

# Spontaneous Neoplastic Lesions in Aged Sprague-Dawley Rats

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**Abstract:** Neoplastic lesions were observed in untreated aged Sprague Dawley (SD) rats throughout their lifespan starting at 5 weeks. Their mean survival times were 89 to 105 weeks of age. The total tumor incidences were 70 to 76.7% and 87 to 95.8% in males and females, respectively. The common neoplasms were pituitary adenoma and adrenal pheochromocytoma in both sexes, testicular Leydig cell tumor in males and mammary gland tumors, thyroidal C-cell adenoma and uterine stromal polyp in females.

**Key words:** rats, spontaneous tumor, Sprague-Dawley

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## Introduction

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There have been many reports about spontaneous neoplastic lesions in aged rats [1, 3, 4, 7–13]. In evaluating the potential tumorigenicity in carcinogenicity studies, it is of importance to distinguish treatment-related lesions from spontaneous ones. In this paper, untreated SD rats aged from 5 weeks to the end of their lifespan were examined for tumor incidence and distribution of neoplastic lesions.

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## Materials and Methods

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A group of 120 male and 120 female SD rats (Group A) was obtained from Japan SLC Inc. (Shizuoka, Japan) and another group of 120 male and 120 female SD rats (Group B) was also obtained from the same sup-

plier. They were housed singly in stainless steel wire mesh cages. Group A and group B were fed *ad libitum* Clea Japan Inc. CA-1 and CE-2, respectively. CA-1 contained approximately 27.1% protein, 4.8% fat, 3.5% crude fiber and had a calculated metabolizable energy value of 342.6 kcal, while CE-2 contained approximately 25.4% protein, 4.4% fat, 4.1% crude fiber and had a calculated metabolizable energy values of 342.2 kcal. Tap water was freely available. These rats were handled during the course of the study according to the Guidelines for Animal Experimentation (Japanese Association for Laboratory Animal Science, 1987).

All rats found dead or sacrificed when moribund were necropsied, and tissues with gross lesions were taken and preserved in 10% neutral buffered formalin from the following organs: thyroid glands, parathyroid glands, salivary glands, thymus, aorta, heart, trachea, lungs,

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liver, spleen, kidneys, adrenal glands, testes, ovaries, prostate, uterus, seminal vesicle, vagina, cerebrum, cerebellum, pituitary gland, larynx, pancreas, tongue, esophagus, stomach, duodenum, jejunum, ileum, cecum, colon, urinary bladder, mandibular lymph nodes, mesenteric lymph nodes, spinal cord, sciatic nerve, femoral muscles, femur with bone marrow, sternum with bone marrow, skin, mammary gland, eyes, and ears. The tissues were subjected to routine histologic processing, staining with hematoxylin and eosin, and were examined microscopically.

## Results

As shown in Table 1, a total of 137 and 170 neoplastic lesions were seen in males and females of group A, respectively. In group B, a total of 135 and 221 neo-

plastic lesions were seen in males and females, respectively. Mean survival times were 89 and 101 weeks of age in males and females of group A, and 92 and 105 weeks of age in males and females of group B, respectively.

In group A, 13 males and 70 females died or were sacrificed moribund, while in group B, 29 males and 78 females were found dead. Most of the dead cases were supposedly the result of neoplasms and pituitary tumor, followed by lymphoma/leukemia in both sexes, and mammary gland tumors in females. Non-neoplastic fatal lesions were of chronic progressive nephropathy.

The distribution of tumor lesions is presented in Table 2, and Table 3 shows those of the organ systems with tumor incidences greater than 1%. In Table 4, the number and percent of what were possibly fatal tumors is

**Table 1.** Incidence of neoplastic lesions in groups A and B

| Items          | Number of cases |         |       |         |         |       |
|----------------|-----------------|---------|-------|---------|---------|-------|
|                | Male            |         |       | Female  |         |       |
|                | Group A         | Group B | Total | Group A | Group B | Total |
| Test animals   | 120             | 120     | 240   | 120     | 120     | 240   |
| Tumor carriers | 84              | 107     | 191   | 104     | 100     | 204   |
| Tumor lesions  |                 |         |       |         |         |       |
| Total          | 137             | 135     | 272   | 170     | 221     | 391   |
| Benign         | 125             | 100     | 225   | 125     | 175     | 300   |
| Malignant      | 12              | 35      | 47    | 45      | 46      | 91    |
| Single         | 48              | 60      | 108   | 56      | 25      | 81    |
| Multiple       | 36              | 47      | 83    | 48      | 75      | 123   |

