

## **A novel technique for cell block using glucomannan extracted from *Amorphallus konjac***

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Cell block preparation is applied for cytological diagnosis of samples with small amounts of cell constituents, such as urinary samples, sputum, body cavity fluids as well as aspirated materials. We evaluated a cell block preparation using glucomannan which was extracted from *Amorphallus konjac*.

## **Materials and Methods**

The material was ascites of a patient with definitively diagnosed clear cell carcinoma of the ovary.

### **Method I: Treatment of the cell sample**

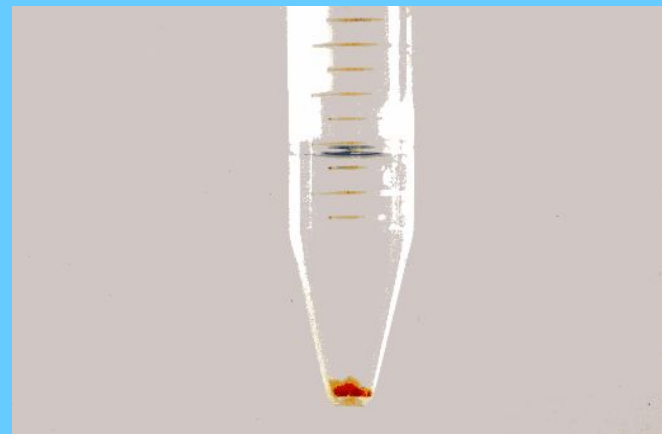
(1) Ascites was centrifuged at 1,500 rpm for 5 minutes, the supernatant was removed, the remnant after the preparation of smear specimens for routine cytological examination was added with 20% formalin, and fixed at room temperature for 1-12 hours.

(2) The fixed ascites was centrifuged at 1,500 rpm for 5 minutes, and the supernatant was removed.

(3) The precipitate was resuspended with 2 ml of eosin solution.



(4) The suspension was mixed with 1-5 ml of 80% alcohol, centrifuged at 1,500 rpm for 5 minutes, and the supernatant was removed.

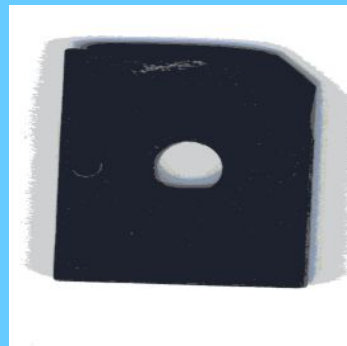


(5) To the precipitate, 1 drop of a glucomannan-formalin solution (Holdgel 110; Asia Kizai Co., Ltd.) was added gently

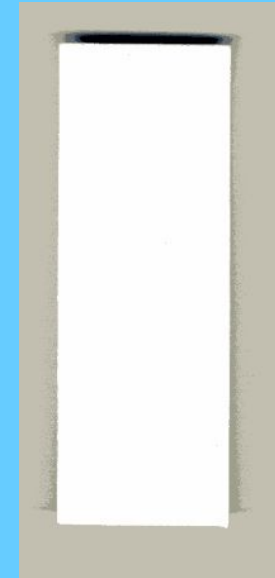


**Method II: A cassette for the preparation of cell blocks**

(6) The glucomannan-formalin solution was applied around the cylindrical hole at the bottom of a frame for the preparation of cell blocks (Asia Kizai Co., Ltd.).



(7) Pieces of filter paper trimmed to 6.5 x 2.3 cm were applied to the upper and lower surfaces of the above cell block frame, and the frame was placed in a cassette for the preparation of tissue specimens (System Cassette G, Asia Kizai Co., Ltd.).



Method III : Preparation of a cell block

(8) The precipitate in (5) was aspirated with a syringe and placed in the hole prepared as in (7).

