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ORIGINAL PAPER

Optimising physiochemical control of invasive Japanese knotweed

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Abstract Japanese knotweed, *Fallopia japonica* var. *japonica*, causes significant disruption to natural and managed habitats, and provides a model for the control of invasive rhizome-forming species. The socioeconomic impacts of the management of, or failure to manage, Japanese knotweed are enormous, annually costing hundreds of millions of pounds sterling (GBP£) in the UK alone. Our study describes the most extensive field-based assessment of *F. japonica* control treatments undertaken, testing the largest number of physical and/or chemical control treatments (19 in total) in replicated 225 m^2 plots over 3 years. Treatments focused on phenology, resource allocation and rhizome source–sink relationships to reduce the ecological impacts of controlling *F. japonica*. While no treatment completely eradicated *F. japonica,* a multiple-stage glyphosate-based treatment approach provided greatest control. Increasing herbicide dose did not improve knotweed control, but treatments that maximised glyphosate coverage, e.g., spraying versus stem injection, and exploited phenological changes in rhizome source–sink relationships caused the greatest reduction of basal cover and stem density after 3 years. When designing management

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D. Jones (✉) · G. Bruce · M. S. Fowler · R. Law-Cooper · D. Eastwood
Department of Biosciences, Swansea University, Singleton Park, Swansea SA2 8PP, UK
e-mail: daniel.ll.jones@gmail.com

G. Bruce
e-mail: gareth@advancedinvasives.com

M. S. Fowler
e-mail: m.s.fowler@swansea.ac.uk

R. Law-Cooper
e-mail: r.p.law-cooper@swansea.ac.uk

D. Eastwood
e-mail: d.c.eastwood@swansea.ac.uk

D. Jones · G. Bruce · R. Law-Cooper
Advanced Invasives Ltd., Institute of Life Science 2, Swansea University, Singleton Park, Swansea SA2 8PP, UK

I. Graham
Complete Weed Control Ltd., Unit 16, Hurworth Road, Newton Aycliffe DL5 6UD, UK
e-mail: ian.graham@completeweedcontrol.co.uk

A. Abel
Complete Weed Control Ltd., Unit 4, Ferry Concrete Site, Off Holden Road, Leckwith, Cardiff CF11 8BS, UK
e-mail: alan.abel@completeweedcontrol.co.uk

F. A. Street-Perrott
Department of Geography, Swansea University, Singleton Park, Swansea SA2 8PP, UK
e-mail: f.a.street-perrott@swansea.ac.uk

strategies, effective control of *F. japonica* may be achieved by biannual (summer and autumn) foliar glyphosate applications at 2.16 kg AE ha^{-1}, or by annual application of glyphosate in autumn using stem injection at 65.00 kg AE ha^{-1} or foliar spray at 3.60 kg AE ha^{-1}. Addition of other herbicides or physical treatment methods does not improve control. This work demonstrates that considering phenology, resource allocation and rhizome source–sink relationships is critical for the control of invasive, rhizome forming species.

Keywords Field trial · Glyphosate · Herbicide · Invasive alien plants (IAPs) · Invasive non-native species (INNS) · Japanese knotweed · Rhizome source–sink

Introduction

Japanese knotweed (*Fallopia japonica* var. *japonica*; referred to as *F. japonica* hereon) is one of a number of herbaceous, rhizomatous, non-climbing perennial *Fallopia* spp., collectively referred to as Japanese knotweed *sensu lato* (*s.l.*) taxa (Bailey and Conolly 2000). Japanese knotweed *s.l.* are significant Invasive Alien Plants (IAPs) across economically developed countries (Bailey 2013; Lavoie 2017). Spread is primarily through asexual (clonal) dispersal, encouraged by both anthropogenic and natural disturbance processes (e.g. disturbance by floods), accelerated by suboptimal control methods and disposal of soil contaminated with knotweed rhizome (Dawson and Holland 1999; Bailey et al. 2009).

F. japonica is a fast-growing competitor (C-strategist; Grime 2001) that exhibits highly plastic growth responses to environmental conditions (Beerling et al. 1994). It forms rhizomes (perennating woody storage organs), that commonly accumulate late in the preceding growing season, year after year (Callaghan et al. 1981). The extensive rhizome network of *F. japonica* is concentrated in the first metre of the soil profile and may extend vertically to a depth of 4.5 and 20 m laterally from the main stand of aboveground growth (Beerling et al. 1994). Above and belowground (dry) biomass values reported in northern Europe (Czech Republic, Germany and UK) range from 0.75–2.53 to 1.19–3.01 kg m^{-2}, respectively (Callaghan et al. 1981; Adler 1993; Brock 1995; Strašil and Kára 2010). Domination of plant communities by dense, monospecific *F. japonica* stands results from a rapid early season development from shoot clump and rhizome buds that allow pre-emptive occupation of space and resource capture (Grime 2001; Lavoie 2017). Dominance of non-native plant communities is maintained through the growing season via escape from herbivory i.e. the Enemy Release Hypothesis (ERH; Maurel et al. 2013) and direct and/or indirect allelopathy through the soil biota i.e. the mutualism facilitation hypothesis (Parepa et al. 2013; Parepa and Bossdorf 2016), while resource sharing through clonal rhizome integration may also aid competition and spread (You et al. 2014). Such invasions displace native flora, reducing floral assemblages and modify ecosystem functioning, e.g. soil nutrient cycling (Lavoie 2017). Socioeconomic impacts include high *F. japonica* control costs that amount to £165.6 million per annum in the UK alone (Williams et al. 2010).

We propose that *F. japonica* control treatments must account for the linkage between above and belowground tissues to inform the correct timing, concentration and intensity, e.g. rhizome dormancy maybe induced by aboveground herbicide application (Nkurunziza and Streibig 2001). The delivery of adequate herbicide into belowground tissues and/or depletion of rhizome reserves are hampered by substantial above and belowground biomass and a deep rhizome system that exhibits a strong seasonal change in source–sink strength.

Management of *F. japonica* in Europe and North America is predominantly chemical, based on a range of active ingredients promoted (Delbart et al. 2012; Clements et al. 2016). The principal active ingredient employed is glyphosate, an aromatic amino acid (AAA) synthesis inhibitor, though synthetic auxins and acetolactate synthase (ALS) inhibitors are also widely used (Online Resource 1, Table S1.1). Beyond this, there are a wide range of herbicide application methods recommended for knotweed control, few of which have been tested quantitatively or at an appropriate scale, despite widespread application (Table S1.2).