**Table 2.** Distribution of tumor lesions in groups A and B\*

| System          | Male         |         |       |       | Female       |         |       |       |
|-----------------|--------------|---------|-------|-------|--------------|---------|-------|-------|
|                 | No. of cases |         |       | %     | No. of cases |         |       | %     |
|                 | Group A      | Group B | Total |       | Group A      | Group B | Total |       |
| Cardiovascular  | 0            | 2       | 2     | 0.83  | 2            | 0       | 2     | 0.83  |
| Hematopoietic   | 4            | 14      | 18    | 7.50  | 19           | 13      | 32    | 13.33 |
| Digestive       | 6            | 4       | 10    | 4.17  | 4            | 4       | 8     | 3.33  |
| Respiratory     | 2            | 1       | 3     | 1.25  | 1            | 1       | 2     | 0.83  |
| Urinary         | 0            | 3       | 3     | 1.25  | 0            | 2       | 2     | 0.83  |
| Genital         | 36           | 29      | 65    | 27.08 | 3            | 13      | 16    | 6.67  |
| Endocrine       | 83           | 65      | 148   | 61.67 | 105          | 123     | 228   | 95.00 |
| Integument      | 5            | 14      | 19    | 7.92  | 35           | 65      | 100   | 41.67 |
| Central nervous | 1            | 2       | 3     | 1.25  | 1            | 0       | 1     | 0.42  |
| Musculoskeletal | 0            | 1       | 1     | 0.42  | 0            | 0       | 0     | 0     |

\*Each group had 120 males and 120 females

**Table 3.** Major tumors\* in groups A and B

| System/organ    | Tumor                              | Male         |         |       |       | Female       |         |       |       |
|-----------------|------------------------------------|--------------|---------|-------|-------|--------------|---------|-------|-------|
|                 |                                    | No. of cases |         |       |       | No. of cases |         |       |       |
|                 |                                    | Group A      | Group B | Total | %     | Group A      | Group B | Total | %     |
| Hematopoietic   |                                    |              |         |       |       |              |         |       |       |
| Multiple site   | Large granular lymphocyte leukemia | 3            | 6       | 9     | 3.75  | 9            | 4       | 13    | 5.42  |
|                 | Myeloid leukemia                   |              | 4       | 4     | 1.67  |              | 6       | 6     | 2.50  |
|                 | Malignant lymphoma                 |              | 4       | 4     | 1.67  | 7            | 3       | 10    | 4.17  |
| Digestive       |                                    |              |         |       |       |              |         |       |       |
| Liver           | Hepatocellular adenoma             | 5            | 3       | 8     | 3.33  | 3            | 2       | 5     | 2.08  |
| Genital         |                                    |              |         |       |       |              |         |       |       |
| Testis          | Leydig cell tumor                  | 33           | 27      | 60    | 25.00 |              |         |       |       |
| Uterus          | Endometrial stromal polyp          |              |         |       |       |              | 10      | 10    | 4.17  |
| Endocrine       |                                    |              |         |       |       |              |         |       |       |
| Pituitary       | Adenoma                            | 41           | 44      | 85    | 35.42 | 77           | 86      | 163   | 67.92 |
|                 | Adenocarcinoma                     | 3            |         | 3     | 1.25  |              | 4       | 4     | 1.67  |
| Thyroid         | Follicular cell adenoma            | 6            |         | 6     | 2.50  |              |         |       |       |
|                 | C-cell adenoma                     |              |         |       |       | 8            | 12      | 20    | 8.33  |
| Parathyroid     | Adenoma                            | 3            |         | 3     | 1.25  |              |         |       |       |
| Adrenal medulla | Pheochromocytoma                   | 19           | 12      | 31    | 12.92 | 13           | 11      | 24    | 10.00 |
|                 | Malignant pheochromocytoma         |              |         |       |       | 2            | 2       | 4     | 1.67  |
| Pancreas        | Islet cell adenoma                 | 8            |         | 8     | 3.33  |              | 3       | 3     | 1.25  |
| Integument      |                                    |              |         |       |       |              |         |       |       |
| Cutaneous       | Keratoacanthoma                    | 3            | 3       | 6     | 2.50  |              |         |       |       |
|                 | Squamous cell carcinoma            |              |         |       |       | 2            | 3       | 5     | 2.08  |
|                 | Zymbal gland carcinoma             |              | 4       | 4     | 1.67  |              |         |       |       |
| Subcutis        | Fibrosarcoma                       |              | 3       | 3     | 1.25  |              |         |       |       |
|                 | Malignant fibrous histiocytoma     |              |         |       |       |              | 4       | 4     | 1.67  |
| Mammary         | Fibroadenoma                       |              |         |       |       | 16           | 39      | 55    | 22.92 |
|                 | Adenocarcinoma                     |              |         |       |       | 13           | 11      | 24    | 10.00 |

\*Showing incidence of more than 1%.

presented.

