Case Report

A Spontaneous Ovarian Immature Teratoma in a Juvenile Rat

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Abstract: A spontaneous ovarian immature teratoma characterized by the presence of tissue derivatives of endoderm, mesoderm and ectoderm was investigated histopathologically in a 6-week-old juvenile Crj:CD(SD)IGS rat. The various kinds of tissue components observed in this teratoma were mainly mature elements derived from three embryonic germ layers, such as nervous tissue, skin, pancreas, salivary gland, gastrointestinal tract, respiratory tracts, striated and smooth muscle, bone with bone marrow, cartilaginous tissue and adipose tissue. Remarkable features were that this teratoma contained immature tissues intermingled with mature tissues, which typically took the form of primitive neuroepithelial rosettes and tubules and islands of immature cartilage, showing evidence of malignancy. A small yolk sac carcinoma-like focus was also observed. Neither invasion nor distant metastases were found in the other organs and tissues. This is the first report of an ovarian immature teratoma in a rat. (J Toxicol Pathol 2004; 17: 211–218)

Key words: ovary, immature teratoma, malignant teratoma, ovarian tumor, spontaneous tumor, IGS rat

The rat is an extremely valuable animal species used in toxicologic and carcinogenic studies. Although abundant pathological background data is available for major strains of rat, spontaneous tumors of the ovary are generally uncommon1–5. Rat ovarian tumors can be roughly divided into 3 main histological categories classified by their presumed histogenesis4–6. Sex cord-stromal tumors are the most common, both spontaneous and induced rat ovarian neoplasms1–3,7–11. Ovarian teratoma classified as germinal cell tumor is extremely rare in rats1–11. In this report, we describe the histopathological characteristics of the first case of an immature (malignant) ovarian teratoma in a juvenile Crj:CD(SD)IGS rat.

For the purpose of a toxicity study for the safety assessment of a new drug in the Toxicology Research Laboratories of Fujisawa Pharmaceutical Co., Ltd., 45 male and 15 female Crj:CD(SD)IGS rats were purchased at 5 weeks of age from Charles River Japan Inc. (Hino Farm, Shiga, Japan). These rats were housed in a room with a barrier system, were allowed free access to standard laboratory diet (CRF-1, Oriental Yeast Co., Ltd., Tokyo) and filtered tap water from an automatic dispenser. All animals were handled in accordance with the Guideline for Animal Experimentation of the Japanese Association for Laboratory Science. During the quarantine and acclimation period, rats were routinely observed for morbidity and mortality, and weighed weekly. On palpation, a large mass was detected in the abdomen of one female, body weight of which was 181.2 g, while the average weight of the other 14 females was 146.1 g. The animal showed no other abnormalities on clinical examination, but was not used for any study, and received a complete necropsy at 6 weeks of age. Necropsy revealed a large mass, which was round and multinodular in shape, mottled dark red color, 25 × 40 × 40 mm in size, and connected to the top of the right uterus horn with no adhesion to the surrounding tissues (Fig. 1). The right side of ovary was indistinguishable. The cut surface of the mass was solid without hemorrhage of cyst formation. Red ascites of about 23 ml was observed, without any abnormalities in the other organs or tissues. The mass and the other organs were fixed in 10% buffered formalin, embedded in paraffin. Paraffin sections at 3 μm were stained with hematoxylin and eosin (H & E) in routine processes. Additional sections of the tumor mass were stained with periodic acid-Schiff (PAS), phosphotungstic acid hematoxylin (PTAH) and Alcian-blue. Microscopically, this tumor was believed to have originated from the right ovary, since it was located at the position of the right ovary, and there was growth of normal ovarian follicles and corpora lutea at a small peripheral part of the tumor mass (Fig. 2). The tumor was characterized by the presence of various tissue elements, including both
mature and immature tissues, derived from three embryonic germ cell layers: endoderm, mesoderm and ectoderm (Table 1, Fig. 3). These findings are consistent with the diagnosis of an ovarian teratoma. The predominant mature elements were skin with dermal appendages, central and peripheral nervous tissues, cartilage, bone with bone marrow, striated and smooth muscle, white and brown adipose tissues, exocrine pancreas- or salivary gland-like glandular structure, gastro-intestinal tract-like ducts, trachea-like ducts and esophagus like ducts. Well-differentiated skin formed several cystic and projective structures lined by keratinized squamous epithelium associated with the presence of hair follicles, hair shaft and sebaceous glands. The nervous tissue component consisted of a hippocampus-like neural tissue (Fig. 4a), and in other places a mixture of neural cells with glial cells were noted. Small foci of choroid plexus, ganglia and peripheral nerve fascicle (Fig. 4b) were also formed. Striated muscle bundles, which showed stripes as clarified by PTAH staining (Fig. 5), formed atypical arrangements. Smooth muscle bundles often encircled ducts or cystic epithelial structures in some parts. Skeletal tissues predominantly formed cartilage islands (Fig. 3). The areas of intra-cartilaginous ossification and bone formation contained bone marrows that were differentiated into various stages of hematopoiesis. Formations of both white and brown adipose nests were scattered (Figs. 3 and 5). In the glandular elements, pancreatic acinus-like glands, salivary gland-like seromucous glands were observed (Fig. 6) and their secretion activities were distinctly visualized by PAS or Alcian-blue stains. Tracts lined by various kinds of epithelial cells such as ciliated or non-ciliated columnar cells, cuboidal cells, goblet cells and Paneth cells, with differentiation into both gastro-intestinal and respiratory tracts, were noted. Ductal structures which had non-keratinized squamous epithelium, similar to the esophagus, were also detected (Fig. 3). The interstitium consisted of abundant loose connective tissues. Areas of primitive neuroepithelial rosettes and tubules formation were also distinguishable (Fig. 7). They consisted of small cells with intensely basophilic nuclei and indistinct cytoplasm, orientated in a regular pattern around the central lumen. Remarkable numbers of mitotic figures were seen in these neuroepithelial cells. Furthermore, some islands of immature cartilage surrounded by the primitive mesenchymal tissue were formed (Fig. 8). A small yolk sac carcinoma-like focus, characterized by nests and ribbons of round basophilic cells which contained PAS positive
eosinophilic droplets embedded in an eosinophilic homogeneous hyalinized matrix, was found in the tumor margin (Fig. 9). Neither metastasis nor abnormal findings were detected in the other organs and tissues.

