Ultrasonogram of Hepatic Abscess in Cattle inoculated with *Fusobacterium necrophorum*

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**ABSTRACT.** *Fusobacterium necrophorum* was inoculated into the portal vein of eight cattle. Multiple necrotic foci were found in the livers of three of them which died acutely. The remaining cattle autopsied 12 to 126 days after the inoculation had abscesses in their livers. In the former three cattle, multiple hyperechoic masses were detected in the liver by ultrasonography. The masses corresponded to the necrotic foci. In the latter five cattle, a liver lesion was similarly detected, first on the 5th to 10th day after inoculation. It consisted of a central hyperechoic mass and a surrounding hypoechoic halo, which corresponded to necrotic tissue and granulation tissue, respectively. In some cases the central hyperechoic area of the halo pattern changed later to be hypoechoic. The ultrasonographic lesions appeared as a spherical hypoechoic pattern. Finally, they disappeared from the hepatic images on around the 50th to 70th day after inoculation. These processes corresponded to diminution of the central necrotic mass, the abscess being gradually replaced by the granulation tissue and capsule and to change of the abscess into the scar. The capsule and scar were imaged as a isoechoic or hypoechoic pattern. Some other lesions of the halo pattern remained and were accompanied by lateral shadows.—**KEY WORDS:** cattle, hepatic abscess, ultrasonogram.

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Cattle affected with hepatic abscesses show no specific clinical symptom. For possible clinical diagnosis of hepatic abscess in cattle, two methods may be available. One is clinico-chemical [6]. The other method is of medical imaging by means of an ultrasonograph, but clear hepatic images are not always obtained in this way (3.5 MHz) from the body surface of large animals such as cattle [2], and also these images do not cover the whole of the liver [12].

Ultrasoundographic observation was made, by Itabisashi [2], on the experimental bovine hepatic abscess induced by inoculation of *Fusobacterium necrophorum* into the portal vein, but in this study the character of the ultrasonographic image was not correlated in detail with the nature of the hepatic lesion. This relationship was examined more fully in the present study.

**MATERIALS AND METHODS**

**Animals:** Eight Holstein cattle 8 to 14 months of age were employed (Table 1). A *F. necrophorum* suspension was inoculated into the portal vein through a needle inserted by a percutaneous-transhepatic route.

**Bacterial suspension:** A *F. necrophorum* suspension was prepared by the method of Motoi *et al.* [7], which in turn was a modification of the method of Takeuchi *et al.* [13]. The well-mixed bacterial suspension was directly inoculated into the portal vein.

**Inoculation procedure:** Feed and water were not withheld from the cattle before surgical manipulation. The standing animal was sedated by an intravenous or intramuscular injection of 2% Celactal solution
(Bayer, West Germany). A small cutaneous area in the 10th or 11th intercostal space was locally anesthetized and incised 5mm just enough for insertion of an inoculation needle of 20cm in length and 1.6mm in outer diameter. The ultrasonic probe, not specially designed for needle biopsy, was improved very slightly as shown in Fig. 1. The small notches were made as A and B of Fig. 1 to locate the inoculation needle exactly in the ultrasonic beam. After setting the target, the intrahepatic portal vein, on a needle line previously drawn on the screen of ultrasonograph, the inoculation needle was inserted through the cutaneous opening and, guided by the notches, down into the portal vein as observed on the ultrasonograph. A polyethylene catheter was inserted through the needle into the vein. In some cases, the needle was withdrawn before bacterial inoculation leaving the catheter in the portal vein.

A bacterial suspension (Table 1) was quantitatively infused at a rate of 0.5ml/min through the catheter except in the case of cattle No. 1 in which it was given by rapid injection. After the catheter was flushed with saline, the catheter and needle were withdrawn one by one. Care was taken not to leave any solution or blood contained in the catheter and needle along the percutaneous-transhepatic route. Sound media for the ultrasonic guidance was sterile saline. Standard aseptic technique was employed during these procedures. No drugs were used except for sedation, local anesthesia and cutaneous disinfection.

**Ultrasonogram:** Hepatic images were obtained by means of an ultrasonograph (SSD-210DX-3.5MHz, Aloka, Tokyo) and recorded with a video recorder (VT-8, Hitachi, Tokyo). After examination on

![Image of ultrasonic probe used for percutaneous-transhepatic bacterial inoculation. A inoculation needle set in small notches made on the hard rubber block (A) and on the edge of the probe (B) could be inserted precisely along the sonic beam.]

