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Spontaneous Osteoarthritic Lesions in a New Mutant Strain of the Mouse

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Abstract: In our laboratory, mice showing signs of osteoarthritic lesions with cinnamon colored (yellowish-brown) hair were discovered in a colony of B6C3F₁ mice. This mouse is characterized by tiptoe walking and swelling and ankylotic changes in the ankle joint. As to radiographic findings, osteoarthritic changes, such as erosion and/or fusion of the bone tissue, were evident in the ankle joints. Histopathological characteristics included irregularity of articular surfaces caused by fissuring and/or erosion with degeneration of articular cartilage, as well as osteophytes with abnormal proliferation of chondrocytes in joint margin regions. Subsequently, ankylotic changes in the ankle joints were completed in the formation of a cartilaginous bridge and fusion of articular cavity with abnormal proliferation of cartilaginous or bone tissues. This mouse strain may provide an additional animal model that is valuable in the study of human osteoarthritis (OA).

Key words: animal model, mice, osteoarthritis, spontaneous articular lesion

The etiology of many forms of osteoarthritis (OA) in humans is still obscure [5]. Therefore, animal models simulating human OA are desired to explain the pathogenesis of this disease. Animal models for studying the pathogenesis of OA have been established, and most of them require the administration of biological, chemical or physical agents and surgical manipulation to produce the osteoarthritis-like lesions [7, 15, 16]. Several spontaneous animal models of OA have been also reported [2, 9–11], but there has been little information on the frankly ankylotic osteoarthritic lesions simulating human OA in animal models [8, 14]. In our laboratory, mice showing signs of progressive ankylotic osteoarthritic lesions with a change in the color of

hair were discovered during brother sister mating of B6C3F₁ mice (generation F₁₈). Protean manifestations of the joint in this mutant mouse open the possibility of its being a valuable model for investigating OA in humans. This report describes morphological features of the ankle joint to develop more characteristic osteoarthritic lesions.

All mice were maintained in barrier rooms conditioned to 24 ± 1°C and 55 ± 10% relative humidity, and given an F2 diet (Funahashi farm Co. Ltd. Japan) with water *ad libitum*. In order to ascertain the full-blown changes, the mutant mice had the ankle joints examined at 20 months of age. As the control, the ankle joints of B6C3F₁ mice of the same age were used. For

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Fig. 1. Macroscopical findings in the hair color in mice: left to right, mutant, CDF₁, B6C3F₁, C3H/He and C57BL/6 mice. The hair color of the mutant mouse is obviously lighter than that of the B6C3F₁ mouse, and the hair color is the same as that of the CDF₁ mouse.

radiological examination, a soft X-ray apparatus (Softex-ESM2 type; Softex Co. Ltd., Japan) was used. For histological examination, the specimens were fixed in buffered formalin, and decalcified with decalcifying fluid (K-CX; Faima Co. Ltd., Japan). Tissues were stained with hematoxylin and eosin (H-E), alcian blue, toluidin blue, safranin O stains and Kossa's method were added as needed.

The mutant mouse was born from a B6C3F₁ mouse (generation F₁₈). Macroscopically, characterizations of this mouse that show cinnamon (yellowish-brown) color of the hair (Fig. 1), and swelling and/or ataxia in the ankle joints were detected. Changes in the ankle joints developed as early as 9 months of age, and the change gradually worsened gradually after that. These abnormalities were characterized by tiptoe waking and motor paresis at about 20 months of age. On radiographic examination, the ankle joints showed fusion of the tarsal bones, bridging between the calcaneus and metatarsal bones, and ossification of the extra-articular soft tissues (Fig. 2). There was joint deformity, followed by progressive osteoarthritic changes and ankylosis of the ankle joints. As histopathological find-

Fig. 2. Radiographical findings in the hind limbs of the B6C3F₁ mouse (upper) and the mutant mouse (lower). Marked ossification of extra-articular soft tissue (small arrow) and further fusion of the tarsal bone (large arrow) is seen in the mutant mouse.