We therefore tested the three main approaches applied to *F. japonica* physiochemical control: physical (e.g. covering), chemical (e.g. application of herbicide) and integrated (e.g. cutting before herbicide spraying; Table S1.3; Child and Wade 2000). Our study combined *F. japonica* physiology (i.e. resource allocation and rhizome source–sink strength) with physical or chemical control method target (i.e. resource depletion, uptake, movement and metabolism) to develop a novel, four-stage mechanistic model to test treatment efficacy (Fig. 1). Briefly, stage 1; early season, pre knotweed emergence disruption of new aboveground growth and depletion of rhizome reserves. Stage 2, spring treatment against metabolism and growth, reducing resource acquisition. Stage 3, summer treatment at maximum height and leaf expansion, targeting the transition point where the rhizome becomes a reserve. Stage 4, late season coupling of aboveground resource translocation to the rhizome with herbicide application, maximising translocation to belowground tissues.

The primary objective of this study was to employ an evidence-based experimental approach to provide a robust, appropriately scaled field assessment of management strategies using *F. japonica* as a model for rhizome-forming IAPs. We tested 19 currently employed control strategies for effectiveness with the aims of optimising *F. japonica* control and informing field-scale management of other IAPs. Limited spatial and temporal scales (less than 2 years) of field trials conducted to date have restricted the interpretation of control outcomes and interpretation of the mechanisms underpinning effective control (Child 1999; Skibo 2007; Delbart et al. 2012). Here we report on the most extensive and comprehensive (in terms of control treatments tested), multi-year field trials of *F. japonica* control, explicitly considering whether targeting the rhizome source–sink switch can provide more effective and sustainable *F. japonica* control, by reducing pesticide application to minimise ecological impact and maximise habitat recovery (Kettenring and Adams 2011).

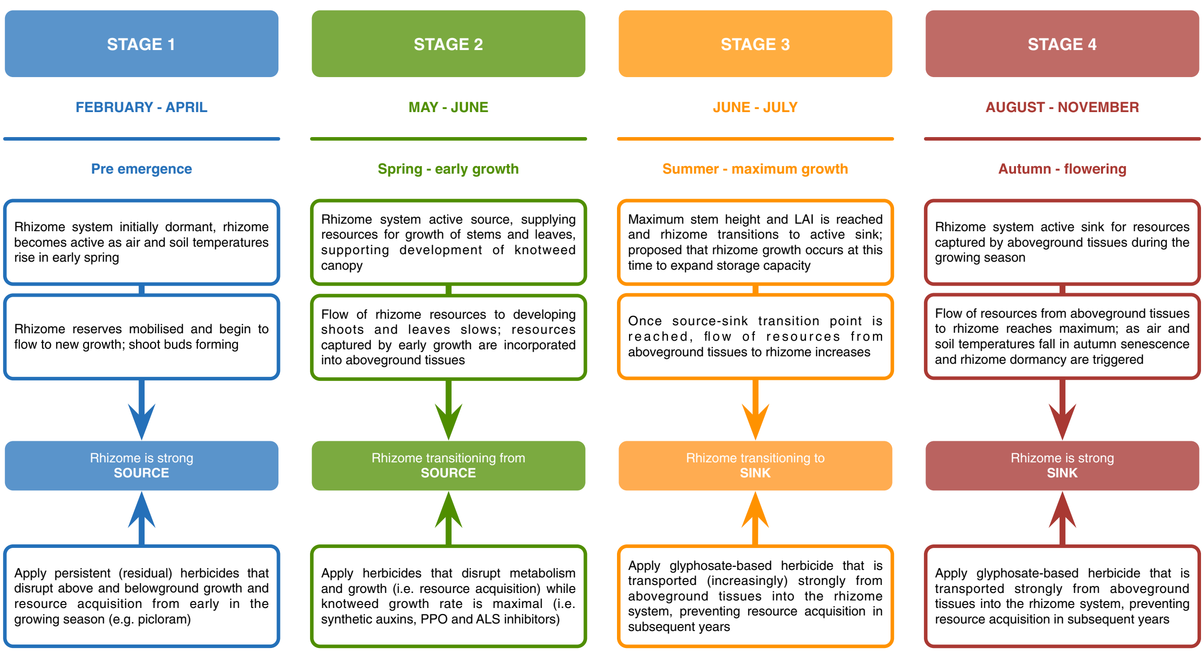

Fig. 1 Four stage mechanistic model of phenological changes in *F. japonica* growth, resource allocation and rhizome source–sink strength during the growing season. *LAI* leaf area index. *Note* linkage of above and belowground growth processes with changes in source–sink strength and that rhizome tissue sink strength increases through the growing season from June, reaching a peak in August–November during flowering and senescence. To maximise physiochemical control outcomes, physical and herbicide control treatment application should account for seasonal changes in rhizome source–sink strength. The precise timing of stages 1–4 are dependent upon local conditions and phenology may vary, impacting upon control (e.g. clones growing at higher altitude will exhibit delayed phenology, relative to lowland clones)

Methods

Field trial site selection

Three sites in south Wales (UK) were selected (Fig. 2), with comparable geological and hydrological conditions (Online Resource 2). For the present study, control methods were applied from 2012 to 2014 at sites 1 (Lower Swansea Valley Woods) and 2 (Swansea Vale Nature Reserve) and from 2013 to 2015 at site 3 (Taffs Well).

Experimental design

Fifty-eight 225 m^2 treatment and control plots were established across all three sites (Online Resource 3) and each plot was surrounded by a 1 m buffer zone. Physical, chemical and/or integrated treatments were applied to the whole of each treatment plot. Each treatment group (TG) was replicated in triplicate (with the exception of the covering treatment) and all sites contained one control plot. No dummy treatments were applied to the control plots as no facilities were available to clean the knapsack sprayer tank at field trial site 1 which may have resulted in application of dilute quantities of herbicide, influencing control plot response. Intra- and inter-site assignment of TGs was semi-randomised, as certain herbicide products could not legally be used near watercourses (e.g. picloram; Online Resource 2).

Annual plot assessment was undertaken in spring or autumn before control treatment application and was based on six randomly assigned 4 m^2 monitoring patches within each field trial plot; pre-treatment assessment commenced in 2012. Data captured included: aboveground *F. japonica* stem density, 4 m^2; *F. japonica* basal percentage cover (%) and whole plant maximum light utilisation efficiency of PSII (F_v/F_m). F_v/F_m was measured using a chlorophyll fluorescence system (Handy Plant Efficiency Analyser (PEA), Hansatech Instruments, King's Lynn, UK; light intensity 3000 μmol m^{-1} s^{-2}; dark adaption time calibrated). Mean whole plant F_v/F_m was derived from leaf measurements taken at 25, 50 and 75% of total plant height (to reflect leaf age); six representative plants were measured within each treatment and control plot.

