

Chapter

Game of Clones: Students Model the Dispersal and Fighting of Japanese Knotweed (*Fallopia japonica*)

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Abstract

Fallopia japonica as an invasive alien species in Europe and North America presents a significant problem to the existing flora as well as to infrastructures and agricultural land. That is why measures and attempts to control the plant are increasing rapidly. However, conservationists are not yet able to agree on the most suitable method. In the research project ‘Game of Clones’, a team of scientists together with the help of high school students is spatially modeling the spreading behavior of knotweed under different circumstances and is creating and providing a board game as well as a computer simulation as an experimental platform. To develop sustainable assumptions to be able to model the responses of knotweed to each control measure, a vast understanding of the plant is necessary. The chapter covers the results of research activities and experiments within the project and gives a comprehensive review about Japanese knotweed.

Keywords: Japanese knotweed, invasive species, dispersal, modeling, computer simulation, board game

1. Introduction

The spread of non-native species and their impact on the environment are a much-noticed topic in science and nature conservation. Recently, also a broader public is becoming increasingly interested, especially as the annual economic loss caused by alien species is estimated to be up to 5% of the world economic output [1]. Moreover, invasive alien species are an important factor in the loss of biodiversity. In fact, an analysis of the IUCN Red List shows that it is one of the most common threats associated with extinct species. Invasive alien species can also lead to changes in the structure and composition of ecosystems that have a significant negative impact on ecosystem services and affect the economy and well-being of humans [2–4]. Although the number of documented invasive species is underestimated in many countries, the introduction of invasive species has increased significantly. In Europe, for example, the number of invasive alien species increased by 76% between 1970 and 2007 (IUCN). Only a few of the thousands of species introduced into new areas actually become invasive, which is why their

identification is the main objective of invasion biology. In Austria, 1110 alien vascular plant species have been identified, which account for 27% of the total Austrian flora. Of these, 17 species are problematic for nature conservation as they invade near-natural habitats [5]. Japanese knotweed, *Fallopia japonica*, is one of them and is considered to cause large changes to the communities and ecosystems it invades. Its large size and its clonal, monocultural growth lead to the visual, structural, and chemical transformation of ecosystems. Wherever the plant takes root, the diversity of plant species decreases. The remaining competing species are mostly non-native [6–9] and show strong reductions in height, biomass, and specific leaf area (SLA) [10]. Once a *F. japonica* stand is established, the clonal connectivity increases its ability to grow further [6]. The vast spreading in riparian areas also results in the reduction of an overall abundance of invertebrates [3, 7]. Therefore, a large-scale invasion of *Fallopia* species is likely to seriously affect the biodiversity and quality of ecosystems and should be prevented [7].

Not only does Japanese knotweed have a negative effect on the environment, but it also causes damage to infrastructure and costs effort and money for removal work. Each year, a considerable sum is spent on vegetation management on railway and road networks [11]. *Fallopia japonica* prefers manmade locations where other plants do not have a chance; in railway structures these are graveled areas, platforms, and loading areas. Weed control is primarily carried out in the track area in order to avoid fine soil and humus accumulation and thus reduce increased water retention capacity. Also, for the treatment of the track-accompanying paths, security is the main reason [12]. The urgent need for action can also be seen in our current projects: the project “Vegetation control on roads and railways” aims for vegetation control of traffic infrastructure areas with a balanced consideration between conventional and effective eco-alternative methods. In another project we are taking over the scientific monitoring for railway embankment grazing on the Koralm railway in order to control Japanese knotweed [13].

In agriculture, in addition to knotweed competing with crops, contaminated goods such as humus landfills pose a real problem. Open soils and disturbed vegetation provide an opportunity for problematic plants to colonize. One centimeter of root is enough for Japanese knotweed to form a new population [14]. According to Section 21 of the Carinthian Nature Conservation Act, the release or sowing of wild plants [...] into areas in which they are not native requires a permit. A permit may only be granted if neither the natural habitats nor the native wild animal and plant species are damaged. Large economic losses can therefore occur if humus landfills partly or fully overgrown with knotweed can no longer be used as such.

At present, there is no fully effective method to control knotweed. Still, in the literature, there is a long list of control methods ranging from mechanical methods such as pulling out and mowing [15] to grazing with sheep and goats [16], planting competitive native species [17, 18], covering the roots with tarpaulin, and using herbicides [18, 19] to biological control such as the use of Japanese knotweed psyllid [20].