Fig. 3. Microscopical finding in the ankle joints in the mutant mouse. Destruction (large arrows) and erosion (small arrow) of the articular cartilage are seen in the cartilage surface. H-E stain. $\times 40$.

ings, osteoarthritic changes in the ankle joints were observed. As early changes, the surface of the cartilage was rough, and thinning, fissuring, and/or erosions were detectable in the cartilage layers (Fig. 3). The presence of marginal osteophytes was observed as cartilaginous projections from periosteal tissues subjacent to the synovial-articular cartilage junction. The marginal osteophyte formation was progressively larger, and extended well away from the epiphysis. Ankylotic changes occurred due to hyperplasia of osteophyte formation and regenerative cartilage, and fusion of the opposing bones. Therefore, many articular cavities had been bridged by cartilaginous hyperplastic tissues (Fig. 4). Subsequently, these bridges of cartilaginous hyperplastic tissues were largely ossified. Finally, the articular cavity and bone marrow space were completely occupied by newly formed cartilaginous and bone tissues (Fig. 5). Marked ossification of extra-articular soft tissues were positive for Kossa's method. In severely affected joints, synovial lining cells seemed to have been essentially eliminated, but no infiltration of inflammatory cells into synovial tissues was observed. About 87% of the male mice and 21% of the female mice were affected (Table 1). The mutant mice begin

at about 14 months of age, and most of them die at about 22 months of age. In contrast, these changes were not seen in all B6C3F₁ mice examined.

The present study failed to clarify details of the cause of the change in the color of hair and the mechanism of osteoarthritic changes. Precise evaluation of this animal model await further studies. Morphological findings reported here, however, indicate that the condition in this mutant mouse apparently resembles in human OA. As osteoarthritic changes, namely, thinning, fissuring and erosion of the articular cartilage, absorption of the subchondral bone, and cartilaginous osteophyte formation corresponded fairly well with changes seen human OA. Changes in the ankle joint simulated, moreover, at least in some aspects, ankylosing osteoarthritis, but lack of proliferation of the synovial lining cells or infiltration of inflammatory cells into the synovial tissues in this mouse appear to be unsatisfactory aspects in relation to human OA. Although the predilection sites of human OA are the knee and hip joints [5], the higher genesis of osteoarthritic lesions in the ankle joint is one of the characteristics of the mouse. Osteoarthritic lesions in the mouse were first reported by Silberberg and Silberberg [9], and have

Fig. 4. Microscopical finding in the ankle joint in the mutant mouse. Cartilaginous osteophyte in the articular lateral part almost bridged. H-E stain. $\times 40$.

Fig. 5. Microscopical finding in the ankle joint in the mutant mouse. An articular structure is grossly deformed and hardly recognizable because of hyperplasia and bone fusion. H-E stain. $\times 40$.

Table 1. Incidence of changes in hair color and ankle joint in mutant mice

Generation	Sex	No. of mice	Hair color		Abnormality of ankle joints* (%)
			Wild [#]	Cinnamon [†]	
1	M	13	0	13	11 (84.6)
	F	8	0	8	2 (25.0)
2	M	8	0	8	8 (100)
	F	11	0	11	3 (27.2)
3	M	21	2 [@]	19	17 (89.5)
	F	16	0	16	3 (18.8)
4	M	18	0	18	16 (88.9)
	F	17	0	17	3 (17.6)
5	M	16	0	16	14 (87.5)
	F	9	0	9	2 (22.2)

*Values for changes in ankle joints to use in histological examinations. [#]The color of the hair is the same as in the B6C3F₁ mouse. [†]Yellowish-brown color. [@]No abnormal changes of joints were demonstrated. M: Male F: Female

now been identified inbred in some strains of the mouse [4]. Many scientists examined these animal models have reported that the knee joints in particular exhibited osteoarthritic changes [2–4, 9]. In most of them, therefore, significant differences from the mutant mouse are noticed. On the one hand, Sokoloff and Habermann [11] and Walton [17] reported that osteoarthritic lesions of the mouse were also observed in the ankle joints. It has been known that the incidence, progress and affected joints with osteoarthritic changes in mice vary with different strains [10–13]. This fact shows a quite different incidence of osteoarthritic lesions in different strains of the mouse [10–13]. Therefore, genetic predisposition in this lesion cannot be denied [9, 18].

This mutant mouse had osteoarthritic lesions, with a special connection to aging and sexuality, and some histopathological characteristics resembled those of human OA. Although the pathogenesis of changes found in the mouse is not yet clarified, analysis of the morphology, immunology, heredity and chemistry of this strain are now under way in our laboratory. Hereafter, this mouse should be useful for studying the process and mechanism of osteoarthritic lesions and destructive or ankylotic osteoarthropathies.

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