

5-S-CYSTEINYLDOPA IN THE URINE OF MELANOMA PATIENTS

G. Agrup, P. Agrup, T. Andersson, B. Falck, J.-A. Hansson, S. Jacobsson,
H. Rorsman, E. Rosengren and A.-M. Rosengren

*From the Departments of Dermatology, Radiotherapy, Lund and Malmö, Surgery, Plastic Surgery, Malmö,
Histology and Pharmacology, University of Lund, Lund, Sweden*

Abstract. A newly discovered amino acid, 5-S-cysteinyldopa, present in the urine of healthy subjects is excreted in pathological amounts in many patients suffering from melanoma metastases. Increased excretion of 5-S-cysteinyldopa may be observed before metastases become clinically evident. Determination of 5-S-cysteinyldopa is superior to determination of dopa+dopamine in the diagnosis of melanoma metastases.

Key words: Dopa; Dopamine; Amino acids; Catechol; 5-S-cysteinyldopa; Melanoma

Melanomas are tumours that form specific products often detectable in the urine. Several substances related to melanin formation have been investigated in the urine of melanoma patients. The classical Thormählen reaction (8) detects indols with an unsubstituted pyrrol ring. This test is positive only in advanced cases of liver metastasis. Dopa, dopamine, 3,4-dihydroxyphenylacetic acid (dopac), and homovanillic acid have all been examined in the urine of melanoma patients and the results concerning these compounds have generally been found more informative than the Thormählen reaction (9, 12, 13, 15).

A new amino acid, 5-S-cysteinyldopa, has recently been detected in human melanomas (2, 3, 6, 14). This substance is excreted in the urine of healthy subjects (1, 4), and the urinary excretion sensitively reflects pigment metabolism (11). A preliminary study has shown that the excretion of 5-S-cysteinyldopa may be increased in patients with melanoma metastases (7).

The present study was undertaken in order to ascertain the value of urinary 5-S-cysteinyldopa determination in the diagnosis of melanoma metastases. For comparison, the excretion of dopa and its decarboxylation product dopamine was also determined.

MATERIAL AND METHODS

The patients studied were 53 men and 67 women previously operated on for melanoma of the skin or eye. Such patients are examined at intervals of 3-4 months during the first 2 years after operation, and subsequently twice yearly for a further 5-10 years. At every visit the entire skin surface and all lymph nodes are examined. A liver scintigram is performed within a year of operation, and the lungs are X-rayed each year.

The patients followed up for melanoma metastases were examined for the presence of 5-S-cysteinyldopa and dopa+dopamine in the urine in accordance with methods previously described (10, 5). All urine specimens were collected in 1973 and 1974, with the exception of the summer months June, July, and August. When several determinations were performed on the same patient, only the first is reported here.

24-hour specimens of urine were collected in plastic bottles containing 50 ml acetic acid and 1 g sodium metabisulphite.

The 24-hour excretion of cysteinyldopa, dopa, and dopamine was examined in 30 men and 46 women controls (1).

RESULTS

It is evident from Figs. 1 and 2 that the excretion of 5-S-cysteinyldopa was greater in 11 of the melanoma patients than in the controls. In 7 of these 11, metastases were diagnosed at clinical examination shortly after the finding of the pathological 5-S-cysteinyldopa excretion. In some cases the finding of increased 5-S-cysteinyldopa excretion actually prompted the examination of the patients and led to the detection of the metastases.

Four patients with increased 5-S-cysteinyldopa excretion have shown no clinical signs of metastases during follow-up periods of 3-12 months.

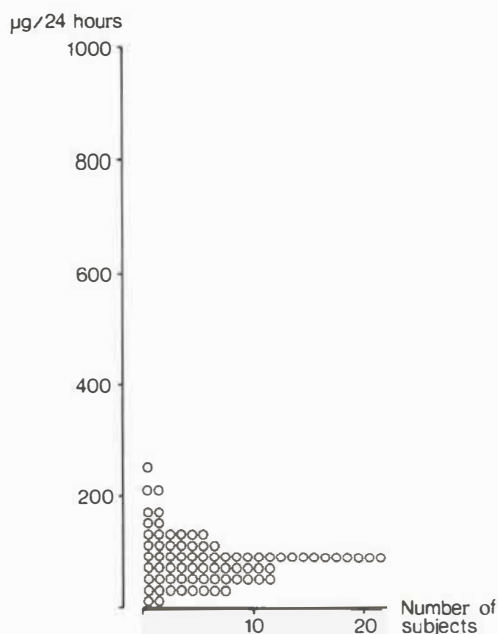


Fig. 1. Excretion of 5-S-cysteinyl-dopa in the urine of 76 healthy subjects.

