Proceeding S.Z.P.G.M.I vol: 6 (1992), pp. 14-19.

# Serum Sialic Acid as a Tumour Marker and the Effect of Therapy in Cancer Patients

**Mansoor ul Hassan Alvi, Sikander Ali Shaikh**
Department of Chemical Pathology, Shaikh Zayed Hospital, Lahore

## INTRODUCTION

Tumour markers are substances that quantitatively reflect the presence of malignant disease. They are either secreted in body fluids or found on cell surface as antigens[1]. Some of these tumour markers are synthetized by the tumour and others are produced in reaction to the presence of a tumour. Chemically the term refers to a molecule that can be detected in plasma or other body fluids[2]. Although sialic acid and siayl transferase levels have been studied as tumour markers, sialic acid has not been included in the established tumour markers[2-5].

Sialic acids are amino sugars widely distributed in tissues and micro organisms. Chemically they are N or O-acylated derivatives of nine carbon 3 deoxy 5 amino sugars called neuraminic acid[6]. N acetyl neuraminic acid (NANA) is the main component of carbohydrates in glycoproteins of cell membrane[7]. They participate in post-translational modifications of proteins. Because sialic acids possess relatively strong carboxyl groups ($pK_a$ 2.6 for NANA) their presence in glycoproteins or at cell surface imparts a negative charge. Mayhew[8] showed that the mean electrophoretic mobilities of some types of tumour cells increased during the mitotic phase because of an increased surface density of sialic acids. The sialic acids in carbohydrate groups of bio-polymers are hydrolyzed in vivo by neuraminidase. The desialation may be a factor in the rate of turnover of plasma glycoproteins including circulating peptide hormones. Sialic acid thus prolongs the half life of peptide hormones and modulates the activities of the enzymes present in the cell[9].

This study consists of the evaluation of serum sialic acid level in 60 control subjects and 195 patients who were diagnosed and treated for various types of cancers.

## MATERIALS AND METHODS

Two hundred and one patients from Oncology and medical wards of Shaikh Zayed Hospital, Lahore, and Oncology Ward of Mayo Hospital, Lahore, were registered. Six patients later on proved to be non-cancer and the remaining 195 patients with different types of malignancies were included in the study. The control group of 60 healthy subjects were staff members, postgraduate students and employees of Shaikh Zayed Hospital.

Five ml of venous blood from anticubital vein was drawn from each control subject. From the patients two samples were drawn, one before the treatment and the other 7 to 10 days after treatment (surgical, chemotherapeutic, radio therapeutic or combined). Blood was allowed to clot for half an hour at room temperature (25-28 °C). Serum was separated by centrifugation and the clear supernatant was removed and stored at -25 °C until assayed. Before assay the serum samples were allowed to thaw at room temperature.

Serum sialic acid concentration was assayed in duplicate by the method of Shamburger[10]. NANA, 1.293 mmol/L (400 mg/l) was used as standard.

The spectrophotometric measurements were made on spectronic 21 UVVIS spectrophotometer. NANA from human urine was obtained from Sigma Chemical Co. (St Louis, MO, USA) and 4 dimethylaminoazo benzaldehyde from E. Meck (Darmsttat Germany). Other reagents were of analytical grade from various sources.

## RESULTS

The data of controls and cancer patients regarding their age and sex is shown in Table 1.

The cancer patients were subdivided into 20 groups according the type of malignancy as shown in the table. The largest group is that of blood disorders grouped under leukemia (43 patients) followed by lymphoma (26 patients) gastrointestinal (19 patients) breast, hepatobiliary and prostate cancer.

The results of assay of serum sialic acid in controls and in cancer patients before and after the treatment are depicted in Table 2.