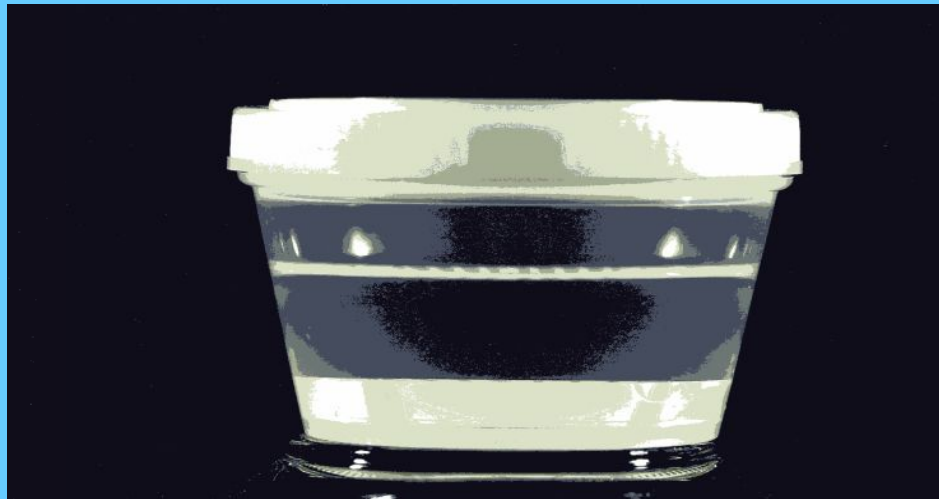


(9) The glucomannan-formalin solution was added gently until the hole was completely filled.



(10) The hole was covered with filter paper, and the lid of the cassette for the preparation of tissue specimens was applied over the filter paper.

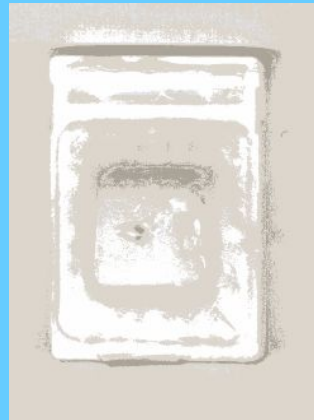
(11) After immersion in methanol for 3 hours, glucomannan is solidified and becomes gelatinous.