In summary, characteristics, effects, and control measures of Japanese knotweed are subject to numerous research projects in Central Europe and North America; “Game of Clones” is one of them and approaches the topic in a somewhat different and playful way. A team of scientists together with the help of high school students aims to spatially model the spreading behavior of knotweed under different circumstances and to create and provide a computer simulation as an experimental platform as well as a board game. Considering that multiple components are required, first, a vast understanding of knotweed, especially regarding its ecological optima, its dispersal strategy, and its response to different control measures, is necessary. Therefrom, sustainable assumptions can be developed to be able to model the responses of knotweed to each control measure. For answering some of

the questions, experiments will be used. The outcomes will lead to the creation of a board game and a computer simulation model based on a cellular automaton to be able to analyze and demonstrate the spreading behavior of knotweed in an interactive manner. Players will try out different measures to eradicate the clones and to keep particularly valuable areas clear from the weed. Doing this, they should go as easy on resources as possible. Depending on the individual starting points, different measures and combinations of measures will lead to success, in other words, reduce or stop the plant growth. The game takes place on actual existing land (satellite images), so the computer simulation can also be consulted for concrete action planning. The students in the research project will also play a part in the browser-based programming of the strategy game; in this way, they will simultaneously be an important reference group regarding its user-friendliness and functionality. The present chapter covers the results of the research activities and experiments within the project and gives a comprehensive review about Japanese knotweed. Section 2 starts with a description of all methods used, and Section 3 will be about the corresponding results. Section 4 finally discusses the question of the necessity of invasive species removal, summarizes the results, and concludes with a range of further recommendations for improving the existing evaluation and monitoring frameworks.

2. Methods

In “Game of Clones”, a multitude of methods have been and are used. This is especially important because it is the only way to fully understand Japanese knotweed in all its parts and behaviors. The following chapter will describe each method with all its limitations and challenges in detail to be able to relate to the results.

2.1 Literature research

As a start, the team of researchers has carried out an extensive literature search. The contributions and articles collected were reviewed and classified as more or less relevant to the research question of the project. With the support of the Regional Museum of Carinthia (Landesmuseum Kärnten), a bibliography of over 200 relevant papers on *Fallopia japonica* was compiled and divided into various topics: classification, taxonomy, identification, characteristics, history, growth, reproduction, spreading, usage, impacts, monitoring, control, management, invasions, and modeling.

2.2 Phenotypic and genotypic identification

For a serious discussion about the plant, the most urgent question that needs to be clarified and cannot be answered by the literature is what exact species we are dealing with in our project area. In Central Europe, there is evidence for two introduced species, *Fallopia japonica* and *Fallopia sachalinensis*; their hybrid *Fallopia* × *bohemica* has begun spreading as well [6, 21, 22]. The two original species are relatively easy to distinguish based on the shape and size of their leaves, but discriminating them from hybrids is challenging, even for experts. Hence, we will make use of DNA-barcoding, a taxonomic method for species identification using the DNA sequence of a marker gene [23]. The sequence of base pairs is used as a marker for a particular species, analogous to the barcode on food packaging. Since the DNA sequence changes by point mutations at a generally uniform rate, more closely related individuals (and species) have more similar sequences. As long as a species remains undivided, i.e., has a common gene pool, differences between different populations are compensated again and again by gene flow. So, if samples from two individuals have clearly different

sequences, this is a sign that they come from different species [24]. The analysis of two marker genes (chloroplast marker and nuclear marker) should provide information on hybridization and distribution of the species in our project area of the Austrian federal states of Carinthia and Styria. The chloroplast marker is inherited from the maternal organism, so by using it we will see what species was maternal. The nuclear marker will indicate if the plant is homo- or heterozygote, therefore a hybrid.

In the months of July and August 2018, 95 leaf samples were collected and sent to the Canadian Centre for DNA Barcoding (CCDB) in Guelph for DNA sequencing. For 3 weeks, 72 of the leaf samples were taken from individuals in Carinthia and Styria. Care was taken to ensure that different locations and morphologically different stands were chosen. If a site was selected, a tissue piece with an area of 1×0.5 cm was sampled with clean forceps. Preference was always given to the youngest and greenest parts of the plant, rich in plastids and meristematic cells such as the tip of a leaf. The samples were then placed in airtight bags of silica gel and kept to dry. Before proceeding to the next sample, it was crucial to ensure that no residual tissue remained on the forceps by rinsing them in 95% ethanol and wiping them with a clean absorbent paper. For each sample, a herbarium voucher of several leaves and flowers was collected, dried, and archived in the Regional Museum of Carinthia. Additional metadata included the assumed species, age, and sex as well as a detailed description of the site consisting of GPS coordinates, address, and site conditions. A photo documentation comprising location, entire plant, leaf surface and underside, and flower complements the sample collection (**Figures 1 and 2**).