In general, rat ovarian spontaneous tumors are uncommon with an overall incidence of 1.0%\(^3\). Rat ovarian tumors are basically classified into 3 major categories, which are named according to their presumed histogenesis and
differentiation, epithelial tumors, sex cord-stromal tumors, germ cell tumors and others\textsuperscript{4–6}, in a manner similar to classifications for the mouse\textsuperscript{4,6,8,9}, domestic animals\textsuperscript{12} and human\textsuperscript{13,14}. Epithelial tumors include cystadenoma/carcinoma, tubulostromal adenoma/carcinoma and mesothelioma. Sex cord-stromal tumors include granulosa cell tumor, thecoma, luteoma, Sertoli cell tumor and sertoliform tubular adenoma. Germ cell tumors include yolk sac carcinoma, choriocarcinoma and teratoma\textsuperscript{4–6}.

Rat ovarian teratomas are defined as tumors containing any combination of well differentiated elements derived from 3 germ cell layers\textsuperscript{2–6}. These tumors are classified into benign/mature or malignant/immature teratomas\textsuperscript{2–6} in accordance with the classification of tumor pathology in the
On the basis of our findings, our case should be diagnosed as a malignant/immature teratoma due to the existence of 3 germ cell elements and primitive neuroepithelial components. The yolk sac carcinoma is one of the germ cell tumors of the rat ovary, and is occasionally seen as combination elements of experimentally induced uterine teratoma of rats. Furthermore, two cases of spontaneous bilateral testicular tumor in rats have been reported, and these tumors showed a combination of teratoma and embryonal carcinoma elements. It is known that germ cell tumors in women occur as a mixture of two or more types, i.e. dysgerminoma, yolk sac tumor, embryonal carcinoma, polyembryoma and immature teratoma, and these combinations of primitive germ cell tumors are diagnosed as mixed germ cell tumors in human pathology. Likewise, it is believed that all types of germ
cell neoplasms could be combined in rodent tumors\textsuperscript{18}, but there is no agreement for the classification as ‘mixed germ cell tumors’ in the rat among toxicologic pathologists. A small yolk sac carcinoma-like focus was detected in our case as a minor component in a large immature ovarian teratoma, and it should be thought that this focus was not so much an element of mixed tumor as an odd element of teratoma.