Metastases were found in 3 patients with high but normal values. In 2 of these, clinical examinations leading to a diagnosis of metastasis were prompted by the 5-S-cysteinyl-dopa findings.

Among 109 patients with normal 5-S-cysteinyl-dopa values, metastases have been detected in 3. Among the 11 patients with abnormal values, metastases were demonstrated in 7. The clinical findings in the 10 patients in whom metastases were diagnosed after the urinary examination are briefly described. Patients are numbered according to 5-S-cysteinyl-dopa excretion (highest value, lowest number).

1) A 39-year-old man operated on for primary skin melanoma in 1972 developed regional lymph node metastases the same year. These were treated by surgery. Routine examination showed urinary 5-S-cysteinyl-dopa in March 1974 (5 900 µg/24 h). In August 1974 liver metastases were diagnosed.

2) A 61-year-old man was admitted to hospital owing to weight loss, confusion, and pain. Suspected primary melanoma of the skin was treated by surgery, and determination of the urinary 5-S-cysteinyl-dopa disclosed strongly pathological values (2 900 µg/24 h). The pigmented spot proved to be a primary melanoma, but no

○ no metastasis observed during follow up period 3-12 months
● metastasis observed after urine finding

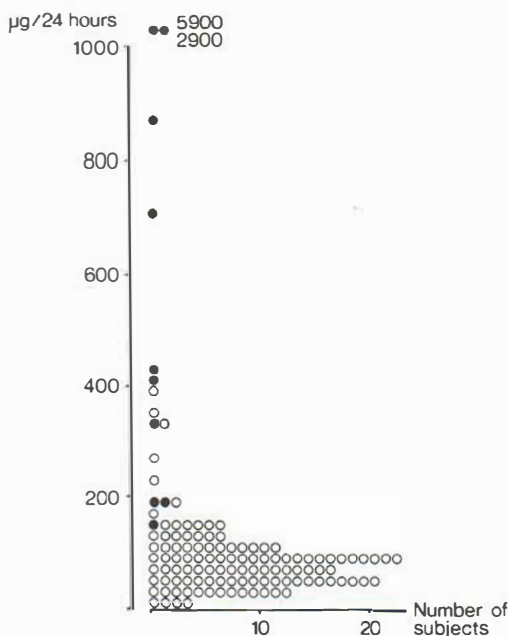


Fig. 2. Excretion of 5-S-cysteinyl-dopa in the urine of 120 melanoma patients.

metastases were found. The patient died one month later and necropsy disclosed widespread melanoma metastases.

3) A 44-year-old man was operated on for melanoma of the eye in 1971. The 5-S-cysteinyl-dopa excretion was 8/0 µg/24 h in September 1973. X-ray showed skeletal metastases.

4) An 80-year-old man with primary melanoma of the esophagus was treated surgically in March 1974. The 5-S-cysteinyl-dopa excretion was 720 µg/24 h in May 1974. A regional lymph node metastasis was diagnosed in June 1974.

5) A 63-year-old woman operated on for primary melanoma of the skin in September 1973. The 5-S-cysteinyl-dopa excretion was 430 µg/24 h in April 1974. Regional lymph node metastases were found in August 1974.

6) A 48-year-old man with no known primary melanoma was operated on for skin metastases and lymph node metastasis in 1972. In May 1974 the 5-S-cysteinyl-dopa excretion was 400 µg/24 h. He developed anaemia and haematemesis, but no metastases were found. He died in December 1974. Necropsy showed intestinal invagination due to melanoma metastases.

7) A 45-year-old man with no known primary melanoma was operated on for lymph node melanoma metastases in February 1973. The 5-S-cysteinyl-dopa excretion was 330 µg/24 h in May 1973. Signs of a cerebellar metastasis developed. He was operated on in August 1973, and the 5-S-cysteinyl-dopa returned to normal.