**Table 1.** Experimental animals, bacterial inoculation and pathological lesions detected

<table>
<thead>
<tr>
<th>Cattle&lt;sup&gt;a&lt;/sup&gt;)</th>
<th>F. necrophorum inoculation</th>
<th>Days from inoculation to autopsy</th>
<th>Hepatic abscess</th>
<th>Extrahepatic abscess</th>
<th>Dermatitis at right breast</th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td>Sex &amp; Body Weight (kg)</td>
<td>Cfu/ml</td>
<td>Volume (ml)</td>
<td>Number</td>
<td>Size (cm)</td>
</tr>
<tr>
<td>1</td>
<td>Male 266</td>
<td>6.8x10&lt;sup&gt;9&lt;/sup&gt;</td>
<td>6.0</td>
<td>Hundreds</td>
<td>1–2</td>
</tr>
<tr>
<td>2</td>
<td>Male 250</td>
<td>5.2x10&lt;sup&gt;7&lt;/sup&gt;</td>
<td>5.0</td>
<td>Hundreds</td>
<td>1–2</td>
</tr>
<tr>
<td>3</td>
<td>Male 264</td>
<td>9.3x10&lt;sup&gt;6&lt;/sup&gt;</td>
<td>5.0</td>
<td>Hundreds</td>
<td>1–2</td>
</tr>
<tr>
<td>4</td>
<td>Male 296</td>
<td>1.8x10&lt;sup&gt;6&lt;/sup&gt;</td>
<td>5.0</td>
<td>7</td>
<td>2–5</td>
</tr>
<tr>
<td>5</td>
<td>Male 200</td>
<td>1.8x10&lt;sup&gt;6&lt;/sup&gt;</td>
<td>3.0</td>
<td>20</td>
<td>2–13</td>
</tr>
<tr>
<td>6</td>
<td>Male 184</td>
<td>3.4x10&lt;sup&gt;6&lt;/sup&gt;</td>
<td>1.5</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>7</td>
<td>Male 197</td>
<td>5.2x10&lt;sup&gt;7&lt;/sup&gt;</td>
<td>1.0</td>
<td>97</td>
<td>5</td>
</tr>
<tr>
<td>8</td>
<td>Male 232</td>
<td>5.2x10&lt;sup&gt;7&lt;/sup&gt;</td>
<td>4.0</td>
<td>126</td>
<td>2–5</td>
</tr>
</tbody>
</table>

<sup>a</sup>) Holstein breed.
them replayed on the screen of ultrasonograph, images were selected and printed (667, Polaroid, U. S. A.). The right body surface of animal was clipped for ultrasonic scanning [12]. Sound media for taking hepatic images was not saline but liquid paraffin, which was later carefully wiped off with a paper towel.

RESULTS

All the animals employed were first infused with 5ml of sterile saline or broth into the portal vein as a control. This made no change in ultrasonographic image of the liver.

Cattle Nos. 1, 2 and 3 had multiple necrotic lesions in their livers.

Cattle No. 1 died on the 2nd day after inoculation. A hepatic ultrasonogram of the 2nd day is shown in Fig. 2a. On the hepatic image echogenic masses marked N were seen. Many small necrotic foci of pale-yellow color and 3mm in diameter were scattered in much larger blackish-red lesions in the liver. Cattle No. 2 died on the 10th day after inoculation. Cattle No. 3 was recumbent when slaughtered on the 14th day. Echogenic lesions 1 to 2cm in diameter were imaged singly or in combination in their hepatic ultrasonograms (Fig. 2b and 2c). They began to be imaged dimly from the 6th day after inoculation. Multiple necrotic foci of pale-yellow or pink color and 1 to 2cm in diameter were found in their livers (Fig. 2b and 2c).

Extrahepatic abscesses were found in cattle Nos. 2 and 3 (Table 1).

Fig. 2. Hyperéchoic masses correspond to necrotic foci (N). In Figs. 2 to 5, photographs of gross hepatic lesions (upper) and ultrasonograms (lower) showing views of the caudal surface of the scanning section were printed in equal reduction (1 cm/div).
Hepatic ultrasonograms had been taken for 12 to 126 days after the inoculation in cattle Nos. 4 to 8. At autopsy up to 20 abscesses were seen in the individual animal (Table 1).

An ultrasonographic pattern consisting of a central hyperechoic mass with peripheral hypoechoic halo was first seen deeply on the 7th day after the bacterial inoculation. This will be called the halo pattern below and it is indicated with arrows in Fig. 3a to 3c, which were obtained from cattle No. 4 on the 12th day after inoculation. The ultrasonographic lesions of this animal were not detected on the 5th day. Ultrasonographic lesions of this type shown in Fig. 3a and 3b appeared singly and that in Fig. 3c had a combination of two central masses. The hyperechoic and hypoechoic regions corresponded to necrotic tissue and granulation tissue, respectively. No capsule had yet been formed. The necrotic tissue was surrounded by a very thin layer of dense pus, but this could not be identified differently from the surrounding tissues on the ultrasonogram. These young hepatic abscesses could be clearly detected as the halo pattern on ultrasonograms even when the abscess was only 1cm in diameter.