The above three responses to physical and chemical treatment were assessed to provide a complete picture of *F. japonica* response, accounting for absolute basal cover reduction, deformed regrowth, potential photosynthetic capacity and whole plant photosynthetic efficiency and physiological state. Importantly, basal cover measurements were made at ground level and recorded deformed regrowth, providing a good indicator of recovery from physiochemical treatments

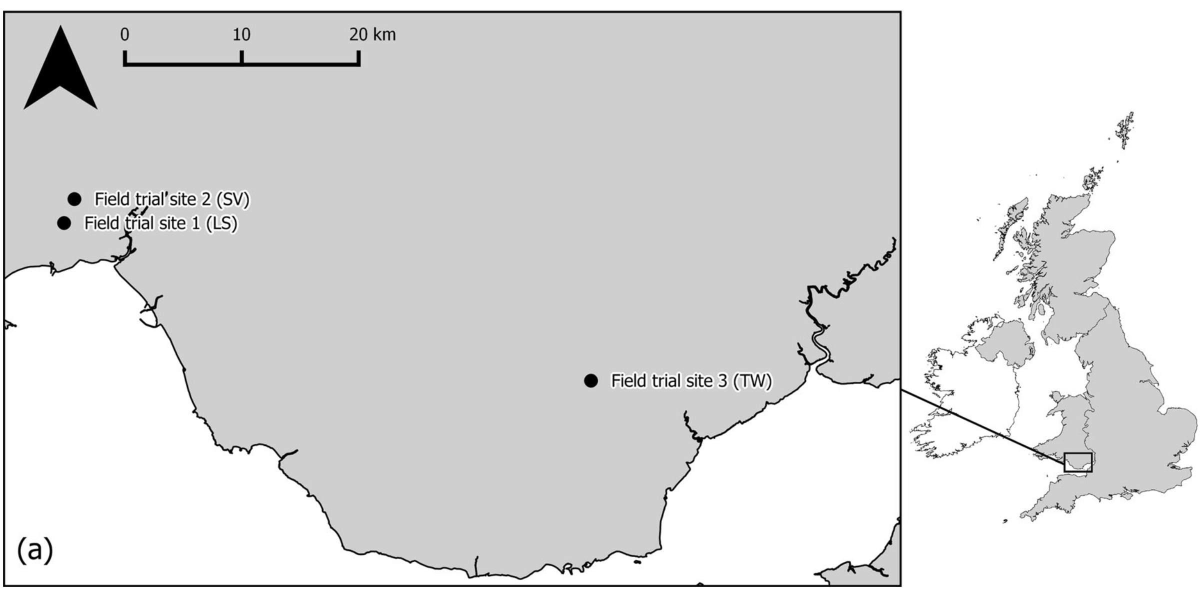

Fig. 2 Map of the study area. **a** Location of field trial sites in south Wales, UK. Field trial sites are assigned: *LS* Lower Swansea Valley Woods, *SV* Swansea Vale Nature Reserve, *TW* Taffs Well

(particularly herbicide). Stem density is a stable measurement throughout the growing period and provides indication of declining aboveground investment by the plant. F_v/F_m determines photosynthetic and carbon fixation efficiency, while also providing an indication of whole plant stress status (Maxwell and Johnson 2000; Dayan and Zaccaro 2012).

Herbicide product selection and control treatment timing

Herbicide product selection and application timing of the 19 treatments (Table 1) was based upon biological understanding of *F. japonica* source–sink relationships (Fig. 1) and existing, untested control treatments reported in the literature (Online Resource 1). The novel inclusion of a PPO inhibitor (HRAC Group E; WSSA Group 14) within the experimental design is the first time that the efficacy of this herbicide group has been reported for *F. japonica* control in the scientific literature (Online Resource 4, Table S4.1 provides herbicide product physical properties, fields of use, legal designations and UK inclusion date; Table S4.2 provides herbicide product and spray adjuvant manufacturers and suppliers).

Details of control treatments

Herbicide control treatments

Soil and foliar spray application (TGs a1 to a13, site 3) Herbicide product(s) were applied at a fixed rate (L or g ha^{-1}), with consistent application of active ingredient(s) per unit area using a Cooper Pegler CP3 (20 L) Classic knapsack sprayer, fitted with a 0.75–1.5 m telescopic lance and Cooper Pegler blue flat fan nozzle (AN 1.8). All soil and foliar spray application herbicide products were applied with dye and adjuvant (Topfilm; 1.2 L ha^{-1}) to ensure even coverage and maximise herbicide active ingredient absorption. Herbicide products containing aminopyralid (Synero, synthetic auxin) were applied with antifoaming agent (Foam Fighter). Weather forecast information was consulted to ensure that no rain was forecast for a minimum of 8 h post-application. Prior to initial soil spray herbicide application of picloram and flazasulfuron (TGs a8 and a12), aboveground *F. japonica* material from previous years, including dead stems and litter was cleared to ensure even coverage of the substratum and facilitate herbicide delivery to the rhizome and emerging shoots.

Cut and fill application (TG b1, site 3) In autumn (stage 4) of the first year of treatment, individual stems were cut at the second node above ground level, with variable rate application of 50% v/v glyphosate solution per stem (5–10 ml dose/stem; equivalent to 87.12 kg AE ha^{-1}), using a Cooper Pegler CP3 knapsack sprayer, standard lance and green anvil nozzle (AN 1.2—anvil removed). Adjuvant (1.2 L ha^{-1}) was included in the tank mix to maximise active ingredient absorption. Cut stems were left in situ to prevent dispersal of *F. japonica* propagules. In subsequent years, foliar spray application of glyphosate at full label rate (FR; 3.60 kg AE ha^{-1}) was undertaken in autumn.

Stem injection application (TG c1, site 3) In autumn (stage 4) of the first year of treatment, each individual stem was injected at the second node above ground level, with variable rate application of undiluted glyphosate per stem (3–5 ml injection volume; equivalent to 65.00 kg AE ha^{-1}), using a Nomix Enviro Stem Master injection system. Adjuvant was not included in the injection system to minimise the likelihood of blockage. In subsequent years, foliar spray application of glyphosate at FR (3.60 kg AE ha^{-1}) was undertaken in autumn.

Integrated physiochemical control treatments

Cutting and foliar spray application of glyphosate in autumn (TG d1, site 3) *F. japonica* was cut in mid growing season (summer; stage 3) to promote stand access and maximise re-growth. Cutting was performed using a Stihl FS-450 Professional 2.1 kW clearing saw and foliar spray application of glyphosate at FR (3.60 kg AE ha^{-1}) was undertaken in autumn (stage 4). In subsequent years, foliar spray application of glyphosate at FR (3.60 kg AE ha^{-1}) was undertaken in autumn.