Each sample was assigned to a Museum ID, which links it to the voucher, the metadata, and the photo documentation. A total of 13 of the 95 samples were collected from reliably identified individuals of all three species from the herbarium in the Regional Museum of Carinthia to serve as a reference. Ten of the 95 samples were not taken from the field, but CCDB offered to organize reference samples from Eastern Asia to have some samples from Japanese knotweed's native range.

The analysis is still ongoing; in case the sequencing will be successful, the data will be fed into the global Barcode of Life Data System (BOLD).



Figure 1.
Required tools for field sampling (E.C.O Institute of Ecology).



Figure 2.
Herbarium voucher of a sample of *Fallopia sachalinensis* (Herbarium collection code: KL—Kärntner Landesherbar).

2.3 Transect monitoring

Growth rates and propagation patterns are crucial parts of the basic data needed for the modeling of *Fallopia* populations. That is why we set up two transects on the campus of the Lakeside Science & Technology Park, a science and technology park in Klagenfurt. A transect is a straight line along which one counts and records occurrences of a species. The main advantage of transect mapping is its repeatability and standardization even under difficult terrain conditions. Both our transects (10 m each) were border on infiltration areas. The exact position of the transects was chosen in such a way that the shoots are rather in the middle of the observation area in order to be able to measure the propagation better. All methods were implemented according to the manual of vegetation-ecological monitoring by Andreas Traxler [25]. The transects were divided into 10 subplots (1 × 1 m each); the measured plants were each marked with a piece of yarn. In a weekly monitoring (April–July), the growth and propagation of *Fallopia japonica* was observed with two methods. On the one hand, three shoots were selected in both transects, in which the shoot width (at a height of 10 cm) and the height itself were measured with a caliper and meterstick. On the other hand, the number of shoots in each subplot was counted, and new shoots were marked and measured for their exact position. The data gain significance if they are interpreted in connection with the weather data for the period in question, as it is intended for the compilation of the logarithms of the computer simulation.

2.4 Rhizome uncovering

A good understanding of the underground processes in the *Fallopia* clone is of central importance for our research. To understand the connection between plant growth above- and underground and the knotweed's reaction to obstacles, we laid bare the entire root network of two stands in a large-scale field experiment. The method we used was already developed and successfully applied for the root exposure of forest trees in the past [26].

The stands are located on the campus of the Lakeside Science & Technology Park in Carinthia that borders directly on the Natura 2000 site Lendspitz-Maiernigg. During construction works 2 years ago, building rubble was piled up and populations of Japanese knotweed were able to colonize the area. The first location is a 4 m high hill with a 2-year-old stand; the second location borders on the parking lot, and its stands already exist for 4 years.

After the excavation work had been carried out and the site on the hill and next to the parking lot had been dug down by 2 m, the manual excavation work began. Together with the students, teachers, and soil experts of our two cooperating schools “BORG Spittal” based in Carinthia and “HBLFA Raumberg-Gumpenstein” based in Styria, the roots were then uncovered in a period of 2 days (**Figures 3 and 4**). The rough work was done with shovels, spades, and picks; the fine work was mainly done with screwdrivers. Bit by bit, the earth was dug away along the rhizomes and roots, thus exposing the roots. The results were documented in writing, in photographically, and in overview and detail drawings. The excavated shoot and rhizome parts were disposed of by the waste management department of the city of Klagenfurt so as not to contaminate further soil. After the two work days, the holes were dug up again by the excavator.

2.5 Rhizoboxes

Rhizoboxes are a non-invasive investigation method, which offers the possibility to survey the root system growth dynamics in time and space. Based on the root uncovering in Carinthia and the knowledge gained about length and width growth of the underground biomass, the experimental arrangements for the rhizoboxes were proposed. After a test experiment, adaptations took place; further experiments will follow. The method of using rhizoboxes aims to answer the following questions: how quickly do the rhizomes of knotweed grow (growth rates and depth and width growth) in vertical and horizontal rhizoboxes? What are the limiting factors (e.g., aboveground biomass, drought, cold, light, etc.)? For this purpose, 10 rhizoboxes in size of 30 × 100 cm were built, five in horizontal and five in vertical alignment (**Figures 5 and 6**).



Figure 3.
Excavation work at location 1 (E.C.O. Institute of Ecology).



Figure 4.
Measuring the length growth of the rhizome (HBLFA Raumberg-Gumpenstein).



