## **Network Analysis**

An abstract from "Network analysis for FAO Project Managers and Project Personnel" by the Management Services Division and Management Information Systems Unit of the FAO, Rome, 1971.

Compiled by Ariena H.C. van Bruggen.

Current information about Network Analysis:

http://www.netmba.com/operations/project/cpm/

Or http://www.management-hub.com/project-management-planning.html

| Activity          | An activity connects two events and is represented by an arrow. An activity has a                     |
|-------------------|---|
| 7                 | duration, it consumes resources and someone is responsible for starting the activity and              |
|                   | for reporting on its progress.  |
| <u>Event</u>      | An event happens instantaneously, it has no duration, it consumes no resources and                    |
|                   | nobody is responsible for it. It is the result of an activity or more activities. It is               |
|                   | represented by a <u>circle</u> with a number. The number must be unique.                              |
|                   | An activity can be described by the numbers of the preceding and succeeding events.                   |
|                   | In a network, there cannot be two similar activities, described by the same numbers.                  |
| <u>Start</u> and  | In each network, there are two special events: <u>Start</u> and <u>Finish</u> events. The Start event |
| <u>Finish</u>     | does not depend on any activity, and the starting activities do not depend on the                     |
| S                 | completion of any other activities. The Start event can take place at an arbitrary time,              |
|                   | which you can determine. Starting activities do not necessarily start at the same time.               |
| Logical           | The third symbol in the network is the dotted arrow, which represents a logical                       |
| <u>constraint</u> | constraint, usually called a dummy. It means that one event must occur before another                 |
| -7                | event can occur. Dummies don't consume time and you can insert them whenever you                      |
|                   | need them.  |



D depends on A and B, E depends on B and C.

Wrong! Because B is described twice.

Wrong! Because in this way, E depends also on

A, and D depends on C

Using dummies is the correct way.

| <u>Logic</u>         | In a network there are two kinds of logic: <u>strict logic</u> and <u>loose logic</u> . <u>Strict logic</u> is<br>imposed on us by nature: a certain activity <u>can only start</u> when other activities are<br>completed. With loose logic an activity can start as described in the network, but<br>alternatives are possible. The choice is up to the planner and is determined by<br>common sense. Loose logic is a function of experience, bias, culture and philosophy<br>of the planner.                     |
|----------------------|--|
| <u>Time analysis</u> | The time needed for each activity must be estimated and indicated along the arrows representing the activities. Based upon the logical constraints, the Earliest Event Time (EET) and Latest Event Time (LET) can be calculated. The EET = the earliest time an event can take place, assuming that all activities prior to it are carried out at their earliest times. It is determined by the longest path from the start event to the event concerned and indicated in the left-hand portion of the event circle. |
|                      | The LET = the latest time an event can take place if the project is to be completed by the specified completion time. It is determined by the longest path from the finish event to the event concerned and indicated in the right-hand portion of the event circle.   |

## Example 2.



| Event Slack      | Event slack = LET – EET   |
|------------------|---|
|                  | An event with no event slack is called a critical event. A critical event has no elbow    |
|                  | room; it must not occur late, or else the project will be delayed.                        |
| <u>EST - LST</u> | The earliest start time of an activity (EST) has the same value as the EET of its         |
|                  | preceding event. Often an activity can start some time after its preceding event          |
|                  | occurs.   |
|                  | The latest time the activity can start without delaying the project is its Latest Start   |
|                  | Time (LST). LST = LET of the succeeding event – duration of activity.                     |
|                  | e.g. (see example 2) LST <sub>H</sub> = 26, not 20.                                       |
|                  | An activity is critical if its earliest start time equals its latest start time. Critical |
|                  | activities have no 'elbow room' ; if they are late, the project will be delayed.          |
|                  | The critical activities in example 2 are: B, G, L and J.                                  |