Excavation (TGs d2 and d3, site 1) Excavation was undertaken in spring (stage 1) using a JCB 3CX backhoe loader (94 cm bucket, 0.3 m^3 capacity) to a depth of 2.5 m, with rhizome material roughly sorted and concentrated at the soil surface by the heavy

Table 1 Physiochemical *F. japonica* control treatments, showing treatment group, herbicide active ingredient (a.i.), application rate, application method and timing

Treatment group	a.i. (g L^{-1})	Application rate (kg AE ha^{-1})	Application method	Application timing
a1	Glyphosate (360)	3.60	Foliar spray	Autumn
a2	Glyphosate (360)	2.16	Foliar spray	Autumn
a3	Glyphosate (360)	2.16	Foliar spray	(i) Summer
				(ii) Autumn
a4	2,4-D amine (500)	4.50	Foliar spray	(i) Late spring
	Glyphosate (360)	3.60		(ii) Autumn
a5	Glyphosate (360)	2.16	Foliar spray	(i) Summer
	Glyphosate (360)	2.16	Foliar spray	(ii) Autumn
	+ 2,4-D amine (500)	+ 4.50		
a6	2,4-D amine (500)	2.80	Foliar spray	(i) Late spring
	Glyphosate (360)	3.60	Foliar spray	(ii) Autumn
	+ 2,4-D amine (500)	+ 2.80		
a7	Glyphosate (360)	2.16	Foliar spray	(i) Late spring
	+ 2,4-D amine (500)	+ 2.80	Foliar spray	(ii) Autumn
	Glyphosate (360)	2.16		
	+ 2,4-D amine (500)	+ 2.80		
a8	Picloram (240)	2.69	Soil and foliar spray	(i) Early spring
	Glyphosate (360)	3.60	Foliar spray	(ii) Autumn
a9	Glyphosate (360)	2.16	Foliar spray	(i) Late spring
	+ Aminopyralid (30) and Fluroxypyr (100)	+ 0.06 and 0.20	Foliar spray	(ii) Autumn
	Glyphosate (360)	2.16		
a10	Aminopyralid (30) and Fluroxypyr (100)	0.06 and 0.20	Foliar spray	(i) Late spring
	Glyphosate (360)	3.60	Foliar spray	(ii) Autumn
a11	Glyphosate (360)	2.16	Foliar spray	(i) Late spring
	+ Flazasulfuron 25% w/w	+ 0.15	Foliar spray	(ii) Autumn
	Glyphosate (360)	2.16		
a12	Flazasulfuron 25% w/w	0.15	Soil and foliar spray	(i) Early spring
	Glyphosate (360)	3.60	Foliar spray	(ii) Autumn
a13	Glyphosate (360)	2.16	Foliar spray	(i) Late spring
	+ Flumioaxazin (300)	+ 0.03	Foliar spray	(ii) Autumn
	Glyphosate (360)	2.16		
b1	Glyphosate (360)	87.12	Cut and fill	Autumn
c1	Glyphosate (360)	65.00	Stem injection	Autumn
d1	*Cutting*	N/A	Clearing saw	(i) Summer
	Glyphosate (360)	3.60	Foliar spray	(ii) Autumn
d2	*Excavation*	N/A	Excavator	(i) Early spring
	Glyphosate (360)	3.60	Foliar spray	(ii) Autumn
d3	*Excavation*	N/A	Excavator	(i) Early spring
	Picloram (240)	2.69	Soil and foliar spray	(ii) Early spring
	Glyphosate (360)	3.60	Foliar spray	(iii) Autumn

Table 1 continued

Treatment group	a.i. (g L^{-1})	Application rate (kg AE ha^{-1})	Application method	Application timing
d4	*Covering*	N/A	Geomembrane	Early spring

Underlined herbicide active ingredients indicate product mix; italicised processes represent physical components of integrated physiochemical control treatments; roman numerals represent multi-seasonal application of physiochemical control treatments. Treatment group codes are assigned: a = soil and foliar spray herbicide application methods; b = cut and fill herbicide application method; c = stem injection herbicide application method; d = integrated physiochemical control treatments. Specific timing of seasonal application was: early spring (stage 1) = March; late spring (stage 2) = May; summer (stage 3) = June; autumn (stage 4) = September

equipment operator. For TG d3, this was immediately followed by soil spray application of picloram at FR (Tordon; 2.69 kg AE ha^{-1}) in spring and for both TGs d2 and d3, foliar spray application of glyphosate at FR (3.60 kg AE ha^{-1}) was undertaken in autumn (stage 4). In subsequent years, excavation was not performed, though soil and foliar spray application of herbicides was maintained.

Physical control treatments

Covering combined with hand pulling (TG d4, site 2) Prior to covering in early spring (stage 1), aboveground *F. japonica* material from previous years was flattened and left in situ. High-density polyethylene (HDPE) geomembrane (Viqueen® 300 μm 1200 gauge) was extended over the treatment area and weighted to remain in position for the duration of the experiment. Subsequent *F. japonica* growth beneath the membrane was flattened, while visible growth emerging around the covering was hand pulled and left in situ underneath the membrane, to prevent dispersal of *F. japonica* propagules. Covering was the only physical control treatment trialled, as other physical control treatments (pulling, digging and burning) were considered too costly, labour intensive and increased the risk of *F. japonica* spread.

Data analysis

F. japonica basal cover (%; 4 m^2) data was arcsine transformed prior to analysis (Sokal and Rohlf 1981). We used Akaike information criteria (AIC) to select the best performing model from the following four candidate models, applied to each response variable (y) for independent comparison across time (t) at each site (i):

$$y_{i,t} = \mathrm{DAT}_t \quad (1)$$

$$y_{i,t} = \mathrm{TG}_i \quad (2)$$

$$y_{i,t} = \mathrm{DAT}_t + \mathrm{TG}_i \quad (3)$$

$$y_{i,t} = \mathrm{DAT}_t + \mathrm{TG}_i + \mathrm{DAT}_t * \mathrm{TG}_i \quad (4)$$

where days after treatment (DAT) is a continuous variable indicating the days after the first treatment was applied and treatment group (TG) is a categorical variable indicating the treatment group applied (including the control). The DAT_t*TG_i term indicates the interaction term between time and treatment.

Inference was based on the parameters estimated from the best performing candidate model(s) at each site (Burnham and Anderson 2002). We used general linear (ANCOVA design) models to analyse arcsine transformed % basal cover and F_v/F_m response data and compared Poisson and Negative Binomial generalised linear models (GLMs) for the stem density response data, considering AIC and goodness-of-fit statistics (comparing residual model deviance with degrees of freedom using a χ^2-test) for the GLMs. In all cases, the Negative Binomial GLM was a more appropriate model, with the Poisson GLMs consistently being overdispersed, showing a significant difference between residual deviance and d.f. ($p < 0.001$). Therefore, only results based on the negative binomial GLMs are presented here.

Within-site comparison of the 'best' predicted treatments at each site with other treatments and respective site controls were made based upon prior knowledge of biological and treatment processes. At site 1, TG d3 (spring dig; spring picloram FR; autumn glyphosate FR) was compared with TG d2 and the control; at site 2 TG d4 (covering) was compared with the control and at site 3 TG a3 (summer and autumn glyphosate half full label rate (HR) foliar spray) was compared with all other TGs and the untreated control.

All data were analysed using R v3.2.5 (The R Development Core Team 2012). The 'MASS' package (Venables and Ripley 2002) was required for negative binomial GLMs.

Results

Basal cover control response

There was no significant change over time or difference between the three sites in % basal cover (arcsine transformed) for the untreated control plots ($F_{3,81} = 1.54$, $p = 0.21$).