Figure 5.
The earth material must be sieved before the boxes are filled (HBLFA Raumberg-Gumpenstein).



Figure 6.
The rhizome was traced to simplify the measuring (HBLFA Raumberg-Gumpenstein).

Attempt 1: For the first experiment, fresh rhizome mass of Japanese knotweed was used to illustrate the growth in length, height, and width. On 13 July 2018, the first two boxes were filled with fresh earth material and the rhizome mass was planted (box 1 = 4 cm piece and box 2 = 7 cm piece). The first attempt was aborted because of glass jumps, mold formation, and too much soil and water.

Attempt 2: The second attempt started on July 27, 2018; four boxes were filled with fresh earth material. The length growth of the rhizome was measured approx. every 3–4 days, and the growth spurts were documented in an Excel file. The alignment of the boxes was optimized; the rhizome parts cast less intensively. After about 3 weeks, the growth directions and lengths were traced with a white marker. The boxes are still filled, and the rhizomes are moving inwards. Next steps will include:

- leaving some earth and rhizome material in the boxes and storing it over the winter (covered with fleece) in order to test whether there will be further growth next year,
- experimenting with two boxes being filled and sampled and simulating a longer growing period in the boarding school at a nearly constant temperature—the results are then evaluated in spring 2019, and

- starting a new rhizobox experiment with fresh material in spring 2019 and precisely documenting length and width growth.

The rhizobox experiments were all conducted by the students Philipp Poier and Julian Heywood and their teachers Renate Mayer and Irene Sölkner from the HBLFA Raumberg-Gumpenstein. Next year, observations will be longer and more regular. The present studies served as first pilot experiments to get familiar with the method.

2.6 Board game and computer simulation

All experiences gained in our research will influence and have influenced the development of both the analog and the digital version of the strategy game ‘Game of Clones’. The game is based on a spatial model using cellular automata (Figure 7) to display dynamic vegetation patterns [27]. The basic approach of cellular automata is a subdivision of the area into equally sized, mostly quadratic fields. The dynamics of the modeling results from an interaction between the neighboring cells, in which a “state” is set to overlap the neighboring field (discretely modeled temporal development). When defining neighborhoods for quadratic cells, the von Neumann neighborhood and the Moore neighborhood are being distinguished. Whereas in the von Neumann neighborhood only cells with common edges are considered neighbors (this results in 4 neighbors per cell), the Moore neighborhood also defines diagonally adjacent cells as neighbors (this results in 8 neighbors per cell). For “Game of Clones”, we chose the approach of a model with hexagonal cells, in which there are always exactly six neighbors. An application of a hexagonal model can be found, for example, in the SCIDDICA model, which models the behavior of landslides during strong water accumulations [28].

The modeling of the game includes biological parameters such as nutrient uptake, growth, and propagation rates as well as system parameters such as the shape and size of the cells to be simulated. This intersection of disciplines requires a close cooperation between expert biologists and modelers. Starting from the literature and empirical findings (reference area and experiments), the model is developed in an iterative process.