**Table 1: Data of normal and cancer patients.**

| Group | Diagnosis | Total patients | Males | Females | Mean Age (Years) |
|---|---|---|---|---|---|
| A | Normal (Control) | 60 | 36 | 24 | 39 |
| B | Cancer patients | 195 | 111 | 84 | |
| 1. | Leukaemia | 43 | 30 | 13 | 23 |
| 2. | Lymphoma | 26 | 21 | 5 | 38 |
| 3. | Gastrointestinal cancer | 19 | 11 | 8 | 53 |
| 4. | Breast cancer | 18 | - | 18 | 50 |
| 5. | Hepatobiliary cancer | 16 | 9 | 7 | 54 |
| 6. | Ovarian cancer | 12 | - | 12 | 53 |
| 7. | Prostate cancer | 11 | 11 | - | 62 |
| 8. | Multiple myeloma | 7 | 4 | 3 | 66 |
| 9. | Urinary bladder cancer | 7 | 4 | 3 | 51 |
| 10. | Lung cancer | 7 | 6 | 1 | 60 |
| 11. | Pancreatic cancer | 6 | 4 | 2 | 51 |
| 12. | Cervix cancer | 6 | - | 6 | 46 |
| 13. | Laryngeal cancer | 3 | 3 | - | 48 |
| 14. | Bone cancer | 3 | 2 | 1 | 40 |
| 15. | Skin cancer | 3 | 3 | - | 50 |
| 16. | Testicular cancer | 2 | 2 | - | 48 |
| 17. | Thyroid cancer | 2 | 1 | 1 | 53 |
| 18. | Malignant ascites | 2 | - | 2 | 19 |
| 19. | Rhabdomyosarcoma | 1 | - | 1 | 5 |
| 20. | Renal carcinoma | 1 | - | 1 | 45 |

**Table 2: Serum sialic acid in normals and cancer patients.**

| Sr. No. | Diagnosis | Mean Sialic acid (mmol/l) before treatment | S.D. | Mean Sialic acid (mmol/l) after treatment | S.D. |
|---|---|---|---|---|---|
| A | Normal (Control) | 1.36 | 0.10 | | |
| B | Cancer patients | | | | |
| 1. | Leukaemia | 2.43 | 0.29 | 1.78 | 0.34 |
| 2. | Lymphoma | 2.42 | 0.29 | 1.56 | 0.17 |
| 3. | Gastrointestinal cancer | 2.42 | 0.20 | 1.63 | 0.42 |
| 4. | Breast cancer | 2.46 | 0.28 | 1.39 | 0.08 |
| 5. | Hepatobiliary cancer | 2.46 | 0.33 | 1.61 | 0.40 |
| 6. | Ovarian cancer | 2.56 | 0.36 | 1.39 | 0.08 |
| 7. | Prostate cancer | 2.40 | 0.24 | 1.47 | 0.18 |
| 8. | Multiple myeloma | 2.62 | 0.23 | 2.23 | 0.43 |
| 9. | Urinary bladder cancer | 2.56 | 0.31 | 1.86 | 0.15 |
| 10. | Lung cancer | 2.66 | 0.20 | 1.88 | 0.34 |
| 11. | Pancreatic cancer | 2.35 | 0.49 | 1.91 | 0.41 |
| 12. | Cervix cancer | 2.52 | 0.23 | 1.51 | 0.19 |
| 13. | Laryngeal cancer | 2.18 | 0.48 | 1.53 | 0.11 |
| 14. | Bone cancer | 2.71 | 0.43 | 1.19 | 0.68 |
| 15. | Skin cancer | 2.26 | 0.39 | 1.79 | 0.28 |
| 16. | Testicular cancer | 2.19 | 0.13 | 1.27 | 0.21 |
| 17. | Thyroid cancer | 2.30 | 0.12 | 1.36 | 0.05 |
| 18. | Malignant ascites | 2.30 | 0.16 | - | - |
| 19. | Rhabdomyosarcoma | 2.52 | - | 2.04 | - |
| 20. | Renal carcinoma | 2.91 | - | 2.64 | - |