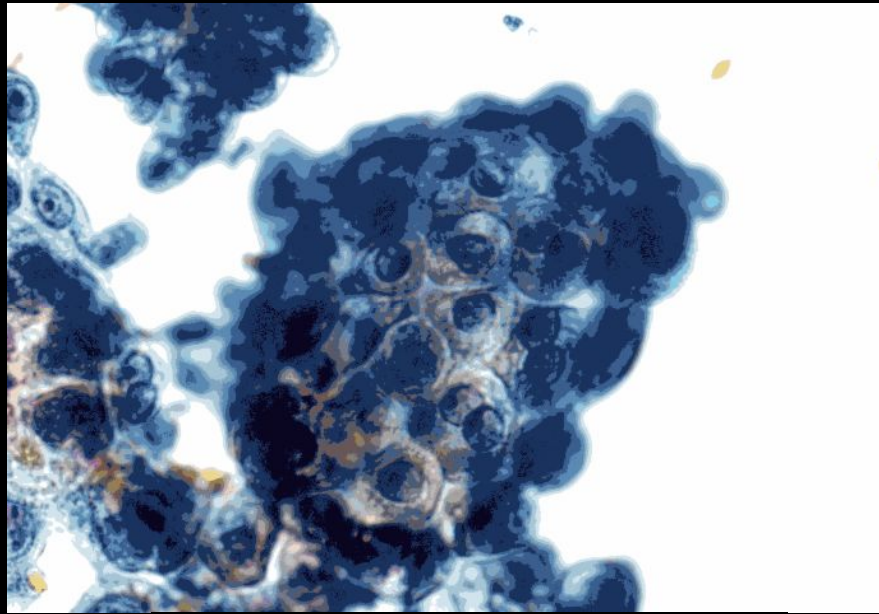




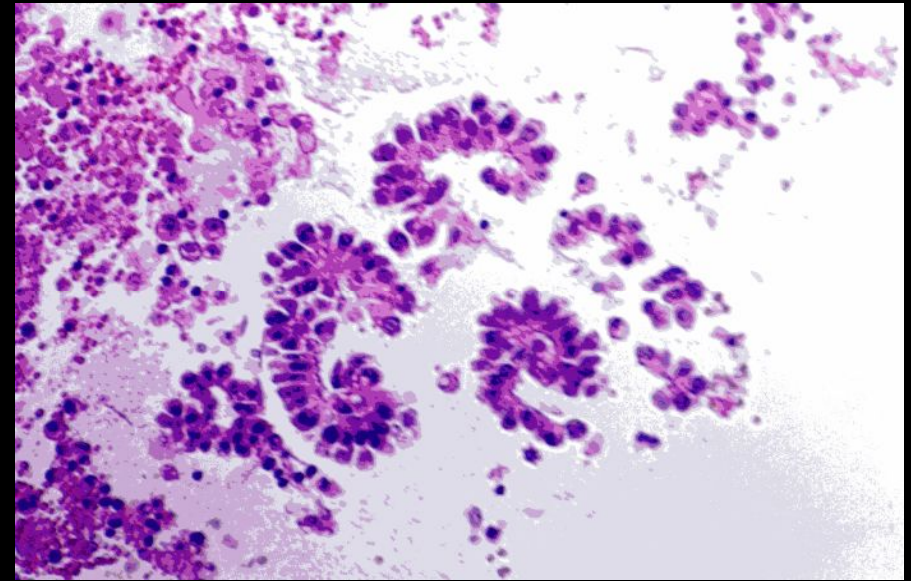
(12) Columnar gel containing the cell sample (cell block) was removed from the hole, and the filter paper was detached.

(13) The cell block was placed in the cassette for the preparation of tissue specimens, dehydrated by the routine method, infiltrated with paraffin, and a paraffin-embedded block was prepared.