|                 | The critical activities join to form a critical path from the start event to the finish  |  |  |  |
|-----------------|--|--|--|--|
|                 | event; this path includes dummy 060-070 and it does not contain H. even though H   |  |  |  |
|                 | ioins the two critical events 060 and 090. There must be at least one critical nath in a   |  |  |  |
|                 | network. The critical path between the start and finish events has no gaps.  |  |  |  |
| Float           | The 'elhow room' in the start time of an activity is called float. There are two types of  |  |  |  |
| <u>110at</u>    | float called Free float (early) and Total Float  |  |  |  |
|                 | $\frac{1}{1} \frac{1}{1} \frac{1}$ |  |  |  |
|                 | Free float (early) = (EEF of s) – (EEF of p) – $a$ .   |  |  |  |
|                 | S = succeeding event   |  |  |  |
|                 | $P = preceding event \ = duration of activity.$  |  |  |  |
|                 | Free float (early) is the maximum period the start of the activity can be delayed from   |  |  |  |
|                 | its earliest start time without affecting the earliest start time of any immediately   |  |  |  |
|                 | following activity.  |  |  |  |
|                 | Total float of activity = (LST of act.) – EST of act.).  |  |  |  |
|                 | Total float is the maximum period by which the start of an activity can be delayed   |  |  |  |
|                 | from its earliest start time without affecting the project duration.   |  |  |  |
|                 |  |  |  |  |
| Updating        | The network must be updated as soon as the information it contains is no longer  |  |  |  |
|                 | accurate enough for the purposes for which the network is used. Once the network   |  |  |  |
|                 | no longer reflects the actual situation, the effectiveness of control will suffer, since   |  |  |  |
|                 | the information necessary for control will be inadequate or even wrong.  |  |  |  |
|                 | The updating involves the following aspects:   |  |  |  |
|                 | 1. Marking those activities which have been completed  |  |  |  |
|                 | 2. Estimating the time to completion for activities currently being carried out  |  |  |  |
|                 | 3. Revising any duration estimates for future activities   |  |  |  |
|                 | 4 Revising the logic where necessary including adding or deleting activities   |  |  |  |
|                 | 5 Recalculating the event times to find the minimum project duration and the   |  |  |  |
|                 | critical nath  |  |  |  |
| Computor        | There are computer programs where you can onter:   |  |  |  |
| <u>computer</u> | Activity name or description   |  |  |  |
|                 | - Activity hame of description   |  |  |  |
|                 | - Preceding event number   |  |  |  |
|                 | - Succeeding event number  |  |  |  |
|                 | - Time estimate for the activity duration  |  |  |  |
|                 | - Code for the person responsible for the activity.  |  |  |  |
|                 | See: <u>http://www.netmba.com/operations/project/cpm/</u>  |  |  |  |
|                 | Or <a href="http://www.management-hub.com/project-management-planning.html">http://www.management-hub.com/project-management-planning.html</a>   |  |  |  |
|                 | The program will run and indicate mistaken links:  |  |  |  |
|                 |  |  |  |  |
|                 | - Duplicate activities   |  |  |  |
|                 |  |  |  |  |
|                 | - Loose ends   |  |  |  |
|                 | Come O-  |  |  |  |
|                 | - Gaps   |  |  |  |
|                 |  |  |  |  |
|                 |  |  |  |  |
|                 |  |  |  |  |
|                 | It will not indicate a mistake in the logic!   |  |  |  |

