

Research on the mechanism of thoracolumbar supernumerary rib development after birth using CT scanning



Makiko Kuwagata DVM PhD
Hatano Research Institute,
Food and Drug Safety Center, Japan



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Our Mission

To investigate the toxicological significance of thoracolumbar supernumerary ribs (TSR) **after birth.**

Thoracolumbar supernumerary ribs (TSR)

- Classified as a variation.
- Observe with relatively high incidence in a rodent study.
- Researchers' opinions split on the significance of TSR after birth.
- Little reliable data on TSR after birth.
- Difficult to distinguish chemically induced effects from spontaneous development based only on statistically significant results.
- Toxicological meaning is still debatable.

Historical control data on rat developmental toxicity test in Japan (2011-2015)

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WILEY 

ORIGINAL ARTICLE

Historical control data on developmental toxicity studies in rats

Makiko Kuwagata | Yuko Sakai | Sho Tanaka | Hiromasa Takashima | Ryuichi Katagiri |
Toshiki Matsuoka | Kenichi Noritake | Mika Senuma | Tatsuya Shimizu | Hitoshi Hojo |
Kanata Ibi | Satoshi Kudo | Takafumi Oota | Masayuki Ube | Yoji Miwa |
Shimpei Kajita | Tohru Uesugi | Kaoru Yabe | Taishi Tateishi | Nao Nakano |
Terumasa Taniguchi | Akihito Yamashita | Takayuki Hirano | Yuka Kirihata | Yumi Sakai |
Shino Nishizawa | Michio Fujiwara | Hiroshi Mineshima | Masao Horimoto | Makoto Ema

According to this survey, TSR(%) is observed at 0.07% to 12.98% in SD rats, and 4.89% to 58.10% in Wistar Hannover rats.

- Data collected from 24 Japanese laboratories, 15 pharmaceutical and chemical companies, and 9 contract research organizations.
- Sprague-Dawley (CrI:CD(SD)) and Wistar Hannover (RccHan:WIST and BrlHan:WIST@Jcl(GALAS)) were used.

Today's talk

- Using CT scanning, monitor TSR morphological changes after birth in the same animal.



5-FC induced TSR rat model

(postnatal observation)

Chemical : 5-flucytocine (5-FC)

Dose : 0, 35 or 75 mg/kg

Treatment : GD9 (orally)

No. dams : 20 dams per group

After delivery, offspring were culled to 8 offspring per litter (4 males and 4 females) on PND4.

CT scanning : PNDs 4, 14, 26, 35 (male), 42 (female), 53 (male), 61 (male) and 62 (female).

5-FC induced TSR rat model

(Cont.)

Developmental landmarks : BW, FC, onset of sexual maturation

Organ weights and histopathology at autopsy at terminal point : liver, spleen, kidneys, adrenal glands, testes, epididymides, ovaries, uterus

Autopsy at Terminal point : PNDs 61-63

Skeletal morphology of all offspring will be observed by double skeletal staining.

Results

- **Dams:** No adverse effects on BW, delivery index, number of pups alive, and nursing.
- **Offspring:** No adverse effects on viability, BW, FC, onset of sexual maturity, OWs.

Postnatal TSR observation by CT scanning

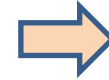
Monitor TSR during postnatal development in the same animal.



3D micro X-ray computed tomography (CT) for laboratory animals
CosmoScan GXII (RIGAKU, Japan)



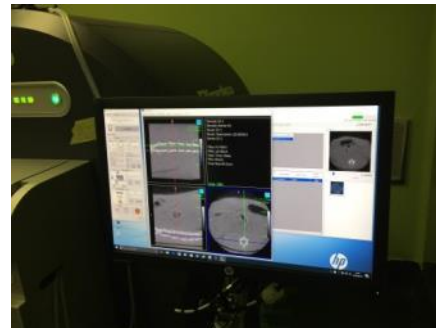
1. Anesthesia



2. Set animal



3. Scanning

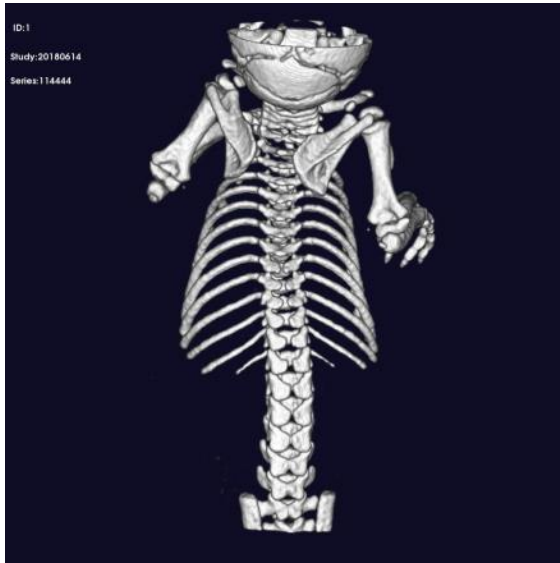


4. Analysis

Postnatal TSR observation by CT scanning

Analysis

1. 3D picture



Monitor rib morphology
(types of TSR; rudimentary, short, full)