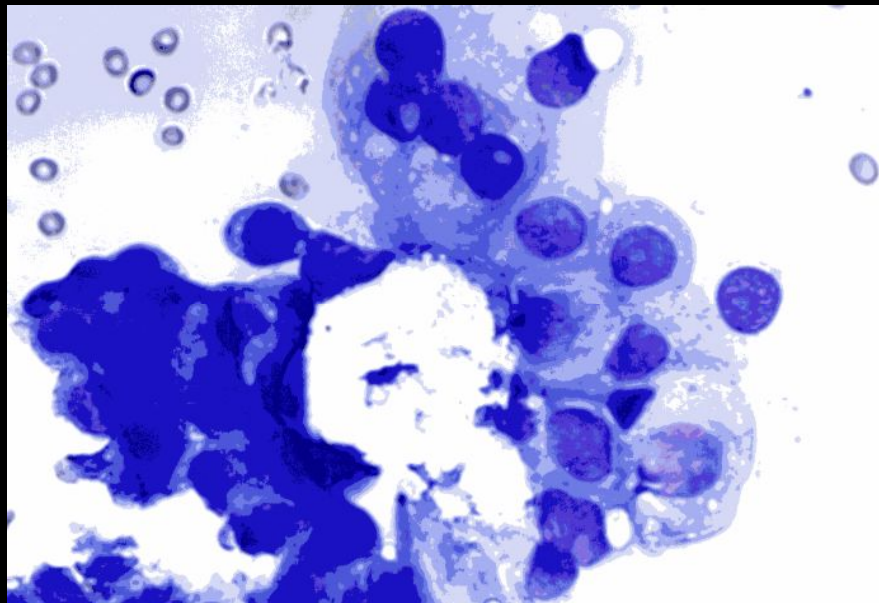




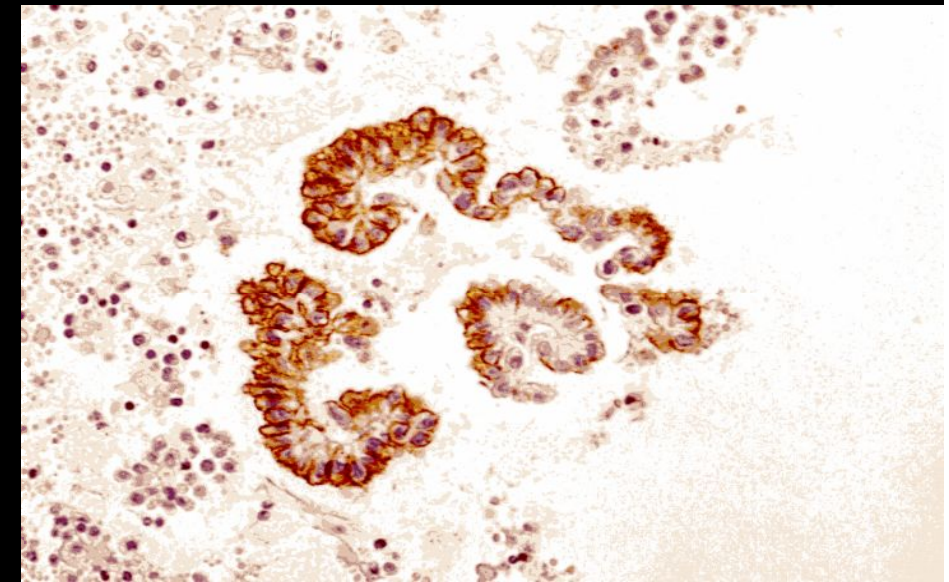
Papnicolaou stain x 40



Hematoxylin & Eosin stain x 20



Giemsa stain x 40



Immunocytochemical stain x 40

## **Results**

On Papanicolaou staining, tumor cells appeared as spherical or papillary aggregates, nuclei were elliptical, chromatin was granular, and nucleoli were clear. The findings on Giemsa staining were similar. Through H&E staining, tumor cells showed a ductal arrangement. On immuno staining, the ovary tumor specific marker CA-125 was positive in the cell membrane.

## **Discussion**

Thin sections were prepared from the paraffin-embedded cell block, and H&E and immuno stain were performed. In this study, we have developed the cell block method using a glucomannan-formalin solution. This method is easy to perform and is considered to be useful as an alternative technique for cytological examination.

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Cell block preparation is applied for cytological diagnosis of samples with small amounts of constituents, such as urinary samples, sputum, body cavity fluids as well as aspirated materials. We evaluated a cell block preparation using glucomannan which was extracted from *Amorphallus konjac*.

**Materials and Methods :** Cellular material was taken from ascites of a patient with definitive diagnosed clear cell carcinoma of the ovary. The specimen was centrifuged at 1,500 rpm for 5 min and the supernatant was removed, remnant after the preparation of smear specimens for routine cytological examination was fixed with 20% formalin. The specimen was centrifuged at 1,500 rpm for 5 min and the supernatant was removed. The residue was resuspended with 2 ml of eosin solution and fixed with 80% alcohol, stirred well. Next, after centrifuged the supernatant was removed, 1 drop of a glucomannan-formalin water solution (Asia kizai Co., Ltd.) was added gently. After immersion with methanol for 3 hours, glucomannan is solidified and becomes gelatinous. Cell block was placed in the cassette for the preparation of tissue specimens, dehydrated by the routine method, infiltrated with paraffin, and a paraffin-embedded block was prepared.

**Results and Discussion :** Thin sections were prepared from the paraffin-embedded cell block, and immunohistochemical stains were performed. We evaluated the cell block method using a glucomannan-formalin water solution, which is less well known. However, the method is easy to perform and considered to be useful as an alternative technique for cell block preparations.