|                 | Calculations by the program:  |  |
|-----------------|---|--|
|                 | - Earliest start time   |  |
|                 | - Earliest finish time  |  |
|                 | - Latest start time   |  |
|                 | - Latest finish time  |  |
|                 | - Total float of each activity.   |  |
| Recommended     | 1. Assumptions about resources: describe which necessary equipment or other         |  |
| <u>approach</u> | resources will be available   |  |
|                 | 2. Make a list of all activities, give short descriptions; indicate the duration of |  |
|                 | each activity, the resources it consumes and the person responsible for the         |  |
|                 | activity  |  |
|                 | 3. Draw the start and finish events   |  |
|                 | 4. Identify those activities which are independent of any other activities, draw    |  |
|                 | them as starting activities   |  |
|                 | 5. Draw the other activities spread out across the paper                            |  |
|                 | 6. Join the activities with dummies to show logical relationships between           |  |
|                 | activities (strict logic)   |  |
|                 | 7. Make assumptions about the sequence of events which are not connected            |  |
|                 | by strict logic (loose logic) and draw the possible dummies to indicate the         |  |
|                 | loose logic between events  |  |
|                 | 8. Identify the final activities and connect them to the Finish event               |  |
|                 | 9. Check that there are no duplicate activities, loose ends, gaps or loops          |  |
|                 | 10. Delete superfluous dummies  |  |
|                 | 11. Give a number to each event, leaving gaps between the numbers, so that          |  |
|                 | more events could be inserted later   |  |
|                 | 12. Calculate the Earliest Event times and the Latest Event Times                   |  |
|                 | 13. Calculate the event slacks and indicate the critical events                     |  |
|                 | 14. Calculate the Earliest Start Times and Latest Start Times of the activities     |  |
|                 | 15. Indicate the critical activities and critical paths                             |  |
|                 | 16. Calculate the Free and total Float of each activity                             |  |
|                 | 17. Try to economize on time or resources from an activity with a large total       |  |
|                 | float to a critical activity.   |  |

## Isolation of Rhizorhapis suberifaciens

Corky root of lettuce (Fig. 1) can be caused by members of several closely related genera of bacteria: *Rhizorhapis, Sphingobium, Sphingopyxis* and *Rhizorhabdus*. The bacteria grow only slowly on low-carbon media; they are oligotrophic. Isolation of these bacteria is very difficult. Sterile filters are used to screen out other bacteria before dilution plating of a suspension. Preparation of all materials needed for isolation takes time and careful planning.

Strains of *Rhizorhapis* and related genera that are pathogenic to lettuce can be isolated from recently infected roots with initial symptoms of corky root (yellow areas on the tap root or main lateral roots). It is difficult to isolate these genera directly from soil, because they are very slow-growing, and truly selective media are not available. This problem can be circumvented by using lettuce seedlings as baits. The best yields of the bacteria occur when only the yellow segments of the infected roots are used for isolation. After the infected roots assume a corky appearance the corky root bacterium is overwhelmed by secondary organisms.

To prepare for isolation of *Rhizorhapis* and related genera from corky root infected material, large, capped test tubes with 20 ml deionized water are autoclaved (as many tubes as samples). In addition, six times as many small, capped test tubes with 9 ml deionized water are autoclaved, as well as several bottles containing 100 ml deionized water. Also, 50-ml empty beakers and small ceramic mortars and pestles need to be autoclaved (as many as the number of samples); beakers and mortars can be placed upside down in an enamel or steel pan for autoclaving. For isolation, S medium (Table 1) is prepared with streptomycin sulfate (30 mg per liter), called S+ medium; cycloheximide (100 mg per liter) can be added but that is not strictly needed. The antibiotics need to be added after the autoclaved medium has cooled to 55°C. The amended medium is mixed and dispensed into sterile plastic Petri plates.

For isolation from the root surface and the root tissues, small sections from young, yellow root lesions are cut and placed into separate large test tubes with 20 ml water. These test tubes are placed in a rack in a small bath-type ultrasonic cleaner and sonicated for 20 minutes. Working inside a sterile transfer hood, the supernatant of each sample (each tube) is poured into a sterile syringe (without needle) with a sterile 0.65 or 0.8  $\mu$ m pore filter attached to the pointed end of the syringe. The filtration will eliminate most fungi and large bacteria. Commercial pre-sterilized disposable syringes and filters or manually assembled and autoclaved filter units can be used. The samples are pushed through the filters with the syringe plunger into sterile beakers underneath. Then 10-fold dilutions are prepared using the 9 ml sterile water in test tubes, vortexing vigorously. *Rhizorhapis* and related genera are sensitive to phosphate buffer! From the 10<sup>-1</sup>, 10<sup>-2</sup>, and 10<sup>-3</sup> dilutions in sterile water, 0.1 ml is plated onto each of five S+ media plates using an alcohol flamed glass, metal or plastic bacteria spreader. Many replications are needed, because you may find just one colony of pathogenic *Rhizorhapis* or related genera amidst 100 other bacteria.

