Ministry of the Environment Project to Promote Monitoring of Important Ecosystem Surveillance Areas (Monitoring Site 1000)

Monitoring Site 1000: Forest and Grassland Surveys

# Manual on Cellulose Filter Decomposition Testing

Revised in July 2011

Nature Conservation Bureau
Ministry of the Environment
Biodiversity Center of Japan

Japan Wildlife Research Center

This manual is an English translation of the Japanese version and has not been reviewed by experts. Some technical terms and expressions may not exactly capture the intended nuances.

# Introduction

The forest ecosystem is highly appreciated because of its contribution to curbing global warming as a principal source of absorption of carbon dioxide in the Kyoto Protocol adopted in 1997. Forest trees absorb carbon dioxide ($CO_2$) and make leaves using carbon from it. Although some leaves are eaten by insects, many of them wither away and are supplied to forest floors. These fallen leaves are decomposed by animals living in soil such as microorganisms, earthworms, and roly-poly bugs. In the process of decomposition, carbon dioxide is released into the atmosphere. Therefore, in the forestry ecosystem, carbon dioxide is absorbed and released at the same time.

This means that it becomes possible to figure out the volume of carbon dioxide maintained in the forestry ecosystem by capturing the volume of leaves produced and fallen onto forest floors as fallen leaves and decomposed, and the rate of decomposition process.

The volume and rate of the decomposition mechanism of forests differs greatly from one place to another. This is likely to be attributable to differences in temperatures, soil conditions, and different types of organisms that facilitate decomposition. Thus, it is possible to figure out what kind of changes are occurring based on environmental changes such as global warming by collecting long-term data. Moreover, data collected from each area is of help for environmental predictions.

To this end, a decomposition test will be carried out so that the core sites nationwide can uniformly measure the decomposition process of organic matter on forest floors. A difference in tree species causes changes in the hardness of fallen leaves and their components, affecting the progress of decomposition. Therefore, cellulose filters are used to unify the testing conditions nationwide, because cellulose is the major component of leaves. A survey is conducted at litter layers where fallen leaves are accumulated and decomposed actively and soil layers directly under.

## Survey method

The Monitoring Site 1000 Forest and Grassland Survey Network Center (A department that handles samples and data of the Monitoring Site 1000 Forest and Grassland Survey (excluding bird species) established in the Hokkaido University Tomakomai Experimental Forest, hereinafter referred to as "the Network Center") sends cellulose filters which are labeled and weighted to each site. Each site buries these filters in soil, digs them out after a pre-determined time passes, and sends them back to the person in charge of the Network Center. The person in charge measures the weight of filters to calculate the reduction in weight during the period when filters were buried in soil. A decomposition rate of organic matters is calculated based on the weight of filters before they were buried and the reduction in volume while they were buried.

This survey is conducted every three years and cellulose filters are buried twice and collected four times a year. The procedures for each work are shown below.

### 1. Burying filters

#### 1.1. Period

- First installation (Filters whose collection date is A~C): When the first pitfall trap survey is conducted
- Second installation (Filters whose collection date is D): When the fourth pitfall trap survey is conducted

#### 1.2. Necessary tools

[Tools sent by the Network Center]

- Cellulose filter (Benchkote 2300-916 (Whatman), 5 x 5 cm, 80 filters per survey site)
- Wire
- Metallic mesh (15 per survey site)

[Tools prepared by each site]

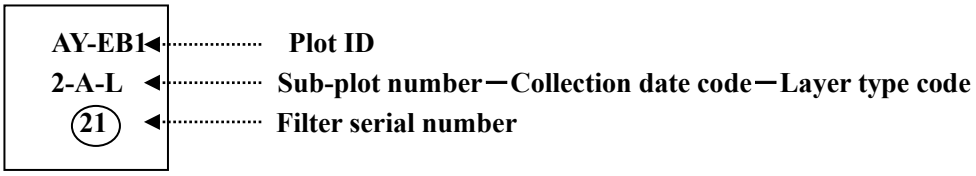
- Hoe (or shovel)
- Indicator tape
- Oil-based pen

### 1.3. Advance preparations

(1) A filter has a cellulose face (paper face) and a resin face (vinyl face).