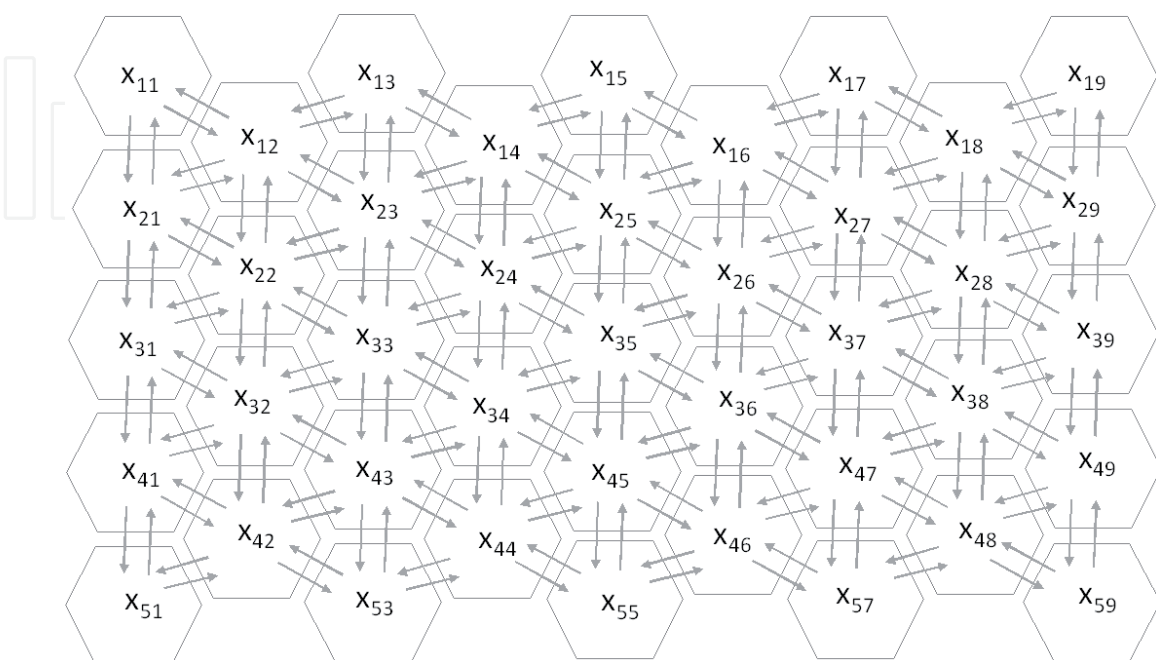


Figure 7. Principle of the cellular automaton with hexagonal cells (Institute of Networked and Embedded Systems, University of Klagenfurt).

To be able to run the model with as many systems as possible, a browser-based implementation using html5 is provided to make the system compatible. Html5 supports the execution on operating systems and is—with certain restrictions with regard to screen size—also suitable for mobile devices. NetLogo, a multi-agent programming language with an integrated modeling environment, will be used for the simulation. The development process requires a repeated feedback of the results with biologists, whereby the model parameters and assumptions are repeatedly adjusted and compared with available findings (literature and experiments). This process is of particular scientific interest and value. The user interface is developed at the same time as the model is created. For this purpose, early user tests prototypes of the user interfaces to ensure ease of operation and an attractive design. The separation of model, view, and controller (Model-View-Controller Design Paradigm) allows a largely independent further development of program parts and supports a later independent use for other projects. The software is developed under an open source license and made available as a project result.

The board game “Game of Clones” is the analog version of the computer simulation and focuses on playability and fun instead of enforcing fully realistic scenarios. In the cooperative game, players work together in order to compete against Japanese knotweed, either winning or losing as a group. The board game was developed during biweekly meetings of the experts of the E.C.O. Institute of Ecology and the Institute of Networked and Embedded Systems from the University of Klagenfurt. A prototype of the board game is already available. During the development process, the students of BORG Spittal played through several test rounds and made a strong contribution to improving the game. The computer simulation will be completed by October 2019. In contrast to the board game, full attention will be paid to the closeness to reality whereby the program will be filled with all recorded data.

3. Results

3.1 Literature research

Fallopia japonica (Houtt.) from the knotweed family (Polygonaceae) has a number of synonyms, which makes literature research more difficult (frequently: *Polygonum cuspidatum* (Sieb. & Zucc.), *Reynoutria japonica* (Houtt.), and *Polygonum japonicum* (Meissn.)) as well as a number of phenotypically similar species and hybrids (in Austria in particular: *F. sachalinensis* and *F. × bohemica*) [29–31]. Until the definitive identification of the species in our study area, we use *Fallopia japonica* as the provisional collective name for these species.

Screening the literature, one of the main findings was that considering that it is only one single species, there is a huge amount of papers that revolve around Japanese knotweed. The articles cover various aspects of the plant, having a focus on morphology, systematics, spread, and control. Following a brief story-time about knotweed’s introduction into Europe, this subchapter will be about the findings which proved to be relevant for the project.

Fallopia japonica was first introduced to Europe in 1825 by Philipp von Siebold, a Bavarian physician who worked for the Dutch government in Japan. Von Siebold had a strong interest in botany and natural history and sent a large shipment of live plants—over 500 different species—from Japan to the Netherlands, one of them being Japanese knotweed (under the name *Polygonum sieboldii*). It was intended to make a career as an ornamental and cattle feed plant and to be used in forestry as a feeding ground for red deer and as a covering plant for pheasants. The career as a useful plant did not start so well: it is of little use as a cover for pheasants, since it loses

its leaves in winter and red deer do not eat it, neither our grazing livestock. However, since in early autumn it is an excellent bee pasture when most of the European native plants have already flowered, the beekeepers have discovered Japanese knotweed for themselves [32]. Although the German Federal Nature Conservation Act and most of the Austrian Federal State Conservation Acts prohibit the planting of alien plants in the wild, beekeepers generously distributed the Japanese knotweed in the area—a first step on the way to a spread that currently places Japanese knotweed at No. 37 in the “Global Invasive Species Database”, a database managed by the Invasive Species Specialist Group (ISSG) of the IUCN Species Survival Commission.