A case of hemangioma was observed in the spleen of a group B male, and hemangiosarcomas were seen in the uterus, mammary gland and thoracic cavity in both groups.

Lymphoma/leukemias were the most common neoplastic lesions in cases more than 1 year of age, including large granular lymphocyte leukemia, myeloid leukemia and malignant lymphoma. Malignant thymoma was observed in a female of group A.

The most common digestive tract neoplasms were hepatocellular adenoma/carcinomas appearing at more than 72 weeks of age. Jejunal and ileal adenocarcinomas and pancreatic acinar cell adenoma were also observed.

Alveolar/bronchiolar adenoma was seen in a male and a female in group A as well as in a female of group B, while alveolar/bronchiolar carcinoma was seen in a

male of group A and a female of group B.

Nephroblastoma, renal cell carcinoma, and liposarcoma were observed in 1 male, 1 female and 2 males of group B, respectively. In the urinary bladder, transitional cell papilloma was seen in a female of group B.

Leydig cell tumor appeared in 33 and 27 males in groups A and B, respectively. Also, testicular yolk sac carcinoma and mesothelioma and prostatic adenoma/adenocarcinomas were observed. In females, the most common tumors were endometrial stromal polyp, followed by endometrial stromal sarcoma and fibrosarcoma. Vaginal stromal polyp was also observed. Granulosa/theca cell tumor and malignant granulosa cell tumor were also observed.

Anterior pituitary adenoma/carcinomas were seen in 44 males and 77 females of group A as well as in 44 males and 90 females of group B. Thus, the incidence in females was twice as many as in males. In the thy-

**Table 4.** Fatal cases of tumors in groups A and B

| System/organ           | Tumor                              | Male         |         |       | Female       |         |       |
|------------------------|------------------------------------|--------------|---------|-------|--------------|---------|-------|
|                        |                                    | No. of cases |         |       | No. of cases |         |       |
|                        |                                    | Group A      | Group B | Total | Group A      | Group B | Total |
| <b>Hematopoietic</b>   |                                    |              |         |       |              |         |       |
| Multiple site          | Large granular lymphocyte leukemia | 3            | 6       | 9     | 9            | 4       | 13    |
|                        | Myeloid leukemia                   | 1            | 4       | 5     | 1            | 6       | 7     |
|                        | Malignant lymphoma                 |              | 4       | 4     | 7            | 3       | 10    |
|                        | Histiocytic sarcoma                |              |         |       | 1            |         | 1     |
| <b>Thymus</b>          |                                    |              |         |       |              |         |       |
|                        | Malignant thymoma                  |              |         |       | 1            |         | 1     |
| <b>Respiratory</b>     |                                    |              |         |       |              |         |       |
| Lung                   | Alveolar/bronchiolar carcinoma     | 1            | 1       | 2     |              |         |       |
| <b>Urinary</b>         |                                    |              |         |       |              |         |       |
| Kidney                 | Nephroblastoma                     |              | 1       | 1     |              |         |       |
| <b>Genital</b>         |                                    |              |         |       |              |         |       |
| Uterus                 | Endometrial stromal sarcoma        |              |         |       | 1            |         | 1     |
| <b>Endocrine</b>       |                                    |              |         |       |              |         |       |
| Pituitary              | Adenoma                            | 4            | 5       | 9     | 35           | 32      | 67    |
|                        | Adenocarcinoma                     | 3            |         | 3     |              | 3       | 3     |
| Thyroid                | C-cell carcinoma                   |              | 1       | 1     |              |         |       |
| Adrenal medulla        | Malignant pheochromocytoma         |              |         |       | 1            | 1       | 2     |
|                        | Complex pheochromocytoma           |              | 1       | 1     |              | 1       | 1     |
| <b>Integument</b>      |                                    |              |         |       |              |         |       |
| Cutaneous              | Zymbal gland carcinoma             | 1            | 2       | 3     |              |         |       |
|                        | Squamous cell carcinoma            |              |         |       | 1            | 1       | 2     |
| Subcutis               | Malignant fibrous histiocytoma     |              | 1       | 1     |              | 3       | 3     |
|                        | Fibrosarcoma                       |              |         |       |              | 1       | 1     |
| Mammary                | Fibroadenoma                       |              |         |       | 4            | 19      | 23    |
|                        | Adenocarcinoma                     |              |         |       | 9            | 3       | 12    |
| <b>Central Nervous</b> |                                    |              |         |       |              |         |       |
| Cerebrum               | Astrocytoma                        |              | 2       | 2     |              |         |       |
| <b>Musculoskeletal</b> |                                    |              |         |       |              |         |       |
| Bone                   | Osteosarcoma                       |              | 1       | 1     |              |         |       |