**The plot (survey site) ID, sub-plot number (1~5), collection date code (A~D), layer type code (L: Litter layer, S Soil layer), and filter serial number (1~80) are written down on the resin face in advance with an oil-based pen**.

Example)




(2) Sort filters into 20 sets (Fig. 1). Each set consists of four filters (Filter type code L: Two filters, S: Two filters) whose sub-plot number (1~5) and collection date code (A~D) are the same. Filters whose collection date code is A~C are buried at the first installation and those whose collection data code is D are buried at the second installation.

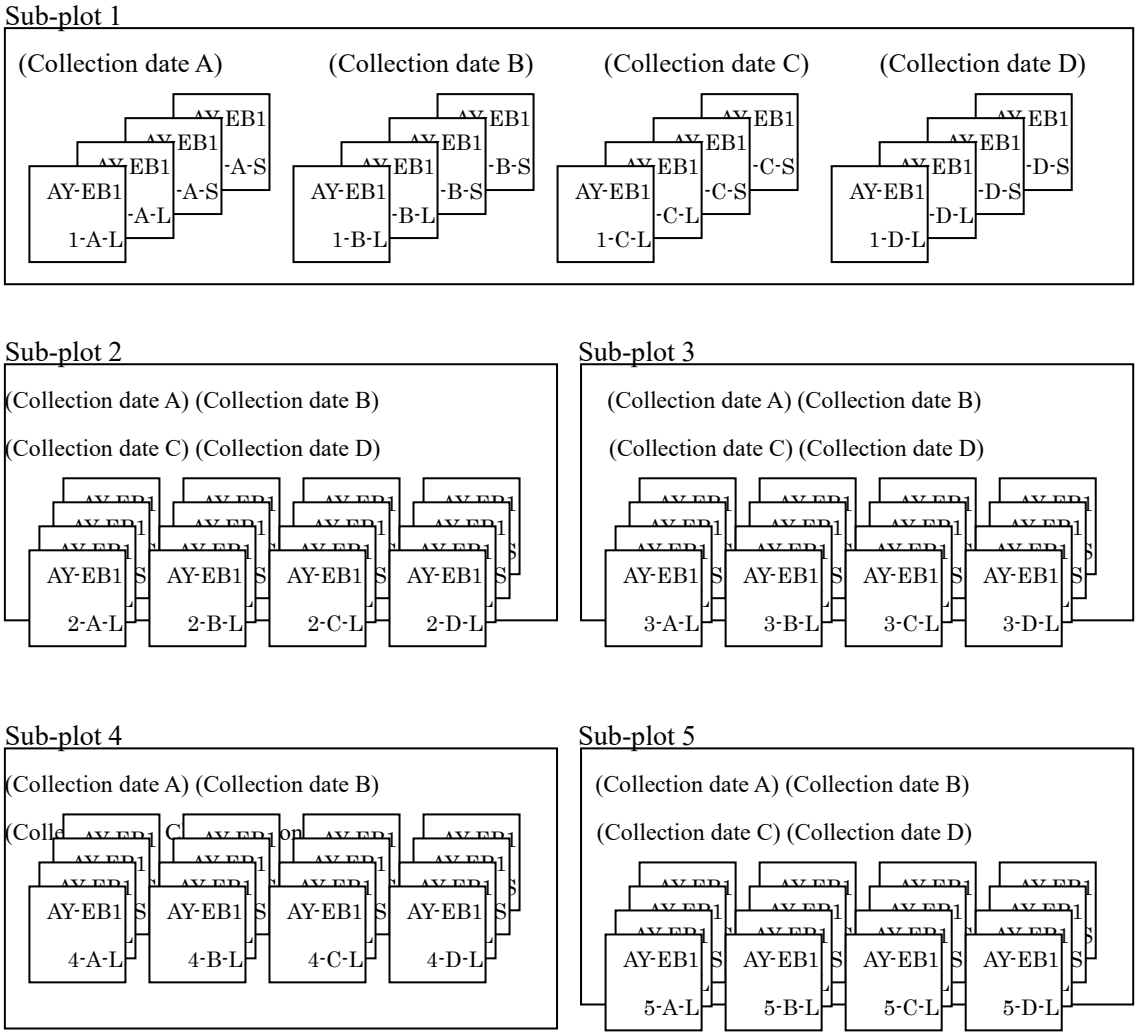



Fig. 1 How to sort filters: Four filters whose sub-plot and collection date are the same form a pair. 80 filters are divided into 20 pairs.