When Japanese knotweed got introduced into Europe, they only introduced a female (male sterile) individual, never the male individual. A significant proportion of knotweed in Central Europe is not *F. japonica*, but the hybrid between it and *F. sachalinensis*—*F. bohemica*. This hybrid can reproduce with either parent and thus can replace the missing male specimens of *F. japonica*. In the same process, the hybrid produces the genetic diversity that *F. japonica* lacks so strikingly [33, 34].

All *Fallopia* species have a strong clonal growth, which allows them to surpass the surrounding species as well as to colonize new areas quickly. The basic unit of the rhizome system is a shoot clump that varies in size in different *Fallopia* species. In general, the apex of a rhizome branch eventually becomes an aerial shoot. When the shoot clump no longer produces new aerial shoots and dies, some lateral buds break the dormancy and begin to grow horizontally as new rhizome branches sometimes extending over 1 m. While *F. japonica* has rather large shoot clumps connected by long thin rhizomes, *F. sachalinensis* produces smaller shoot clumps that are more closely connected and grow in rows. *F. × bohemica* combines the characteristics of both parents and has an intermediate patch structure with smaller shoot clumps than *F. japonica* and longer rhizome connections between individual shoot clumps than *F. sachalinensis*. The fragmentation and spread of rhizomes by flooding or human activity are the most important means of propagation, as rhizome fragments of 1 cm length and 0.7 g weight can regenerate. *Fallopia* species can also regenerate from stem parts, but with lower regeneration rates. *F. × bohemica* had the highest regeneration rate of all taxa (61%) and is the most successful in regenerating and establishing new shoots. *F. japonica* and *F. sachalinensis* show lower regeneration rates (39 and 21%, respectively) [35].

The ability to regenerate in very poor soils with low nutrient requirements allows the plant to occur in a variety of habitats. It is not unusual for *F. japonica* to grow at the foot of buildings or on concrete surfaces [36]. The plant achieves its competitive superiority primarily by limiting access to light [37]. A factor, Japanese knotweed is very sensitive to, is frost. The plant is exposed to significant damages by late spring frosts when the shoots appear and by early frosts in autumn when the leaves senesce at the end of the growing season. This situation suggests that minimum spring temperatures may limit its range expansion [38]. However, climate change will open up habitats within threshold values, and frost conditions in these areas will be less severe and restrictive [39].

All these circumstances and many more make it extremely hard to get control over the invasive species. In the literature, there are many control methods and attempts described, but there are none that are completely convincing, and it amounts to a combination of different methods. Mechanical regulations focus on mowing, and although mowing during the vegetation period reduces the height and the diameter growth of shoots, the total weight of the biomass more or less stays the same [15]. The combination of cutting or mowing and using glyphosate has shown to be the most efficient and least time-consuming strategy so far [19, 40]. It is important to replant the area immediately with competitive native species to fight against invasive recolonization. On average, a suppression of knotweed is necessary

for 2 years, before native species can be successfully established [18]. Another option is a long-term grazing of cows, sheep, or goats to keep the area knotweed-free [16]. A strategy which is becoming more popular is biological control, not least since Japanese knotweed was introduced without all its natural enemies. *Aphalara itadori* (*itadori* being the Japanese word for *Fallopia japonica*), a species of psyllid from Japan which feeds on Japanese knotweed, is the subject of an application for release into the wild in Great Britain. It has been licensed by the UK Government for the biological control of Japanese knotweed in England; this is the first time that biological control of a weed has been sanctioned in the European Union [41]. Other biological controls include a leaf beetle, *Gallerucida bifasciata* [42] or snails, *Succinea putris*, and *Urticicola umbrosus* [43].

3.2 Phenotypic and genotypic identification

So far, the leaf samples have been sent to the Canadian Centre for DNA Barcoding (CCDB), the analysis is not yet complete though. The expected results should clarify which *Fallopia* species occur in Carinthia and Styria and in what proportion. The phenotypic determination suggests that only sporadic samples of *F. sachalinensis* are expected. Due to insufficient morphological differences, the phenotypic discrimination between *F. japonica* and *F. x bohemica* was not possible.