Observation of this halo pattern was made for a longer period in cattle No. 5. In this animal, ultrasonographic lesions of the halo pattern were visualized first on the 5th day after inoculation. Some of them were not seen on the 50th day, but some had been observed up to autopsy, being accompanied by lateral shadows (Fig. 4b). In the former case, the central hyperechoic mass of the halo pattern changed first to hypoechoic one which could not be differentiated from the hypoechoic halo. The lesion was, therefore, visualised as a spherical hypoechoic pattern (Fig. 4a and 4c). This finally disappeared as noted above.

A large abscess 13cm in diameter consisted of central necrotic masses, dense pus

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Fig. 3. Halo pattern consisting of central hyperechoic mass and peripheral hypoechoic halo. The debris had been removed from the lesion (b). For further explanation see the caption to Fig. 2 and the text.
and surrounding granulation tissue. The central necrotic mass and the peripheral pus and granulation tissue were imaged as hyperechoic and hypoechoic areas, respectively. A very small pus-spot surrounded by the granulation tissue and capsule (Fig. 4c) was also seen in the liver of cattle No. 5 at autopsy. Some of the encapsulated hepatic lesions appeared as ultrasonographic lesions with lateral shadows (Fig. 4b). Both scar and capsule were not always imaged differently from the surrounding hepatic parenchyma and therefore could not be detected on the screen. They gave a hypoechoic image in some cases (Fig. 4c).

Cattle No. 5 at autopsy had an abscess in the lung (Table 1). At autopsy, 56 to 126 days after inoculation, cattle Nos. 6, 7 and 8 had medium-sized abscesses about 5cm in diameter, as shown in Fig. 5. In addition to the ultrasonographic lesion corresponding to this medium-sized abscess, cattle No. 8 showed about 20 small ultrasonographic lesions of the halo pattern. They were detected first on the 6th day after inoculation and changed into hypoechoic lesions on the 50th day. They were not detected on the 70th day. Two of the three abscesses in this animal (Table 1) were not visualized on the screen throughout the observation, because they were under the lung.

In cattle No. 6, an ultrasonographic lesion was detected, first on the 7th day after inoculation as a halo pattern. On the 19th day an ultrasonographic lesion was found consisting of small central hyperechoic masses, a peripheral slightly hypoechoic region, lateral shadows and an enhanced posterior echo. It resembled that shown in Fig. 5a taken on the 56th day.

In cattle No. 7, an ultrasonographic lesion similar to that of cattle No. 6 without the enhanced posterior echo was detected first.

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Fig. 4. Changes of halo pattern. Plain hypoechoic lesions (a and c) and halo pattern with lateral shadows (b) are indicated with arrows. For further explanation see the caption of Fig. 2 and the text.
on the 5th day after inoculation. Lateral shadows developed around this lesion on the 9th day. The lesion enlarged from 2 to 3cm in diameter on the 34th day. On the 44th day, its diameter was 5cm, and it remained at this size until autopsy on the 97th day (Fig. 5b).

In cattle No. 8, a medium-sized ultrasonographic lesion was seen first on the 10th day after inoculation. The lesion gradually enlarged up to the 102nd day and then stabilised until autopsy on the 126th day (Fig. 5c). Lateral shadows appeared clearly on the 102nd day. During the ultrasonographic observation on this animal for 126 days, this lesion did not change to a cystic anechoic pattern. Small hyperechoic masses had been seen in hypoechoic region.

The abscesses of cattle Nos. 6, 7 and 8 shown in Fig. 5 were found at the autopsy to be composed of a central necrotic mass, dense pus, a thin layer of granulation tissue and a capsule. Scar surrounded the capsule in the cases illustrated in Fig. 5b and 5c. Not all of these components was well reflected in different echo characteristics on ultrasonograms. The necrotic masses and size of the spherical capsule were indicated as central hyperechoic masses and distance between the lateral shadows, respectively. The granulation tissue, dense pus and scar were imaged as a hypoechoic pattern.

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**Fig. 5.** Ultrasonographic lesions corresponding to chronic hepatic abscesses of medium size. For further explanation see the caption to Fig. 2 and the text.
Cattle No. 6 had an abscess in the intercostal space. For a period between the 25th to 40th day after inoculation, cattle Nos. 7 and 8 were suffering from intraepidermal vesiculopustular dermatitis on the right body surface (Table 1).