The full model (Eq. 4) predicting the effects of time (DAT) and treatment groups (TG) (including their interaction) on basal cover was selected as the best model at all sites, explaining up to 70% of the variation in the data (Table 2, Online Resource 5, Table S5.1). Basal cover decreased across all TGs, except the untreated controls at sites 1 and 3, which showed no change over time (Tables 2, S5.2–5.4; see Table S5.5 for measured initial and final mean % basal cover values for each TG at each field trial site). There were also significant differences among TGs with some treatments reducing basal cover more than others (Fig. 3a, Tables S5.2–5.4).

At site 1 ($R^2 = 0.70$), spring dig, spring picloram full rate (FR), autumn glyphosate FR foliar spray (TG d3) showed a faster decrease in cover over time than spring dig, autumn glyphosate FR foliar spray (TG d2), with both treatment groups performing significantly better than the untreated control (Table S5.2). At site 2 ($R^2 = 0.27$) the untreated control showed a significant increase in basal cover over time, while covering (d4) showed no significant change over time (Table S5.3).

At site 3 (TW, $R^2 = 0.61$), summer and autumn glyphosate half rate (HR) foliar spray (TG a3) showed a faster decrease in basal cover over time than all other treatment groups except autumn glyphosate FR foliar spray (TG a1) and autumn glyphosate stem injection (TG c1, Fig. 3a, Table S5.4); no significant difference in basal cover decrease over time was observed between autumn glyphosate FR foliar spray (TG a1) and autumn glyphosate stem injection (TG c1).

Stem density control response

Full models examining change in stem density over time under different treatments (and their interaction) were the best models for all sites (Tables 3, S5.6–S5.9; see Table S5.10 for measured initial and final mean stem density values for each TG at each field trial site). At site 1, spring dig, spring picloram FR, autumn glyphosate FR foliar spray (TG d3) stem density

Table 2 ANCOVA results for arcsine transformed *F. japonica* % basal cover at each site, for the best model selected by AIC (AIC value for selected model; see Table S5.1 for AIC comparisons)

Site/model fit	Model term	d.f.	Sum sq.	Mean sq.	F value	Pr (> F)
1	DAT	1	4593.8	4593.8	144.043	< 0.001
AIC = 801.7	TG	2	2912.7	1456.4	45.666	< 0.001
$R^2 = 0.70$	DAT * TG	2	1467.0	733.5	22.999	< 0.001
	Residuals	120	3827.0	31.9		
2	DAT	1	44.72	44.72	1.071	0.3099
AIC = 209.4	TG	2	160.61	160.615	3.847	0.0602
$R^2 = 0.27$	DAT * TG	2	203.43	203.435	3.872	0.0360
	Residuals	27	1127.4	31.756		
3	DAT	1	35,261	35,261	1084.153	< 0.001
AIC = 6519.0	TG	2	9476	592	18.210	< 0.001
$R^2 = 0.61$	DAT * TG	2	6445	403	12.384	< 0.001
	Residuals	27	32,264	33		

DAT days after treatment, *TG* treatment group

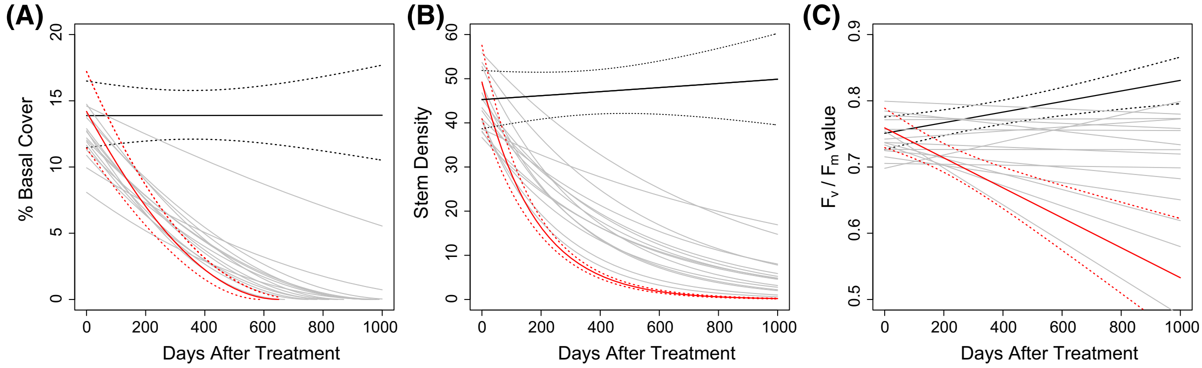

Fig. 3 Response of *F. japonica* **a** % basal cover ($R^2 = 0.61$), **b** stem density and **c** light utilisation efficiency (F_v/F_m, $R^2 = 0.23$) to 16 different treatments over time at site 3 (Taffs Well). Lines show model predicted values for the effects of each different treatment group over time. Solid black lines show values from control plots (no treatment applied). Red lines show results from the best overall performing treatment group a3 (summer and autumn foliar spray application at 2.16 kg AE ha^{-1} per application; 4.32 kg AE ha^{-1} annually). Grey lines show all other treatment groups. Dashed lines indicate 95% confidence intervals (CIs) for control and a3 treatment groups. Linear model predicted values for arcsine transformed % basal cover were back transformed for presentation in (**a**), negative binomial GLM values were used in (**b**) and untransformed linear model values used in (**c**). Coefficient estimates for all treatments are given in Supplementary Tables (Online Resource 5)

Table 3 GLM (with negative binomial error distribution) results for *F. japonica* stem density (4 m^2) at each site, for the best model selected by AIC (AIC value for selected model; see Table S5.6 for full AIC comparisons)

Site/model fit	Model term	d.f.	Deviance	Residual d.f.	Residual deviance	Pr (> χ)
1	NULL			125	291.80	
AIC = 1868.0	DAT	1	24.683	124	267.12	< 0.001
	TG	2	80.166	122	186.95	< 0.001
	DAT * TG	2	58.810	120	128.14	< 0.001
2	NULL			35	703.29	
AIC = 340.1	DAT	1	0.716	34	651.15	0.201
	TG	1	0.087	33	670.77	0.656
	DAT * TG	1	1.997	32	588.31	0.033
3	NULL			1025	1435.6	
AIC = 8278.1	DAT	1	317.33	1024	1118.2	< 0.001
	TG	16	183.38	1008	934.8	< 0.001
	DAT * TG	16	154.79	992	780.1	< 0.001

DAT days after treatment, *TG* treatment group

decreased faster over time than spring dig, autumn glyphosate FR foliar spray (TG d2) or the untreated control (Table S5.7). There was no change in stem density over time at site 2 under covering (TG d4) compared to the untreated control (Table S5.8).

Stem density did not change over time for the untreated control at site 3, but declined in all other treatments (Fig. 3b, Table S5.9). Summer and autumn glyphosate HR foliar spray (TG a3) showed significantly faster declines in stem density than any of the other treatments (Fig. 3b). Autumn glyphosate stem injection (c1) outperformed all remaining treatments except picloram-based treatments (TGs a8 and a11); however, these treatments did not perform as well as TG a3 (Table S5.9).