## 1.4. Outdoor works

When filters are buried twice a year, the following works are carried out at five sub-plots (a range of 5 m x 5 m including four pitfall traps) in the survey site (Fig. 2).

(1) Select a flat forest floor where soil is stable and less disturbed and trampled artificially as a site where filters are buried.

(2) **Install four filters whose collection date is the same** (three pairs (A, B, C) at the first installation and one pair (D) at the second installation, Fig. 1) **in a range of 15 cm x 15 cm**. Then, **place litter layers on those four filters installed and place metallic meshes on top of that**. Follow the following procedures for installation of filters in accordance with layer type code.

(3) Filters whose layer type code is S (Soil) are used to measure a decomposition late of soil layers, particularly, the volume of decomposition by microorganisms in soil. Remove litter on the forest floor surface when filters are buried to expose dirt. **Make vertical cuts at the depth of about 5 cm** using a hoe, etc. **Insert a filter in a gap of cuts made**. At this time, make sure to insert a filter so that its upper end comes to the same height as the soil surface so that the filter does not protrude from the soil surface. After the filter is inserted, **compress soil with both hands from the two sides of the cuts** to fill the gap between soil and the filter. If a width of the filters is kept narrow as much as possible (about a width allowing a filter to be inserted), it becomes easier to fill the gap.

(4) Filters whose layer type code is L (Litter) are used to measure a decomposition rate of litter layers, particularly, the volume of decomposition by microorganisms that use litter layers. Remove litter on the forest floor surface near the cut in which the filter (S) for soil layer is inserted to expose dirt. **Place the filter horizontally on the exposed dirt with the resin face on which the code is written down (vinyl face: undecomposable face)**. However, be careful not to cover the cut in which the filter for soil layer is inserted. After checking that the cellulose face comes to the upper surface (paper face: composable part), **cover the filter with litter removed at the beginning**.

*(5)* **Bend four wires in the U shape and pierce vertically into the four corners of the metallic mesh**. Here, make sure to pierce the wires into a stable part of soil so that the metallic mesh is fixed. Although the purpose is to prevent litter and filters from being flown from forest floors using wires and metallic meshes, this method may be insufficient in some sites. Thus, consult the person in charge for measures against disturbance by mammals.

(6) **Indicate the collection date (A, B, C) on the metallic meshes with indicator tapes, etc.** to be able to distinguish at the time of collection.

(7) Enter survey records in the “all survey records” sheet of a survey sheet (Excel file).