2. MIP (maximum intensity projection) picture



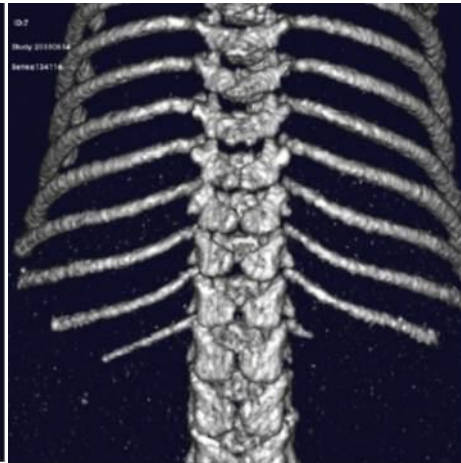
Measurement the length of ribs
(ratio of 14th rib to 13th rib)

Postnatal TSR observation by CT scanning

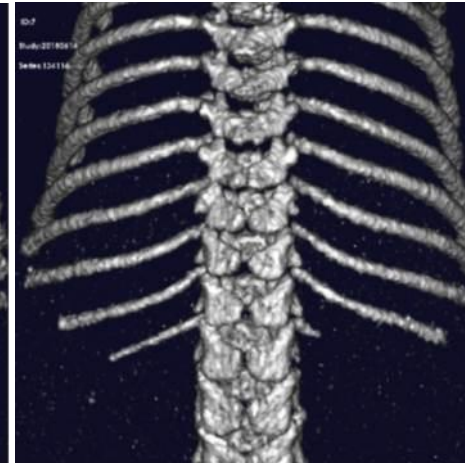
1. 3D picture



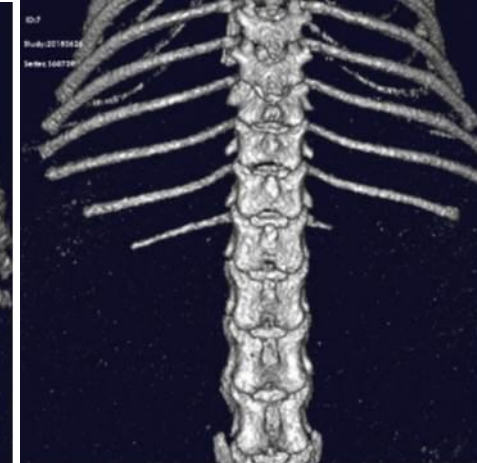
PND4



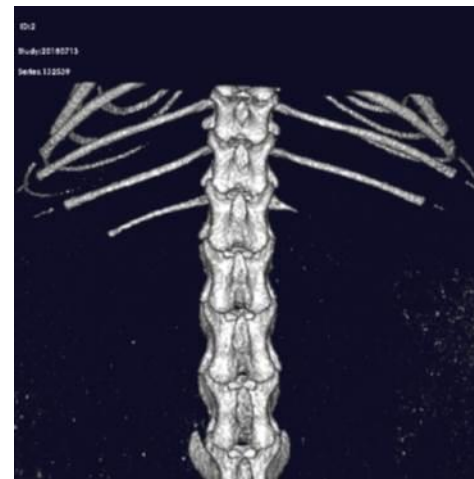
PND14



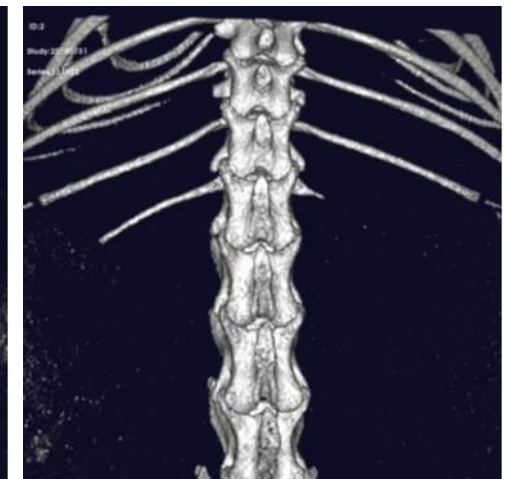
PND26



PND35 (pre-puberty)