3.3 Transect monitoring

The weekly monitoring of the two transects provided information on growth rates in height and diameter. The average height growth of the six plants studied decreased as the vegetation period progressed. While the growth of the shoots in April was averagely 21.7 cm per week, it dropped to an average of 14 cm in May and to an average of 1.2 cm in June. As a percentage, the plants grew by 40, 1, 15, and 0.9%, respectively. In **Figure 8**, the growth rate is visualized for each individual, and the initial length of the shoots is as follows: 117.6 cm (A.4.1.), 35 cm (A.5.1.), 77.5 cm (A.6.1.), 32 cm (B.6.1.), 26 cm (B.9.1.), and 36.7 cm (B.9.1.). The average diameter growth was between 1.2 and -0.5 mm per week. The negative values result

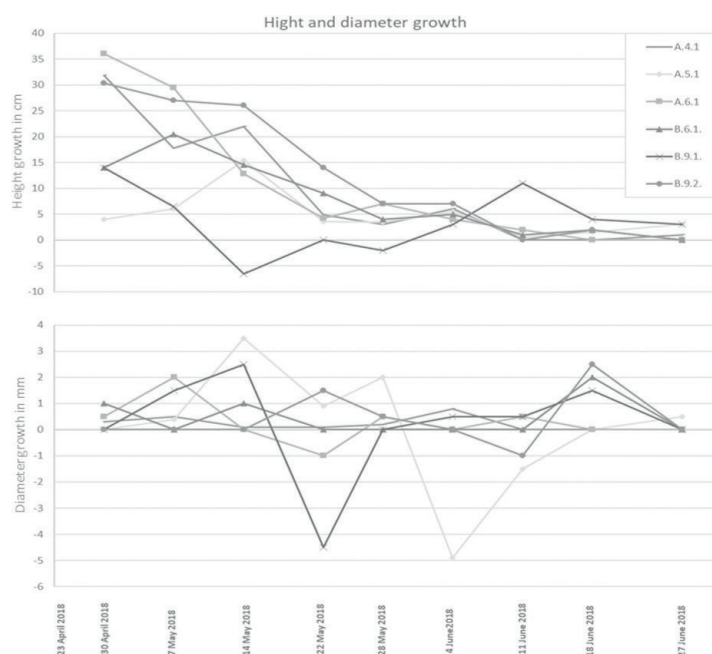


Figure 8.
Height and diameter growth of shoots in two transects (A&B).

from the fact that knotweed does not have woody shoots and the diameter size depends strongly on the water balance of the plant. Thus, it can happen that the diameter shrinks temporarily. While the growth of the diameter in April was averagely 0.3 mm (5%) per week, the growth rose to 1.8 mm (17%) in May. The plants lost biomass in the end of May/beginning of June resulting in negative values of -0.25 mm (-33%) and recovered in June with 1 mm (13%) growth rate. The initial diameters of the shoots as visualized in **Figure 8** are the following: 12 mm (A.4.1.), 2.1 mm (A.5.1.), 8.5 mm (A.6.1.), 3 mm (B.6.1.), 3 mm (B.9.1.), and 6 mm (B.9.1.). B.9.1. has low values starting in May; this results from a sudden wind break in the shoot, which has shortened the shoot and weakened the plant.

3.4 Rhizome uncovering

The rhizome uncovering could not confirm the assumption that the largest biomass of Japanese knotweed is underground. At site 1, the 2-year-old stand grew at a height of 4 m—the longest rhizomes reached a depth of 80 cm and were mainly horizontal. The reason to assume is that the plant mainly invests in the above-ground mass in the first few years. Site 2, a 4-year-old stand, underlines this assumption. The rhizomes reach 2 m into the deep until they stand at the groundwater body, which is generally high in Klagenfurt.

3.5 Rhizoboxes

The initial length of the rhizome pieces put in the rhizoboxes ranges from 5 to 24 cm. The results show that there is no correlation between initial rhizome length and growth rate. The boxes have been positioned horizontally and vertically, which showed a slight advantage for the rhizomes put in the horizontal boxes. All rhizomes started growing at a slow pace and speeded up at the end. These preliminary results have been conducted by students; further experiments are planned (**Figures 9–11**) and (**Table 1**).

3.6 Board game and computer simulation

All data and experiences gained in the project result in the development of a computer simulation. Along the way, we also created a board game for children and adults from the age of 10 years, which is currently on its way to a game publisher. The cooperation game allows players to work together as teammates against the opponent, Japanese knotweed. The game starts with a landscape full of differently



Figure 9.
Overview drawing Site 1 (E.C.O. Institute of Ecology).