Light utilisation efficiency control response

Full models examining change in light utilisation efficiency over time under different treatments (and their interaction) were the best models for all sites (Tables 4, S5.11–S5.14; see Table S5.15 for measured initial and final mean F_v/F_m values for each TG at each field trial site). At site 1, only spring dig, spring

Table 4 ANCOVA results for *F. japonica* whole plant maximum light utilisation efficiency of PSII (F_v/F_m) at each site, for the best model selected by AIC (AIC value for selected model; see Table S5.11 for full AIC comparisons)

Site/model fit	Model term	d.f.	Sum sq.	Mean sq.	F value	Pr (> F)
1	DAT	1	0.0642	0.0642	14.008	< 0.001
AIC = 801.7	TG	2	0.0440	0.0220	4.796	0.010
R^2 = 0.24	DAT * TG	2	0.0229	0.0114	2.492	0.088
	Residuals	92	0.4218	0.0046		
2	DAT	1	0.001	0.001	0.042	0.840
AIC = − 13.9	TG	1	0.149	0.149	4.878	0.036
R^2 = 0.20	DAT * TG	1	0.050	0.050	1.633	0.212
	Residuals	26	0.793	0.030		
3	DAT	1	0.021	0.021	4.703	0.031
AIC = − 2274.9	TG	16	0.738	0.046	10.167	< 0.001
R^2 = 0.23	DAT * TG	16	0.388	0.024	5.353	< 0.001
	Residuals	27	3.939	0.005		

DAT days after treatment, *TG* treatment group

picloram FR, autumn glyphosate FR (TG d3) showed a significant decline in F_v/F_m readings over time (Table S5.12). There were no differences in the effects of different treatment groups over time on F_v/F_m values at site 2 (Table S5.13). At site 3, only four TGs caused a significant reduction in F_v/F_m readings over time: summer and autumn glyphosate HR foliar spray (TG a3), spring 2,4-D amine FR, autumn glyphosate FR (TG a4), summer glyphosate HR, autumn glyphosate HR and 2,4-D amine FR (TG a5) and spring glyphosate and 2,4-D amine HR, autumn glyphosate and 2,4-D amine HR (TG a7). Untreated control and a8 were both associated with an increase in F_v/F_m readings over time (Fig. 3b, Table S5.14).

Cross-site comparisons

Given the lack of significant differences over time or sites for untreated control basal cover ($F_{3,81} = 1.54$, $p = 0.21$), we tentatively highlight the following cross-site results for preliminary comparison (Tables S5.2–S5.4). At site 2, the estimate of spring dig, spring picloram FR, autumn glyphosate FR (TG d3) was comparable to summer and autumn glyphosate HR foliar spray at site 3 (TG a3) (Fig. S5.1, Tables S5.2 and S5.4). However, while the change in basal cover under the covering treatment at site 2 (TG d4) performed significantly better than the untreated control at site 2, which saw an increase in basal coverage (Table S5.3), covering did not lead to a significant reduction in basal cover over time and therefore performed more poorly than the physio-chemical treatments employed at other sites (Fig. S5.1). Given the differences in untreated control stem density and F_v/F_m values across the sites (Fig. S5.2), we do not make any further cross-site comparisons here.

Discussion

Our study represents the largest field-based assessment of *F. japonica* control treatments to date, employing experimental designs at appropriate spatial and temporal scales needed for field-appropriate control of invasive, perennial, rhizome-forming species, such as *F. japonica*. Limited information can lead to excessive herbicide use, and costly, labour intensive and unsuccessful management strategies (Kettenring and Adams 2011). We show that later season (summer/stage 3 onwards, Fig. 1) glyphosate application provides the best control and that consideration of the above and belowground source–sink relationship increases the potential treatment window from June to October.

Through assessment of 58 treatment plots (225 m^2) and 348 sampling plots (4 m^2), this study aimed to account for extensive lateral extension of the rhizome from the aboveground stands and provide appropriate scale for the parameters measured. Sampling over 3 years following herbicide treatment ensured data was available for the recovery of vegetation, often lacking in other studies, which may overestimate the negative impact of treatments (Kettenring and Adams 2011). Due to difficulties in obtaining accessible field sites of sufficient scale (Kabat et al. 2006), previous studies have been affected by small treatment plots (Skibo 2007), geographically discrete, individual stands (Delbart et al. 2012) and split-plot designs (Child 1999). In our study, annual assessment of all treatment, control and sampling plots over 3 years (pre and post-treatment) delivered a robust and scale-appropriate dataset to support our conclusions.

Physical, chemical and integrated control treatment application was married with biological understanding of *F. japonica*. In spring (stages 1 and 2, Fig. 1), all control methods applied were intended to maximise resource depletion, through tillage (excavation), resource restriction (light; covering, PPO and ALS inhibitors) and/or disruption of above (synthetic auxins and ALS inhibitors) and belowground growth (picloram, synthetic auxin). Later season glyphosate application (stages 3 and 4, Fig. 1) aimed to maximise herbicide transit by coupling to the mass flow of photosynthates through the phloem to the rhizome (Price et al. 2002).

Greatest control of aboveground *F. japonica* growth, defined by reduced basal cover and stem density (Fig. 3a, b), was obtained using glyphosate alone, where application timing was coupled to photosynthate flow to the rhizome (Fig. 1). It is notable that stem injection required 15.07 times more glyphosate per unit area than either spray treatment and was more labour intensive to apply. In plants, glyphosate (N-(phosphonomethyl)glycine) inhibits 5-enolpyruvylshikimimate-3-phosphate synthase (EPSPS) disrupting the synthesis of aromatic amino acids (e.g. tryptophan), secondary products, plant growth substances, carbon metabolism, mineral nutrition, oxidative processes and plant–microbe-interactions (Gomes et al. 2014). Specifically, inhibition of tryptophan synthesis in the shikimate pathway, results in suppression of indole-3-acetic acid (IAA) biosynthesis (Jiang et al. 2013). Upon foliar application, glyphosate penetrates rapidly through the plant cuticle prior to slow symplastic uptake. Glyphosate then moves to metabolically active sink tissues with high expression of EPSPS, i.e. *F. japonica* rhizome meristems (active shoot clump and rhizome buds), while aboveground tissues display limited herbicide injury. Although there is a linear relationship between glyphosate dose and tissue concentration (Feng et al. 2003), the distribution across leaf, stem and root tissues in *F. japonica* is independent of dose and is determined by sink strength (Buschmann 1997). This contrasts with smaller, annual dicotyledonous plants that respond in a dose-dependent manner at the whole plant level (Gomes et al. 2014). Mature *F. japonica* leaves provide a strong source of glyphosate and its relatively slow mode of action means that translocation to active rhizome sink tissues can be achieved (Cerdeira and Duke 2006).