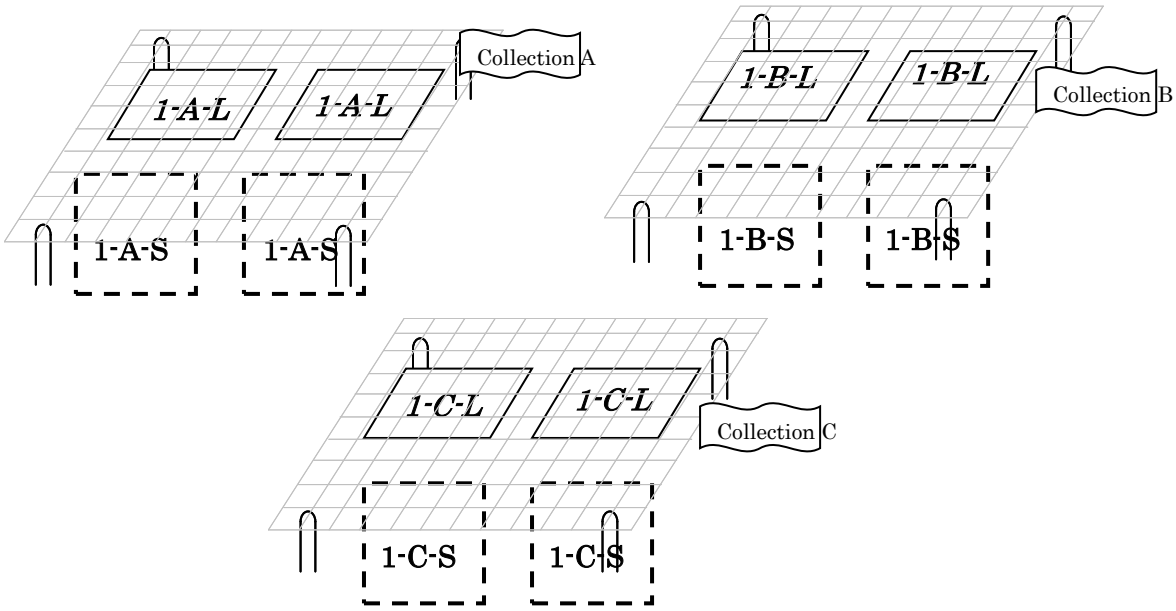



Fig.2 Pattern diagram of forest floors of sub-plot 1 with the filters buried (1st installation). In reality, the filters for litter layers (L) are installed with the resin face (the face on which the code is written) down.

## 2. Collection of filters

### 2.1. Period

- Collection of filters buried at the first installation (Collection date A~C): When the 2nd, 3rd, and 4th pitfall trap surveys are conducted of the relevant year.
- Collection of filters buried at the second installation (Collection date D): When the 1st pitfall trap survey of the following year is conducted.

### 2.2. Necessary tools

【Tools each site should prepare】

- Plastic bags for collecting and sending contents
- Drying trays

### 2.3. Outdoor works

(1) **When the second pitfall trap survey is conducted, collect the filters whose collection date is A (four filters under the metallic mesh A)**. They are collected from five sub-plots, so **a total of 20 filters are collected each time**. The metallic meshes and wires should be collected and stored by each site.

Likewise, collect

- the filters whose collection date is B at the third pitfall trap survey,
- the filters whose collection date is C at the fourth pitfall trap survey, and
- the filters whose collection date is D at the first pitfall trap survey of the following year.

(2) Enter survey records in the "all survey records" sheet of a survey sheet (Excel file).

(3) **Remove soil and litter attached to the surface of the collected filters by lightly rubbing in water so that they are not damaged**.

(4) Immediately **dry the cleaned filters at 60 degrees C for around 24 hours** using a fan dryer. At the time of drying the filters, **extend them with the resin face down avoiding overlapping** to prevent the cellulose face from touching the tray or another filter. Make sure not to leave for a long time under wet condition.

(5) **Put 20 dried filters together in a plastic bag** and make sure to write **the survey site name and the dates on which the filters were buried and collected** on that bag.

(6) The dried filters put in the plastic bag should be sent to the Network Center by mail. It is not necessary to send filters by mail immediately as long as they are dried. Thus, it is allowed to send filters collected at different times all at once.

* In cases where it is difficult to dry filters because there is no fan dryer at the site, please consult the person in charge of the Network Center. Send the filters immediately after their collection by refrigerated transportation service and send email indicating that they have been sent.

## 3. Timing of survey and working schedule

The timing of burying and collecting filters is shown below.

| | Relevant year | | | | Following year |
|---|---|---|---|---|---|
| | Late April ~ Mid-June | Mid-June ~ Early July | Early September ~ Early October | Early October ~ Late November | Late April ~ Mid-June |
| Pitfall traps survey | First | Second | Third | Fourth | First |
| Burying filters | First (A-C) | | | Second (D) | |
| Collecting filters | | A | B | C | D |