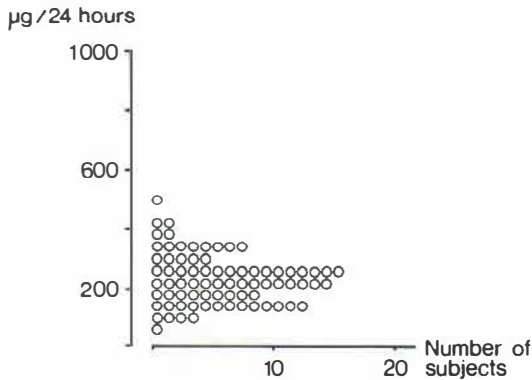


Fig. 3. Excretion of dopa+dopamine in the urine of 76 healthy subjects.

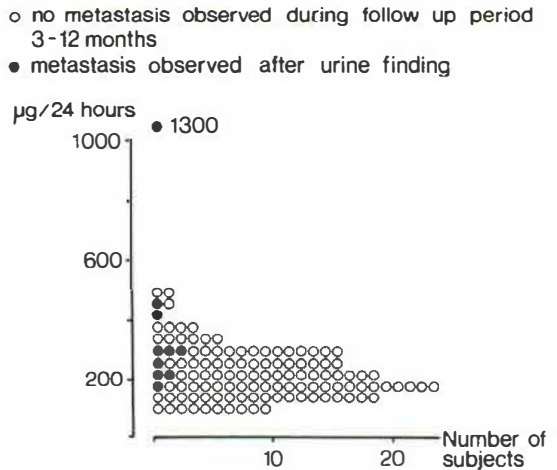


Fig. 4. Excretion of dopa+dopamine in the urine of 120 melanoma patients.

8) A 51-year-old man was operated on for primary skin melanoma in 1968 and for a pulmonary metastasis in 1972. A 5-S-cysteinyl-dopa value in the upper normal range (190 $\mu\text{g}/24\text{ h}$) was found in January 1973, and a lymph node metastasis was detected immediately afterwards.

9) A 27-year-old woman was operated on for a primary melanoma in August 1972. The 5-S-cysteinyl-dopa concentration was in the upper normal range (190 $\mu\text{g}/24\text{ h}$) in April 1974. Distant lymph node metastases were detected in May 1974.

10) A 49-year-old woman was operated on for a primary melanoma of the skin in October 1973. A 5-S-cysteinyl-dopa value in the normal range (140 $\mu\text{g}/24\text{ h}$) was noted in December 1973, and immediately afterwards regional lymph node metastases were detected.

Figs. 3 and 4 show the results of dopa+dopamine determination in controls and in melanoma patients. Dopa+dopamine excretion was clearly increased in patient No. 3, who also showed a high 5-S-cysteinyl-dopa value. The other 9 patients in whom metastases were found had normal dopa+dopamine values.

DISCUSSION

No previously published chemical methods for the detection of melanoma metabolites in the urine have been reported to give positive results prior to the detection of melanoma metastases (8, 9, 12, 13, 15). Our study shows that the determination of the urinary 5-S-cysteinyl-dopa can provide important information in following up patients operated on for melanoma: a pathological finding will in some cases

antedate symptoms and signs of metastases, giving the signal for new surgical intervention, and may well prove to be even more valuable in subsequent chemo- and immuno-therapy.

Dopa and dopamine determinations were less helpful in the prediction of metastasis. This is not surprising, since dopa is formed not only in the melanocytes but also in the neural system, and dopamine is the decarboxylation product of dopa. Only a small proportion of the dopa reaching the circulation is excreted by the kidneys. The greater part of the dopa is decarboxylated, and the dopamine formed is excreted as such or as oxidation products.

Our study was performed during the autumn, winter, and spring months. We excluded the summer months because we had observed marked excretion of 5-S-cysteinyl-dopa in the urine of healthy subjects during the summer, and therefore made a systematic investigation of the normal seasonal variations in 5-S-cysteinyl-dopa excretion (11). There is a pronounced increase in urinary 5-S-cysteinyl-dopa in summer, and the excretion of this amino acid reflects the response of the pigmentary system to UV light. The fact that 5-S-cysteinyl-dopa so sensitively reflects also the normal pigment metabolism of the body limits the usefulness of 5-S-cysteinyl-dopa determination in patients exposed to sunlight. However, the measurement of 5-S-cysteinyl-dopa excretion seems to be the best laboratory method at present available for the detection of early metastases in melanoma patients.