For isolation from lettuce root tissues, the initial steps are the same as in the previous paragraph. One sonicated root section and 1ml sterile water are added into a sterile mortar, and the tissue is ground with a sterile pestle until well macerated. Additional sterile water (8 ml) is added and vortexed. The suspension is pushed through 0.65 or 0.8  $\mu$ m pore size sterile filters as described above. From the filtrate 1 ml is transferred to the first test tube with 9 ml sterile water and ten-fold dilutions are prepared down to 10<sup>-5</sup> dilution. From the 10<sup>-3</sup>, 10<sup>-4</sup>, and 10<sup>-5</sup> dilutions, 0.1 ml is plated onto each of five S+ media plates. The dilutions are streaked in one waving pass on S+ plates. The periphery of individual plates is covered by parafilm to avoid drying out during the long incubation time at 28°C. It is very likely

that bacteria other than the corky root pathogens will grow on the plates. They are usually fast growing. The corky root bacterium will be visible as very small clear or white colonies in 2-3 weeks. Be patient!

After two to three weeks of incubation at 28°C, the colonies are at the most 1 mm in diameter. Colonies of various species of *Rhizorhapis* and closely related genera will become firm, initially smooth but later wrinkled, and cream to yellow, sometimes producing a brown pigment on S-medium (Fig. 2 and 3). The most common isolates of *R. suberifaciens* are creamy white. *Sphingobium mellinum* colonies are honey-yellow, *Sphingobium xanthum* colonies bright yellow, and *Rhizorhabdus argentea* silvery white (Francis et al. 2014). It's all in the name! Very carefully select and transfer compact and slow-growing colonies to S medium. The colonies are so compact that a complete colony sticks onto the loop. It is wise to check cells of putative *Rhizorhapis* colonies under the phase contrast microscope; the cells should be elongated and narrow (0.3 X 0.8-1.1  $\mu$ m), sometimes in strings.

• Francis, I.M., Jochimsen, K.N., de Vos, P., and van Bruggen, A.H.C. 2014. Reclassification of rhizosphere bacteria including strains causing corky root of lettuce as *Rhizorhapis suberifaciens* gen. nov., *Sphingobium mellinum* sp. nov., *Sphingobium xanthum* sp. nov., *Sphingopyxis sp., and Rhizorhabdus argenteus* gen. nov., sp. nov. Int. J. System. Evol. Microbiol. 64: 1340-1350.

| Ingredient                           | g / L distilled H <sub>2</sub> O |
|--------------------------------------|----------------------------------|
| Enzymatic casein hydrolysate         | 5.0                              |
| (Sigma C1026 or N-4517 or N-4642)    |                                  |
| Glucose                              | 2.5                              |
| K <sub>2</sub> HPO <sub>4</sub>      | 1.0                              |
| KNO <sub>3</sub>                     | 0.5                              |
| MgSO <sub>4</sub> 7 H <sub>2</sub> O | 0.5                              |
| $Ca(NO_3)_2 4 H_2O$                  | 60.0                             |
| Streptomycin sulfate *               | 50.0                             |
| Agar Noble                           | 11.0                             |

Table 1. Recipe for S-medium for isolation of *Rhizorhapis* and related genera from roots and soil.

\* Add to the medium when isolating the corky root bacterium from field or greenhouse grown roots. Filter-sterilize and add to the medium after autoclaving.



Fig. 1. Initial and final symptoms of corky root on an iceberg lettuce seedling in the greenhouse (A) and on a mature plant in the Salinas Valley, California (B). Photographs taken by Ariena van Bruggen.





Fig. 2. One-month old colonies of *Rhizorhapis suberifaciens* CA1 on the left and *Rhizorhabdus sp.* CA15 on the right. Photographs by Ariena van Bruggen and Jeff Hall.



Fig. 3. Isolates of *Rhizorhapis, Rhizorhabdus, Sphingobium,* and *Sphingomonas* species on S agar. Photographs by Jeff Hall and Ariena van Bruggen.