**Table 3: Effects on sialic acid after therapy.**

| Sr. No. | Diagnosis | Total Patients | Expired/ LAMA | No response | Positive response % |
|---|---|---|---|---|---|
| 1. | Leukaemia | 43 | 5 | 6 | 85 |
| 2. | Lymphoma | 26 | 3 | 2 | 91 |
| 3. | Gastrointestinal cancer | 19 | 2 | 1 | 94 |
| 4. | Breast cancer | 18 | 1 | 0 | 100 |
| 5. | Hepatobiliary cancer | 16 | 3 | 1 | 92 |
| 6. | Ovarian cancer | 12 | 2 | 0 | 100 |
| 7. | Prostate cancer | 11 | 2 | 0 | 100 |
| 8. | Multiple myeloma | 7 | 2 | 1 | 80 |
| 9. | Urinary bladder cancer | 7 | 0 | 0 | 100 |
| 10. | Lung cancer | 7 | 2 | 1 | 80 |
| 11. | Pancreatic cancer | 6 | 2 | 1 | 80 |
| 12. | Cervix cancer | 6 | 1 | 0 | 100 |
| 13. | Laryngeal cancer | 3 | 1 | 0 | 100 |
| 14. | Bone cancer | 3 | 0 | 1 | 66 |
| 15. | Skin cancer | 3 | 1 | 1 | 50 |
| 16. | Testicular cancer | 2 | 0 | 0 | 100 |
| 17. | Thyroid cancer | 2 | 0 | 0 | 100 |
| 18. | Malignant ascites | 2 | 2 | 0 | - |
| 19. | Rhabdomyosarcoma | 1 | 0 | 0 | 100 |
| 20. | Renal carcinoma | 1 | 0 | 1 | 0 |

**Table 4: Response depending upon the nature of therapy.**

| Sr. No. | Nature of treatment | Male patients | Female patients | Total patients | Number of patients response | % response to therapy | % decrease is sialic acid after treatment |
|---|---|---|---|---|---|---|---|
| 1. | Surgical | 36 | 44 | 80 | 76 | 94 | 41 |
| 2. | Chemotherapy | 32 | 18 | 52 | 44 | 85 | 28 |
| 3. | Radiotherapy | 18 | 4 | 22 | 19 | 86 | 22 |
| 4. | Chem + Radio | 10 | 1 | 11 | 9 | 82 | 39 |

In the control group mean sialic acid level was 1.36±0.1 (SD) mmol/L as compared to 1.74 mmol/L reported by Shamburger[10]. Comparing the means of the serum sialic acid level of patient groups with each other, there was no significant difference ($P > 0.05$) while the difference between the mean of the control group and those of cancer groups is highly significance (P values in all case $< 0.001$). Serum sialic acid level appears to be a reliable indicator of all types of malignancies.

Out of a total 195 patients, 188 patients had the serum sialic acid level more than 2.00 mmol/L. Three of the remaining seven had serum levels > mean control +2 SD and the remaining four levels slightly below this value. There is no case below the mean of the control group comprising zero percent false negative.

The sensitivity of sialic acid as a tumour marker is shown in Table 3.

It is seen that sialic acid is a very sensitive tumour maker for all types of cancer. These patients were selected randomly as they presented themselves in the clinics. The number of patients in groups 13 to 20 is 3 or less each and the 100% sensitivity in these patients is not statistically valid. However in this series serum sialic acid level appears to be a very sensitive tumour marker.

The patients were treated with chemotherapy, radio therapy or a combination of both. There was a decrease in the mean sialic acid levels in all the groups indicating the response to the treatment. The number of patients responding to different therapies is given in Table 4.