roid gland, follicular cell adenoma and C-cell adenoma/carcinomas were observed, and adenoma was observed in the parathyroid gland. Benign/malignant pheochromocytomas and complex pheochromocytoma were common in the adrenal gland, followed by cortical adenoma/carcinomas and islet cell adenoma and carcinoma in the pancreas.

The incidence of integumentary neoplastic lesions was relatively high in both males and females. In addition to benign basal cell epithelioma, keratoacanthoma, trichoepithelioma, sebaceous adenoma, and clitoral gland adenoma, carcinomas of the squamous or basal cells, Zymbal gland and clitoral gland were seen. In the subcutaneous tissue, benign lipoma and fibroma as well as malignant fibrosarcoma and malignant fibrous histiocytoma were seen. In the female mammary gland,

fibroadenoma was the most common, followed by mammary adenoma/carcinomas.

Granular cell meningioma and astrocytoma were observed in both groups, and osteosarcoma was detected in a male of group B.

## Discussion

In the both A and B groups of this study, pituitary adenoma was the most frequent tumor in males and females, followed by testicular Leydig cell tumor and adrenal pheochromocytoma in males and mammary tumors including fibroadenoma and adenocarcinomas, pheochromocytoma and thyroidal C-cell adenoma and uterine stromal polyp in females. These occurrences of tumors are quite comparable with these of previous re-

ports [1, 4, 5, 8–13].

In males of group A, deaths seemed to occur due to pituitary tumor (7 cases), leukemia/lymphomas (4 cases) and alveolar/bronchiolar carcinoma (1 case) and Zymbal gland carcinoma (1 case). In females, deaths were attributable to pituitary tumor (35 cases), leukemia/lymphomas (17 cases), mammary tumor (13 cases) and Zymbal gland carcinoma, malignant pheochromocytoma, histiocytic sarcoma, malignant thymoma or endometrial sarcoma (1 case each).

Fatal cases of group B males might have resulted from leukemia/lymphomas (14 cases), pituitary tumor (5 cases), astrocytoma or Zymbal gland carcinoma (2 cases each) and malignant fibrous histiocytoma, complex pheochromocytoma, osteosarcoma, nephroblastoma, pulmonary alveolar/bronchiolar carcinoma, or C-cell carcinoma (1 case each). In females, pituitary tumor (35 cases), mammary tumor (22 cases), leukemia/lymphomas (13 cases), malignant fibrous histiocytoma (3 cases), pheochromocytoma (2 cases) and subcutaneous fibrosarcoma or cutaneous squamous cell carcinoma (1 case each) seemed to be fatal.

Although the fatal neoplastic lesions were distributed in this study as described in previous reports of carcinogenicity studies using Sprague-Dawley derived rats [2, 6], the incidence of fatal neoplasms was relatively low in this study, probably because there were more cases of chronic progressive nephropathy causing earlier death. The tumor incidences in males and females of both groups in this study were comparable with those reported by Muraoka *et al.* [10], but much higher than in Prejean *et al.* [12] who reported incidences of 34% and 58% in males and females, respectively.

In group A of this study, tumor occurred singly in 40% of males and 46.7% of females, while multiple tumors occurred in 30% of males and 40% of females. In group B, single tumor occurred in 50% of males and 20.8% of females, and multiple tumors in 39.2% of males and 62.5% of females. Muraoka *et al.* [10] reported that multiple tumor incidences were 31.2% and 58.9% in males and females, respectively. Prejean *et*

*al.* [12], however, reported that single tumor incidences in males and females were 81.7% and 68.6%, respectively, after observation up to 19 months of age.

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