Glyphosate accumulation in rhizome meristems causes extensive localised cell and tissue death via blocking of IAA biosynthesis (Gomes et al. 2014). Regrowth tissue showed limited chronic stress in numerous treatment plots (F_v/F_m) when compared to untreated control plants, including autumn full rate foliar spray (TG a1) (Fig. 3c, Table S6.12) suggesting that while active meristems are poisoned effectively, regrowth occurs from healthy (previously dormant) buds of low sink strength, to which lateral rhizome translocation of herbicide is limited. Sub-lethal effects of insufficient glyphosate accumulation include aboveground tissue survival within the season of herbicide application and deformed regrowth due to retention of glyphosate in (previously) active meristems in subsequent years, due to insufficient glyphosate accumulation and/or retention (Fig. 3; Feng et al. 2003; Cerdeira and Duke 2006).

Significantly reduced stem density and F_v/F_m measurements recorded with summer and autumn glyphosate foliar spray application (TG a3) compared with autumn full rate foliar spray (TG a1, Fig. 3b, c) suggests translocation and poisoning of active buds from June onwards (summer/stage 3) onwards, prior to mass transit of photosynthate in autumn (stage 4). Reduced TG a3 F_v/F_m measurements by the end of the field trials may demonstrate a chronic stress response resulting from disruption of mid-season rhizome expansion that limits its storage (source) capacity in subsequent years. Further research should aim to determine whether excess resource translocated in summer (stage 3) might support rhizome growth, while mass transit at stage 4 is used to store acquired resources to support growth in the following season. Interestingly, combining glyphosate and 2,4-D amine (TGs a4, 5 and 7) in summer and autumn also significantly reduced F_v/F_m measurements compared with the untreated control, yet effective control of aboveground *F. japonica* growth was not recorded (Fig. 3).

The application of synthetic auxins 2,4-D amine, picloram, aminopyralid and fluroxypyr (TGs a4 to 10, d3), ALS inhibitor flazasulfuron (TGs a11 and 12), and PPO inhibitor flumioxazine (TG a13) did not significantly reduce long-term basal cover or stem density compared with two foliar glyphosate treatments (TG a3, Fig. 3). This poses a potential challenge for the future management of Japanese knotweed *s.l.* taxa: while *F. japonica* is a single female clone throughout much of the invasive range, other invasive hybrid knotweeds (particularly *Fallopia* × *bohemica*) possess greater genetic diversity (Bailey 2013). Consequently, reliance upon a single herbicide (glyphosate) may lead to resistance development in these hybrid populations. Accordingly, further research should be performed to find alternative effective herbicides to slow or avoid glyphosate resistance development in these species.

Integration of excavation with picloram and glyphosate (TG d3) showed a greater reduction in basal cover than without excavation (TG a8, Fig. 3). This was presumably through picloram suppression of active and dormant rhizome buds brought to the surface during excavation. However, TG d3 performance was comparable with summer and autumn glyphosate HR foliar spray (TG a3), despite d3's greater labour and equipment requirements and cost. Additionally, picloram was deregulated without replacement within the EU in 2015, prohibiting use over a significant part of the invasive range. Reduction in stem density caused by pre-emergence (stage 1) and mid-season (stage 2) herbicide application allows better access to stands and has the appearance of immediate *F. japonica* control. However, basal cover remains high, indicating regrowth and recovery of aboveground growth without further treatment (i.e. late season glyphosate). Therefore, stage 1 and 2 treatments may not achieve sufficient resource depletion due to significant reserves held in the above and belowground *F. japonica* biomass.

Geomembrane covering (TG d4) was the least effective control treatment in reducing the response parameters (Online Resource 6). Integrating physical control methods with glyphosate treatments did not improve *F. japonica* control compared with glyphosate alone, i.e., summer cutting and autumn glyphosate application (TG d1), spring excavation and autumn glyphosate (TG d2) and autumn cut and fill (TG b1). Summer cutting has been recommended to enhance stand access (Gover 2005) and deplete rhizome energy reserves (Child and Wade 2000). However, telescopic lance spray equipment should provide access to all but the most inaccessible *F. japonica* stands and cutting-induced rhizome depletion has not been demonstrated empirically under field conditions. Longer-term analysis may demonstrate that excavation allows poisoning of a greater number of rhizome buds and biomass which was not detected in this 3 year study. Stem density reduction caused by autumn cut and fill treatment (TG b1) did not differ from the glyphosate spray treatments (TGs a1 and a3), despite using 20.37 times more glyphosate per unit area (87.12 kg AE ha^{-1}). Cut and fill application is restricted to stems largely located around the rhizome crowns with a diameter that can accept the equipment nozzle; therefore, overall coverage of active buds with glyphosate is low. While localised poisoning of crown buds occurs, regrowth away from the crown is unaffected, indicating that lateral translocation of glyphosate is limited (Bromilow and Chamberlain 2000) which is compounded by the removal of the aboveground biomass that drives herbicide translocation. As such, the effect on growth is not proportional to herbicide dose—there is no evidence for a classical dose–response relationship (Streibig 2013).

Approximately 75% of active ingredients used as plant protection products (PPPs) in Europe before 1993 have been withdrawn from the market following the introduction of the Pesticide Authorisation Directive (PAD) 91/414/EEC in response to public concern and medical evidence demonstrating the harmful effects of pesticides on human and wildlife health (Hillocks 2012, 2013). In turn, less toxic or less persistent molecules have been produced (Hillocks 2013) and the herbicide production industry has withdrawn support for older molecules, as sales do not support the costs involved in further (mandated) testing and re-registration. Withdrawal of certain herbicides, such as glyphosate, without suitable replacement would compromise the ability of the amenity sector to control rhizome-forming IAPs to the detriment of the wider native biodiversity and ecosystem services.

Conclusions: management of rhizome-forming IAPs

Knowledge of herbicide mode of action, appropriate dose, application timing and coverage are the most important factors for successful *F. japonica* control and this is relevant to other rhizome-forming IAPs such as *Gunnera* spp. (Gioria and Osborne 2013) and agricultural weed species such as *Convolvulus arvensis* (Tautges et al. 2016). Importantly, the addition of the transitional phenological source–sink stage (summer/stage 3, Fig. 1) may increase the logistically challenging narrow autumn treatment application timeframe and further optimisation could focus on glyphosate application and its effect on rhizome biology. Though no control treatment delivered complete eradication of *F. japonica* within 3 years of the first treatment application, glyphosate applied at an appropriate dose, phenological stage (Fig. 1) and level of coverage (using foliar spray and stem injection application) was found to be the most effective control treatment. An immediate recommendation for stakeholders is to discontinue the use of other widely used herbicides for control of *F. japonica* (particularly synthetic auxins) and unnecessary physical control methods (cut and fill, summer cutting and excavation) that add equipment and labour costs and increase environmental impacts, without improving control compared to spraying alone. While we recommend glyphosate use, it is acknowledged that there is a need to identify further herbicides or control approaches to reduce the potential risk of invasive hybrid knotweed populations developing resistance to the single effective herbicide. Rhizome-forming invasive species incur long-term ecological and socioeconomic costs, while few effective management tools are available, as shown by this study. Crucially, this experiment warns of further deregulation of herbicides, such as glyphosate and picloram, without equivalent replacement will lead to the application of greater quantities of ineffective herbicide products and reduce the viability and sustainability of *F. japonica* control.