PND43 (after-puberty)

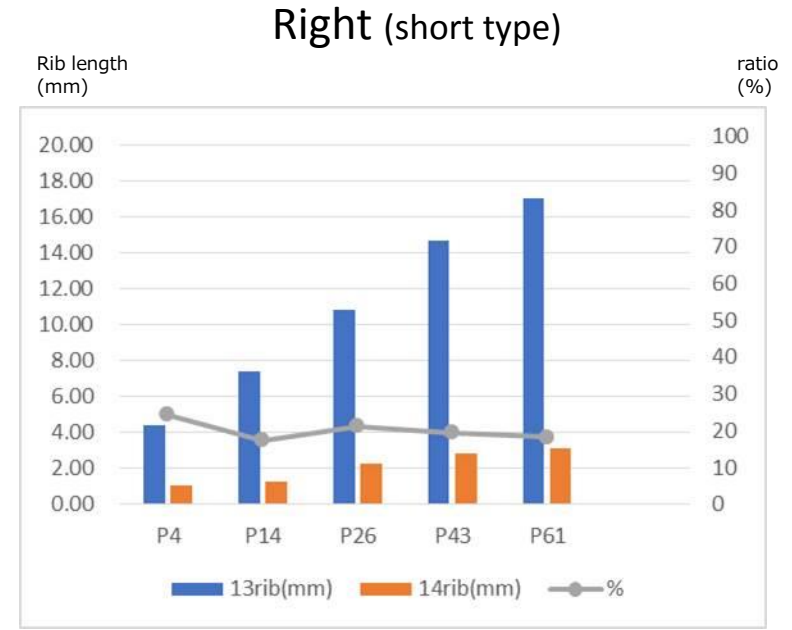
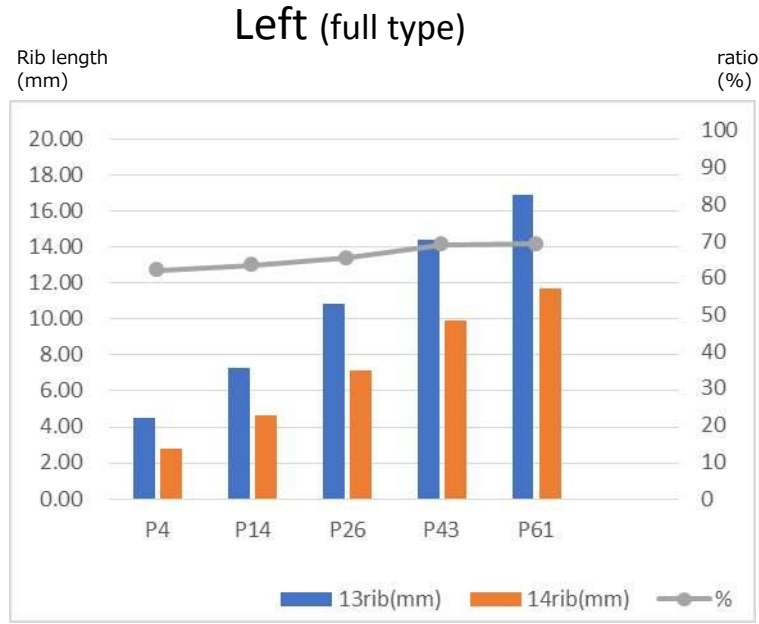


PND61

Animal no.5FC-H2, F7
Left side: full type
Right side: short type

Postnatal TSR observation by CT scanning

2. MIP picture



- Measure length of the 13th and 14th ribs (mm)
- Calculation of the ratio of 14th rib to 13th rib (%)



Animal no.5FC-H2, F7
Left side: full type
Right side: short type

TSR develop within the normal range, but do not exceed the normal range after birth.

- Sexual maturation did not affect the features of TSR.

Ongoing project

- Using CT scanning, monitor TSR morphological changes after birth in the same animal. (Today's talk)
- Characteristics of 5-FC-induced TSR (The critical window is narrow and earlier than the ordinary administration time : *under submission*).
- Researching the mechanism of TSR induced by 5-FC (Contribution on HOX10 gene : *in preparation*).

Final goal

Determine the toxicological significance of TSR in ReproTox studies.

A Goal to reach for...

The results obtained hint at the significance of TSR in reproductive and developmental toxicity studies.

Now, proceeding with the analysis.



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