DISCUSSION

A case report is available on the ultrasonography of bovine hepatic abscess [15]. Itabisashi [2] later presented ultrasonographic images of clinical bovine hepatic abscess and cyst, both of which appeared as anechoic spherical patterns with an enhanced posterior echo.

The hepatic abscesses examined in the present paper were experimental rather than natural in origin. Ultrasonographic information collected from experimental abscesses serve just as a reference. Ultrasonographic images of field cases should, therefore, be collected chronologically to define the standard clinical picture of the bovine hepatic abscess. The collection of hepatic image can be made on a few animals in a herd selected in simple and easy ways such as determination of blood sialic acid [3, 6] or mucoprotein [6].

It was observed that echogenic masses similar to those seen in the cases of acute death of the present study (Fig. 2) fused into a large abscess, which retained its form for long periods without leading to death of the animal [2]. Whether the abscess develops in this way may depend on the severity of hepatic damage.

In cattle Nos. 4 to 8, ultrasonographic lesions, large or small, changed variously in texture. They were detected as the halo pattern first on the 5th to 10th day after inoculation. Some of them disappeared from the screen on the 50th to 70th day. Others were enlarged or did not change in size and were accompanied by lateral shadows or an enhanced posterior echo.

No anechoic cystic pattern with an enhanced posterior echo was visualized even in the most prolonged observation (126 days). Central necrotic tissue suspended in pus may need a still longer time to liquefy into pus, or some unknown regulating factors may exist.

Gross pathology on the experimental bovine hepatic abscess was made by Jensen et al. [4], Scanlan and Berg [11] and Nakajima et al. [9]. Multiple necrotic foci were seen in the lesions at an earlier stage. They later changed variously and were finally composed of pus with or without necrotic tissue, granulation tissue and capsule, from the center to the periphery. Some of them were transformed to scar tissue. These various components was not imaged on ultrasonograms as having different echo characteristics. But the lesion as a whole was easily detected by ultrasonography even if it was as small as 1cm in diameter.

The ultrasonographic images recorded in the previous and present studies may not be specific to the hepatic abscess [1, 10]. The ultrasonographic image of a hepatic abscess in a field case was the same as that given by a hepatic cyst as already noted above. We have no experience of how foci of hepatic teleangiectasis appear on an ultrasonogram. Ultrasonograms of the lesion caused by the liver fluke were examined by Suganuma et al. [12]. More information is needed on the ultrasonographic appearance of hepatic focal lesions to allow differential diagnosis.

An effect of the extrahepatic abscesses noted above on the acute phase reactants such as sialic acid in blood is known [3], but their effect on hepatic abscess formation is not clear. No difference in hepatic abscess formation was found between animals with or without extrahepatic abscesses and dermatitis. It may depend on severity of inflammation and host defense ability.

Multi-reflected echo appeared over the hepatic ultrasonograms in our previous
study of cattle inoculated with *F. necrophorum* by means of laparotomy [2]. For some days after inoculation, therefore, we could not read hepatic images due to this obstruction. The percutaneous-transhepatic inoculation by the ultrasonic guidance, a simple and easy technique [5, 14], did not cause such a multi-reflect echo at all.

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REFERENCES


要 約

*Fusobacterium necrophorum* 接種牛における肝臓癌の超音波所見：板広外茂雄・山本理恵子・佐藤真澄（農林水産省家畜衛生試験場）—— *Fusobacterium necrophorum* 培養菌を用いて12～16ケ月のHolstein牛8頭の肝内膿瘍に，超音波観察下穿刺法によって，経皮・経肝的に接種した。接種後2～14日に死滅し，肝に壊死膿瘍が発生していた。他の5頭では，接種後12～126日の剖検時に1～20個の肝臓癌がみられた。急性期死亡牛では，接種後2～6日に肝壊死膿瘍に対応する高エコー斑塊が認められた。肝臓癌形成牛では，接種後5～10日に肝画像に腫瘍所見がみられ，壊死塊に対応する高エコー斑塊を認めた。肉芽組織に対応する低エコー層を認めた。この超音波パターンは直径約1cmの小さい膿瘍でも確認でき，一部の例では，中心の高エコー層は消減し，肉芽組織と被膜（等または低エコー）のみの残存に対応する単純な低エコー弓となり，接種後50～100日では崩壊（等または低エコー）形成に対応して画像中に指摘できなくなった。エコーパターンのまま側方陰影を伴って長く残存する例もあり，低エコーパターンを示す膿痰は無エコーパターンとはならなかった。