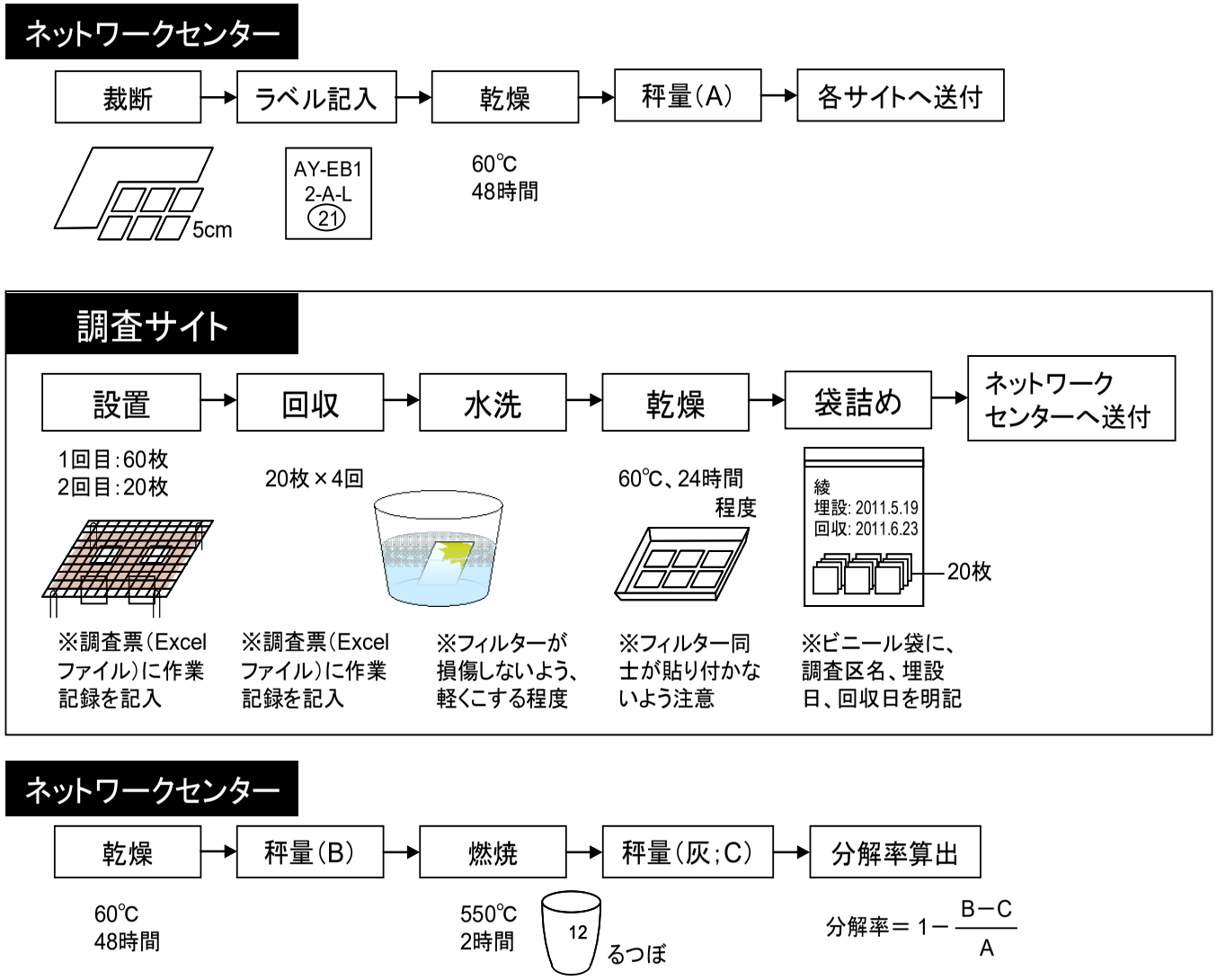



Fig. 3 Workflow

図内用語

ネットワークセンター：Network Center

裁断：Cutting

ラベル記入：Filling in label

乾燥：Drying

秤量：Weighing

各サイトへ送付：Sent to each site

48 時間：48 hours

60° ：60 degrees C

調査サイト：Survey site

設置：Installation

回収：Collection

水洗：Water washing

乾燥：Drying

袋詰め：Packing

ネットワークセンターへ送付：Sent to the Network Center

1 回目：60 枚：1st installation: 60 filters

2 回目：20 枚：2nd installation: 20 filters

20 枚 x4 回：20 pieces x 4 times

24 時間程度：About 24 hours

埋設：Buried on

回収：Collected on

20 枚：20 filters

*調査票（Excel ファイル）に作業記録を記入：Enter work records in a survey sheet (Excel)

*フィルターが損傷しないよう、軽くこする程度：Rub gently to prevent filters from being damaged

*フィルター同士が張り付かないように注意：Be careful to prevent filters from being attached each other

*ビニール袋に、調査区名、埋設日、回収日を明記：Write clearly on the plastic bag the name of survey site and the dates on which filters were buried and collected

ネットワークセンター：Network Center

乾燥：Drying

秤量：Weighing

燃焼：Burning

秤量（灰；C）：Weighing (Ash; C)

分解率算出：Calculation of decomposition rate

分解率：Decomposition rate

48 時間：48 hours

2 時間：2 hours

るつぼ：Melting pot

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