Spontaneous ovarian tumors are rare in many strains of rats\textsuperscript{1–5}, and almost all of them are reported in aged animals\textsuperscript{2}. In many strains of rats such as the F344 strain, nearly all ovarian neoplasms (95%) are gonadal stromal, primarily granulosa cell tumor\textsuperscript{11}. Similar results have been reported concerning spontaneous ovarian tumors in the ACI, Wistar, F344 and Donryu strains\textsuperscript{19}. While in Sprague-Dawley (SD) rats, it has been reported that approximately 73% of all ovarian tumors were Sertoli cell or sertoliform tubular tumors\textsuperscript{7}. Small incidence of (0.3–3%) of Sertoli cell tumor\textsuperscript{20}, thecoma\textsuperscript{21–23}, granulosa cell tumor\textsuperscript{21,22,24} and luteoma\textsuperscript{22} are recorded as spontaneous background data in the Crj:CD(SD)IGS rat, which is the same strain of our case. Germ cell tumors in the rat ovary are more rare than sex cord-stromal tumors and epithelial tumors. Of the 210 spontaneous ovarian tumors seen in 7748 female SD rats, none were germ cell tumors\textsuperscript{7}. In F344 rats, only 2 out of the 204 ovarian tumors from 39851 females were germ cell: one was a yolk sac carcinoma and the other was a choriocarcinoma\textsuperscript{8}. There is no description of spontaneous ovarian teratomas in major text books covering rat ovarian neoplasms\textsuperscript{3,11}, and only 2 spontaneous ovarian teratomas have been recorded by A. Maekawa\textsuperscript{2}. One of the 2 cases was benign in an old Wistar rat, and the other was malignant teratoma in a 114-week-old Donryu rat. In the latter case, various mature tissues, with atypical feature of adenocarcinomatous, sarcomatous and osteosarcomatous areas, and lung metastasis were reported. However, since this case was a particularly pronounced malignant teratoma, it may be not suitable to classify it as an immature teratoma because no primitive tissues such as neuroepithelial tissues were recorded and it was found in an aged rat. Though we can unfortunately access few pathological records of this case, we think the malignant teratoma was likely to have been a mature teratoma which had undergone malignant transformation resembling human pathology cases\textsuperscript{13,14}. On the other hand, a special mutant rat named the Tera strain produces exceptionally ovarian and testicular teratomas with high incidence (25%)\textsuperscript{25}. A few cases of rat spontaneous teratomas which originate outside of the ovary, whether they are benign/ mature or malignant/ immature types, are limited in males. Similar to rats, naturally occurring ovarian teratomas are extremely rare in most animal species\textsuperscript{2,12}, except for women\textsuperscript{13,14}, and an inbred strain of mice\textsuperscript{31,32}. In LT mice, it has been reported that 146 ovarian teratomas develop spontaneously and their incidence rose to about 50% in animals at 90 days of age\textsuperscript{32}. Many methodologies of inducing ovarian tumors in the rat have been reported. For instance, allograft of ovary to entopic site, irradiation and carcinogen can experimentally induce ovarian tumors\textsuperscript{2,3}.

| Table 2. Summaries of Rat Spontaneous Teratomas in the Literature |
|------------------|--------|--------|-------------------|-----------------|
| Strain           | Sex    | Age    | Tumor origin/ Malignancy | Components                          | Special character                   | Reference |
|                  |        |        | location            |                                |                                      |          |
| Wistar           | Female | Old    | Ovary              | Benign                          | Derived from 3 germ layers           | –         | 2        |
| Wistar           | Female | 110 wk | Ovary              | Malignant                       | Derived from 3 germ layers mixed with adenocarcinomatous and sarcomatous tissues | Cellular atypia and mitotic figures | 2        |
| Crj:CD(SD)IGS    | Male   | 8 wk   | Left testis        | Malignant/ immature             | Derived from 3 germ layers with primitive neuroepithelium | Combined with a embryonal carcinoma in the same side of the testis | 16       |
| Crj:CD(SD)IGS    | Male   | 10 wk  | One side of testis | Malignant/ immature             | Derived from 3 germ layers with primitive neuroepithelium | Combined with a embryonal carcinoma in the same side of the testis | 16       |
| Sprague-Dawley   | Male   | 6 wk   | Retroperitoneum    | –                               | Derived from 3 germ layers           | –         | 26       |
| Donryu           | Male   | 5 wk   | Pituitary gland    | Malignant/ immature             | Derived from 3 germ layers with primitive neuroepithelium and immature cartilage | –         | 27       |
| Wistar           | Male   | 5 wk   | Adrenal gland      | –                               | Derived from 3 germ layers           | –         | 28       |
| Wistar           | Male   | 10 wk  | Intracranial       | Malignant/ immature             | Derived from 3 germ layers with primitive cartilaginous matrix | Metastasis to meninges | 29       |
| SHR Wistar       | Male   | 9 wk   | Left Kidney        | –                               | Derived from 3 germ layers           | –         | 30       |

–: Not described
Whereas experimental induction of ovarian teratoma is unsuccessful, it is known that fetectomy operation experimentally induces uterine teratomas in rats.\cite{14}

The authors present a description of morphologic features of an ovarian immature teratoma naturally occurring in a juvenile rat as well as associated discussions. We hope it may stimulate further progress of expert knowledge in toxicologic pathology.

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References


