matsumoto and Suzuki (1989) ''the interest in this subject declined specially in the U.S. because of difficulty in obtaining grant support for such studies''. On the other hand, urinary polyamines have been officially recognized as tumor markers in Japan (Matsumoto and Suzuki, 1989) and commercially diagnostic polyamine kits were produced. In France, Moulinoux, continued to use polyamines as a tumor maker.

It is generally accepted that the determination of polyamines in biological fluids can be a useful tool for the assessment of cancer chemotherapy and the detection of remission and relapse. This could certainly lead to the improvement of the treatment of cancer patients.

Inhibition of polyamine biosynthesis and chemoprevention

The pharmaceutical industry became interested in polyamines and various inhibitors of polyamine biosynthesis were produced. This subject will be discussed by H. M. Wallace, elsewhere in this issue. Of all the inhibitors α-difluoromethylornithine (DFMO), an irreversible inhibitor of ornithine decarboxylase (ODC), appeared to be most promising. However, this drug failed to cure cancer, mainly because of the amplification of the ODC gene and rescue of the cells by increased rates of polyamine uptake.

Cancer chemoprevention and *in vitro* chemosensitivity tests

As polyamines are apparently essential for tumor growth, it was reasonable to presume that inhibition of polyamine biosynthesis can cure and/or prevent cancer. Studies with animals demonstrated that DFMO can inhibit skin tumor production, by inhibiting polyamine biosynthesis (Takigawa et al., 1983). It has, therefore, been suggested

that DFMO can be used as a cancer preventive agent. However, cancer prevention in humans was hampered because of toxicities, which were not observed in animals (Meyskens and Gerner, 1999). Extensive studies are now carried out, to define the dose of DFMO, which does not cause audiometric side effect (Meyskens et al., 1998). Further studies, including combined therapy are certainly warranted.

For the past decade attempts have been made to determine the *in vitro* chemosensitivity of tumor cells to anticancer drugs. Most of these attempts failed because of low plating efficiency and false positive results. Recently, a new *in vitro* chemosensitivity test, based on the disappearance of ornithine decarboxylase (ODC) has been proposed (Wang et al., 1999). The presence of ODC in drug-treated cells can be monitored by determining enzyme activity (Assaraf et al., 1994) or by the detection of ODC by immunohistochemical methods (Wang et al., 1999). This approach opens new avenues for improved cancer therapy.

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Authors' address: Uriel Bachrach, Ph.D., Department of Molecular Biology, Hebrew University-Hadassah Medical School, Jerusalem, 91120, Israel,

Fax: 972-2-678 4010, E-mail: bachur@md.huji.ac.il