**Table 5: Comparison of various tumour markers with sialic acid.**

| Sr. No. | Type of malignancy | Tumour marker | Reference | Sensitivity | % elevated sialic acid |
|---|---|---|---|---|---|
| 1. | Hepatocellular carcinoma | Alpha fetoprotein | Brumm et al. 1989[25] | 24 | 94 |
| 2. | Carcinoma of cervix | CA-125 | Duk et al. 1990[12] | 40 | 100 |
| 3. | Renal cell carcinoma | Tissue polypeptide antigen | Gohji et al. 1990[27] | 42 | 100 |
| 4. | Carcinoma of prostate | Prostatic antigen | Hudson et al. 1989[28] | 44 | 100 |
| 5. | Renal cell carcinoma | Basic fetoprotein | Gohji et al. 1990[27] | 47 | 100 |
| 6. | Hepatocellular carcinoma | Alpha fucosidase | Bukofzer et al. 1989[24] | 75 | 94 |
| 7. | Renal cell carcinoma | Immunosuppressive protein | Gohji et al. 1990[25] | 79 | 100 |
| 8. | Carcinoma of cervix | Carcinomembryonic antigen | Duk et al. 1989[26] | 80 | 100 |
| 9. | Carcinoma of breast | Carcinomembryonic antigen | Coombes et al. 1977[30] | 81 | 100 |
| 10. | Carcinoma of prostate | Prostatic antigen | Powell et al. 1989[31] | 90 | 100 |
| 11. | Carcinoma of thyroid | Calcitonin | Uddelsman et a. 1989[32] | 90 | 100 |

The extent of response between individuals or patient groups cannot be compared because of the type of cancer, the type of therapy and the dosage of therapy. The response varied between 82 to 94 percent of different groups indicating that sialic acid level is a reliable indicator of response in individuals.

## DISCUSSION

Biochemical tumour markers have been studied extensively[2,5]. They may be highly specific for a particular tumour or may shown a generalized trend. They may be useful for one or more of the modalities such as screening, diagnosis, prognosis, monitoring therapeutic response and detecting relapse. A tumour marker can be most useful if it can detect malignancy in asymptomatic persons before the disease reaches a symptomatic and often incurable stage. However, their role in monitoring therapy and detecting relapse are equally important. Nearly all markers can be elevated in benign disorders (false positive) which decreases their specificity. Serum sialic acid levels and those of the associated enzymes have been studies by a number of workers[11-20] but no comprehensive study has been reported for sialic acid as a tumour marker. The present study is a comprehensive study of serum sialic acid levels in various tumours and the response to therapy. As no previous study is available in the local population 60 normal controls were included in the study comprising 36 males and 24 females. One hundred and ninety five cancer patients included 111 males ad 84 females with comparable male:female ratio of control.

In transformed cells sialic acid concentration was 60% of the normal[21]. Decrease in sialic acid in cells is due to the loss of o-acetyl substitute in sialic acid[21]. This decrease cellular concentration removes the inhibition in the malignant or transformed cells, to unrestricted growth. Serum sialic acid in different studies was found elevated in the cancers of the breast[15], genito urinary tract[17] bronchus[18], malignant ascites[19], lung cancer, leukemia, lymphoma and melanoma[22]. The serum levels in all the groups of cancer patients were significantly elevated ($P<0.01$) from the mean control level.

Mean sialic acid level in our controls, was 1.36±0.11 SD, compared to 1.74±0.24 reported by Shamburger[10], 1.6±0.3 by Alvi[23] and 1.75 mmol/L by Loranze[24]. Similarly the mean level of cancer patients was from 2.35 to 2.66±0.2 to 0.48 (SD), in groups 1 to 12 in 178 patients out of 195 excluding the groups of 3 patients or less, The ratio of patient mean/control mean is 1.8 in this study compared to 1.7 in Shamburger's[10]. The results of the two studies are comparable. The elevation in our study corresponded to the stage of malignancy and to the state of metastases. Values as high as 3.16 to 2.90 mmol/L were obtained in acute lymphoblastic leukemia, carcinoma of the breast and multiple myeloma. The higher the serum sialic, acid, the more unfavourable the prognosis. The patient with the carcinoma of breast had highest serum sialic acid level of 3.16 mmol/L and died shortly after taking the pre-treatment blood sample.