Date	Rhizobox 1	Rhizobox 2	Rhizobox 3	Rhizobox 4
27 July 2018	5	5	24	4
30 July 2018	6	7	27	5.5
2 August 2018	6	7.5	30.5	5.5
6 August 2018	6	7.5	31	5.5
10 August 2018	7	8.5	31.5	6.5
31 August 2018	13.5	15.5	42.5	9.5

Table 1.

Rhizome length growth in rhizoboxes (rhizome pieces in cm); rhizoboxes 1 and 2 are positioned horizontally, and rhizoboxes 3 and 4 are positioned vertically (HBLFA Raumberg-Gumpenstein).

suitable habitats for knotweed, occupied by randomly distributed *Fallopia* clones. The players will try out different measures to eradicate the clones and to keep particularly valuable areas clear from the weed. Doing this, they should go as easy on resources as possible. The player team wins if they manage to displace all plants from the game plan and loses if one of the nature conservation areas is overgrown or destroyed by clones of knotweed. The game is based on event and action cards. Each round starts with an event card, meaning Japanese knotweed moves in a specific speed and a specific spreading mechanism. Then, it is the players' turn and they can choose between action cards that portray control methods such as mowing, pulling out, sheep grazing, glyphosate, cover foil, or biological control (*Aphalara itadori*). Depending on the individual starting points, different measures and combinations of measures will lead to success, i.e., reduce or stop the plant growth.

In several test rounds, we could see that the players started to realize how fast and determined Japanese knotweed can spread and how little can be done about it, if one does not take it seriously. The only way is to cooperate, to combine control measures and to act as soon as possible. Whenever the population is little, it is still quite easy to get rid of, and once the board is mostly overgrown by knotweed, it is extremely hard to push back the plant. The game is designed close to reality, and in terms of controlling knotweed, it shows that mechanical methods are time-consuming and inefficient, and that poison and cover foils are more efficient, but that they are not resource-saving and that one has to live with the consequences. Thus, Game of Clones creates awareness of invasive species in a playful way. The digital version is still in process, and the students in the research project will also play a part in the browser-based programming; in this way, they will simultaneously be an important reference group regarding its user-friendliness and functionality.

(E.C.O. Institute of Ecology, Institute of Networked and Embedded Systems at the University of Klagenfurt)

To our knowledge, the presented game is the only board game applying a cellular automata model to depict the spread of invasive plant species. While modeling plant growth with cellular automata is a well-established approach (a good overview can be found in [27]), there are only a few examples for usage of cellular automata simulations in board games: Franzel describes the usage of a board game to assess farmers' preferences among alternative agricultural technologies in [44]. Kang et al. [45] depict a computer simulation model addressing evolutionary game theory within a five-species jungle game, which is based on a Chinese board game. The work most related to our approach is the board game "Alien Invaders!" which teaches students how introduced species can affect native species with the example of native birds being affected by introduced species [46].

4. Conclusion

The project at the time of this publication still has a duration of 1 year, which means that many of the results are not yet complete and require further research.

The current results show that the issue of Japanese knotweed is very complex, and numerous studies and research projects have already been carried out and many are still ongoing. Due to the complexity and the costs of the control, the question arises whether the extensive control of knotweed is really necessary. But it turns out that even if one is of the opinion not to additionally intervene in the ecosystem and let nature take its course, *Fallopia japonica* also has significant economic effects, which cannot be ignored [1]. The need for further studies also arises from several disagreements in the literature such as the one regarding the gender of the species. Some say that the plant is clearly dioecious with distinct male and female individual organisms, and others speak of the plant being gynodioecious, which is the existence of male sterile and hermaphrodite individuals. There is not so much literature on the underground growth of Japanese knotweed. Hence the root uncovering was very informative, which is exactly why it would be advantageous to uncover the roots at another site, especially with older stands in order to present comparisons and observe the underground growth after the initial years.

The one method to fight knotweed does not exist. For every area, every situation, and every circumstance, a different strategy makes sense and mostly only the combination of different methods achieves an impact. When combating invasive species, however, one must always think in years. The computer simulation “Game of Clones” will be able to be overlaid with satellite images in order to establish a relationship to real areas. On the basis of the simulation, control strategies can be considered in advance of a measure concept. The project will also result in a practical guide and an explanatory video because the best way to combat an invasion is prevention and environmental education. During the collection of samples, there were numerous encounters with neighbors who were not aware of the problem and planted Japanese knotweed as a screen or threw plant remains into the compost. We hope that as many people as possible can be picked up by the game and sensitized to this topic. It would also be interesting to test whether the cooperative game method could be applied to other invasive plants in an adapted form.

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Conflict of interest

The Authors declare that there is no conflict of interest.

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