ACKNOWLEDGEMENTS

This investigation was supported by the Swedish Cancer Society (project 626-B74-03X) and by grants from the John & Augusta Persson Foundation for Scientific Medical Research and the Walter, Ellen & Lennart Hesselman Foundation for Scientific Research.

REFERENCES

1. Agrup, G., Falck, B., Kennedy, B.-M., Rorsman, H., Rosengren, A.-M. & Rosengren, E.: Dopa and 5-S-cysteinyl-dopa in the urine in healthy humans. *Acta Dermatovener (Stockholm)* 53: 453, 1973.
2. Agrup, G., Falck, B., Jacobsson, S., Rorsman, H., Rosengren, A.-M. & Rosengren, E.: 5-S-cysteinyl-dopa in melanomas of Caucasians. *Acta Dermatovener (Stockholm)* 54: 21, 1974.
3. Agrup, G., Andersson, T., Falck, B., Persson, K., Rorsman, H., Rosengren, A.-M. & Rosengren, E.: 5-S-cysteinyl-dopa in the plasma of melanoma patients and the renal clearance of this amino acid. *Acta Dermatovener (Stockholm)* 55: 5, 1975.
4. Agrup, G., Falck, B., Fyge, K., Rorsman, H., Rosengren, A.-M. & Rosengren, E.: Excretion of 5-S-cysteinyl-dopa in the urine of healthy subjects. *Acta Dermatovener (Stockholm)* 55: 7, 1975.
5. Anton, Aa. & Sayre, D. F.: The distribution of dopamine and dopa in various animals and a method for their determination in diverse biological material. *J Pharmacol Exp Ther* 145: 326, 1964.
6. Björklund, A., Falck, B., Jacobsson, S., Rorsman, H., Rosengren, A.-M. & Rosengren, E.: Cysteinyl dopa in human malignant melanoma. *Acta Dermatovener (Stockholm)* 52: 357, 1972.
7. Dahlqvist, I., Falck, B., Jacobsson, S., Rorsman, H., Rosengren, A.-M. & Rosengren, E.: 5-S-cysteinyl-dopa in the urine of melanoma patients. Communications from the Department of Anatomy, University of Lund, Sweden, No. 7, 1972.
8. Duchön, J. & Pechan, Z.: The biochemical and clinical significance of melanogenuria. *Ann N Y Acad Sci* 100: 1048, 1963.
9. Hinterberger, H., Freedman, A. & Bartholomew, R. J.: Precursors of melanin in the blood and urine in malignant melanoma. *Clin Chim Acta* 39: 395, 1972.
10. Rorsman, H., Rosengren, A.-M. & Rosengren, E.: A sensitive method for determination of 5-S-cysteinyl-dopa. *Acta Dermatovener (Stockholm)* 53: 248, 1973.
11. Rorsman, H., Agrup, G., Falck, B., Rosengren, A.-M. & Rosengren, E.: Exposure to sunlight and urinary excretion of 5-S-cysteinyl-dopa. *In* *Pigment Cell*, vol. 2. Karger, Basel. Unpubl. data.
12. Scott, J. A.: 3,4-Dihydroxyphenylalanine (dopa) excretion in patients with malignant melanoma. *Lancet* II: 861, 1962.
13. Takahashi, H. & Fitzpatrick, Th. B.: Quantitative determination of dopa; its application to measurement of dopa in urine and the assay of tyrosinase in serum. *J Invest Dermatol* 42: 161, 1964.
14. Vogel, C. L., Dhru, Dh., Rorsman, H., Rosengren, A.-M. & Rosengren, E.: Dopa and 5-S-cysteinyl-dopa in melanoma in Ugandan Africans. *Acta Dermatovener (Stockholm)* 54: 19, 1974.
15. Voorhess, M. L.: Urinary excretion of dopa and metabolites by patients with melanoma. *Cancer* 26: 146, 1970.

Received February 13, 1976

G. Agrup, M.D.
Department of Dermatology
Lasarettet
S-221 85 Lund
Sweden