The response to therapy is shown in Table 3. The cancers responded to all forms of therapy with 19 to 46% decrease in serum sialic level of pre-treatment level after 7 to 10 days after the treatment as shown in the table. Further follow up of treatment and follow up of the patient was not made in this study.

Serum sialic acid is not specific but very sensitive tumour marker. Its sensitivity varies from 82 to 100% for various types of tumours. In table 5 the sensitivity of the serum sialic acid is compared with the sensitivity reported for other tumour markers.

It is seen that serum sialic acid is a more sensitive tumour marker than any other reported tumour marker. Colli et al.[19] also reported the sensitivity of serum sialic acid level to be 82% for malignant ascites. Serum sialic acid level has the advantage that it is independent of age and sex[24]. Sialic acid determination by the method of Shamburger[10] is also very cost effective and therefore can be a valuable screening procedure. Serum sialic acid thus has the advantage of reliability of malignancy, sensitivity, cost effectiveness, and precision over other tumour markers.

## REFERENCES


1. **Marshall WJ, Clayton BE.** Illustrated Textbook of Clinical chemistry. Philadelphia. J.B. Lippincott, 1988: 280.
2. **Liu FJ, Duben-Von Laufen JL, Bishop M.** In: Bishop ML, Duben-Von Laufen JL and Fody EP eds. Clinical Chemistry Principles, procedures correlations, Philadelphia, JB Lippincott 1985. 495-522.
3. **Silver KHB, Karim KA, Archibald EL, Salinas FA.** Serum sialic acid and Sialyltraneferase as Monitors of Tumour Burden in Malignant Melanoma. *Cancer Research 1979;* **39**: 5036-5042.
4. **Henn KH, Gressner AM.** Zur Klnischen Wetigkeit der Sialinsaure in serum bei beningen und malignant Erkrankungan. *J Clin Chim Clin Biochem 1987;* **25**: 423-430.
5. **Bates E.** Clinical applications of serum tumour markers. *Annals of Internal Medicine 1991;* **115**: 623-38.
6. **Orten JM, Neuhaus OW.** Human Biochemistry 9th. ed. ST Lauis CV Mosby 1975; 244-6.
7. **Warran L, Felsenfield H.** The biosynthesis of sialic acid. *J Biol Chem 1962;* **237**: 1421-31.
8. **Mayhew E.** Cellular electrophoretic mobility and the mitotic cycle. *J Gen Physiol 1966;* **19**: 717.
9. **Jaffe WL, Kaitukaitis JL.** Human Choriogonadotrophic secretory tumors. The Clinical Biochemistry of Cancer, Fleisher M. ed. Washington DC American Association of Clinical Chemistry 1979; 215.
10. **Shamberger RJ.** Serum sialic acid in normal and in cancer patients. *J Clin Chem Clin Biochem 1984;* **22**: 647-51.
11. **Shahanglan S, Fritsche HA, Hughs JI, Foemmel RS, Katopodia N.** Plasma protein bound sialic acid in patients with colorectal polyps of known histology. *Clin Chem 1991;* **37/2**: 200-204.
12. **Susheela VK, Das TK, Khurana JS, Jayawala A.** Circulating levels of Sialic acid, and glycoaminoglycans: a diagnostic test for ankylosing spondolytis. *Annals of Rheumatic Disease 1988;* **47**: 833-37.
13. **Hoon DBS, Ng SK, Ranshaw IA.** Analysis of mammary tumor cell metastases and release of bound n-acetyl neuraminic acid: *J Cancer 1985;* **51**: 775-781.
14. **Gross HJ, et al.** Transfer of synthetic sialic acid analogues to N and O linked glycoprotein glycans using four differing mammalian sialyl transferases. *Biochemistry 1989;* **28**: 7386-92.
15. **Hogan-Ryan A, Fennelly JJ, Jones M, Canwell B and Duffy MJ.** Serum sialic acid and CEA concentrations. in human breast cancer. *Br J Cancer 1980;* **41**: 587-92.
16. **Plucinsky MC, Riley WM, Prorok JH, Alhadeff J.** Total and lipid associated serum sialic acid levels in cancer patients with different primary sites and differing degrees of metastatic involvement. *Cancer 1986;* **58**: 2680-5.
17. **Erbil KM, Sen SE, Zineke H, Jones JD.** Significance of serum proteins and lipid bound sialic acid as a marker for genitourinary malignancies. *Cancer 1986;* **57**: 1389-94.
18. **Seber O, Aydilek R, Erbil K, Demirci N.** Diagnostic value of lipid-bound sialic acid levels in serum and bronchial lavage fluid in bronchial cancer. *Asian Med J 1989;* **32**: 109-115.
19. **Colli A, Buccino G, Cocciolo M, Parravinci R, Mariani F, Scaltrini G.** Diagnostic accuracy of sialic acid in the diagnosis of malignant ascites. *Cancer 1989;* **63**: 912-916.
20. **Grimes WJ.** Sialic acid transferases and sialic acid levels in normal and transformed cells. *Biochemistry 1970;* **9**: 5083-91.
21. *Sugihara K, Juss RJ.* Colorectal goblet cell sialomucin: its relation to malignant disease. *J Clin Path 1986;* **39**: 1088-95.
22. **Dnistrian AM, Schwartz MK.** Plasma lipid bound sialic acid and carcinoembryonic antigen in cancer patients. *Clin Chem 1981;* **27**: 1737-9.
23. **Alvi MH, Amer NA, Sumerin I.** Serum 5-nucleotidase and serum sialic acid in pregnancy. *Obstetrics and Gynaecology 1988;* **72/2**: 171-174.
24. **Lorenz K, Weiss J, Krass E.** Serum sialic acid in human serum and cerebrospinal fluid. Comparisons of method and reference values. *J Clin Chem Clin Biochem 1986;* **24**: 189.
25. **Brumm C, Schulze C, Charles K, Morohashi T, Kloppel G.** The significance of alpha-fetoprotein and other tumour markers in differential immunocytochemistry of primary liver tumors. *Histopath 1989;* **14**: 503-13.
26. **Duk JM, Brujin HWA, Croenier KH, Gleuren GJ, Alders JG.** Adenocarcinoma of uterine cervix. *Cancer 1990;* **65**: 1830-7.
27. **Gohji K, Ishi M, Nagata H, Matsumoto O, Kamidono S.** Serum basic fetoprotein in patients with renal cell carcinoma. *Cancer 1990;* **65**: 1405-11.
28. **Hudson MA, Bahmson RR, Catalona WJ.** Clinical use of prostate specific antigen in patients with prostate cancer. *J Urol 1989;* **142**: 1011-7.
29. **Bukofzer S, Stass PM, Dobeer M, Groenveld HT.** Alpha-L-fucosidase as serum marker of hepatocellular - carcinoma in Southern African Blacks. *Br J Cancer 1989;* **59**: 417-20.
30. **Coombes RC.** Biochemical markers in human breast cancer. *Lancet 1977;* **1**: 132-4.
31. **Powell CS, Fielding AM, Rosserk, Ames AC, Vanghton KC.** Prostatic specific antigen: a screening test for prostatic cancer. *Br J Urol 1989;* **64**: 540-6.
32. **Udelsman R, Mohjimini OA, Soper NDW, Dudley NE.** Medullary carcinoma of the Thyroid: management of persistance hypercalcitonemia utilizing Tc(V) dimercaptosuccinic acid scintiagraphy. *Br J Surg 1989;* **76**: 1278-81.

**The Authors:**

Mansoor ul Hassan Alvi,
Professor,
Department of Chemical Pathology,
Shaikh Zayed Hospital,
**Lahore**

Sikander Ali Shaikh
Department of Chemical Pathology,
Shaikh Zayed Hospital,
**Lahore**

**Address for Correspondence:**

Mansoor ul Hassan Alvi,
Professor,
Department of Chemical Pathology,
Shaikh Zayed Hospital,
**Lahore**