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Data accessibility We (I) agree to archive the data associated with this manuscript should the manuscript be accepted at https://figshare.com.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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Authors: Daniel Jones[1,2], Gareth Bruce[1,2], Mike S. Fowler[1], Rhyan Law-Cooper[1,2], Ian Graham[3], Alan Abel[4], F. Alayne Street-Perrott[5] and Daniel Eastwood[1]
[1]Department of Biosciences, Swansea University, UK
[2]Advanced Invasives Ltd., Swansea University, UK
[3]Complete Weed Control Ltd., Newton Aycliffe, UK
[4]Complete Weed Control Ltd., Cardiff, UK
[5]Department of Geography, Swansea University, UK
Corresponding author email address: daniel.ll.jones@gmail.com

Online Resource 1: Physical control methods, herbicides and herbicide application methods for control of *F. japonica*

Supplementary Table S1.1 Summary of key herbicide groups employed within the present field trial. The Herbicide Resistance Action Committee (HRAC) and Weed Science Society of America (WSSA) classification systems are based on the site of action and mechanism of action, respectively (Mallory-Smith and Retzinger 2009). PPO inhibitors are investigated here for the first time; all other herbicide groups are currently used for the control of *F. japonica* in Europe and North America. Examples of active ingredients (a.i.), mode of uptake, plant mobility and legislative requirements are shown. Note that only specific (biactive) formulations of glyphosate are approved for use in or near water in the UK. a.i. = active ingredient; PRE = pre-emergent; POST = post-emergent.

HRAC Group	WSSA Group	Site of action	Mechanism of action	Example of a.i.	Mode of uptake	Plant mobility	Use near water?
B	2	Acetolactate synthase (ALS)/acetohydroxy acid synthase (AHAS) inhibitor	Inhibition of ALS/AHAS - inhibits amino acid formation	Flazasulfuron	Leaves	Phloem; xylem	No
E	14	Protoporphyrinogen oxidase (PPO) inhibitor	Inhibition of PPO - inhibits growth; tissue bleaching; necrosis	Flumioxazine	Leaves; roots	Phloem	No
G	9	Aromatic amino acid (AAA) synthesis inhibitor	Inhibition of the enzyme 5-enolpyruvylshikimimate-3-phosphate synthase (EPSPS) - inhibits amino acid formation	Glyphosate	Leaves; roots	Phloem	Yes
O	4	Synthetic auxin	Synthetic auxin growth regulator - disrupts normal cell and tissue formation	2,4-D - dimethlamine salt	Leaves; roots	Phloem; xylem	No

Supplementary Table S1.2 Summary of herbicide application methods (Child and Wade 2000; Gover 2005; Kabat et al. 2006; Skibo 2007; EA 2013; Clements et al. 2016).

Application method	Description	Advantages	Limitations
Foliar and soil spray	Herbicide application using a range of sprayer equipment, including: hand-held, knapsack and large volume sprayers	• Efficient • Cost effective	• Certain herbicides may not be used near water
Cut and fill	Stem is cut and herbicide is administered directly into stem cavity, there are substantial differences in reported cut and fill methods	• Perceived as a more targeted application method by landowners and the general public	• Labour intensive • Poor dosage control • Large volumes of herbicide required • Should not be used near water due to production of *F. japonica* propagules • Foliar spot treatment of regrowth required in subsequent years
Stem injection	Herbicide is applied directly into the stem cavity using an injection device, there are substantial differences in reported cut and fill methods	• Can be used in inclement weather • Perceived as a more targeted application method by landowners and the general public	• Labour intensive • Large volumes of herbicide required • Foliar spot treatment of regrowth required in subsequent years
Weed wiping	Herbicide is applied directly into the leaf surface using a variety of devices, there are substantial differences in reported weed-wiping methods	• Perceived as a more targeted application method by landowners and the general public	• Labour intensive • Foliar spot treatment of regrowth required in subsequent years

Supplementary Table S1.3 Summary of *F. japonica* physical control treatments and their suitability for integration with chemical control treatments (Child and Wade 2000; Gover 2005; Kabat et al. 2006; Skibo 2007; EA 2013; Clements et al. 2016).

Method	Desired effect	Timing	Frequency
Cutting, using strimmer (1, 2 and 3), mower (3 and 4) and thrasher (2). Grazing may be applicable	1. Removal of dead stems	1. Autumn/winter	1. Annually
	2. Reducing plant height prior to chemical treatment	2. March - August (allow plants to re-grow to 0.5-1.0 m before herbicide application)	2. As required
	3. Reducing vigour of plant	3. March - October	3. Four times a year
	4. Prevent spread of *F. japonica*	4. Throughout the growing season (October - March)	4. In case of mowing, repeat fortnightly and allow livestock to graze throughout growing season, prior to stocking
Pulling	• Removal of individual knotweed stems	• All year	• As shoots emerge
Digging	1. Elimination of knotweed	1. All year, preferably spring and summer	1. Once, if carried out correctly
	2. Disturb rhizome, promoting growth and susceptibility to chemical control	2. During late autumn/winter or early in growing season (March - October)	2. Annually, as required
Covering (1), barrier membranes (2) and encapsulation (3)	1. Covering of knotweed using a geotextile is intended to smother knotweed, depleting energy resources and causing death	1. All year	1. Requires cover to be maintained for at least one growing season
	2. Barrier membranes involves laying geotextiles to minimise/prevent lateral spread of rhizome	2. Permanent	2. Permanent
	3. Encapsulation involves burial of infective material within a geotextile barrier, preventing knotweed regrowth	3. Permanent	3. Permanent
Burning	1. Reduce total biomass 2. Reduce knotweed tissue viability	• All year	• Once, before burial

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Authors: Daniel Jones[1,2], Gareth Bruce[1,2], Mike S. Fowler[1], Rhyan Law-Cooper[1,2], Ian Graham[3], Alan Abel[4], F. Alayne Street-Perrott[5] and Daniel Eastwood[1]
[1]Department of Biosciences, Swansea University, UK
[2]Advanced Invasives Ltd., Swansea University, UK
[3]Complete Weed Control Ltd., Newton Aycliffe, UK
[4]Complete Weed Control Ltd., Cardiff, UK
[5]Department of Geography, Swansea University, UK
Corresponding author email address: daniel.ll.jones@gmail.com

Online Resource 2: Desk-based site geological, hydrological and historical surveys

GroundSure® geographical, geological, hydrological, current and historic landuse data layers for all sites (MapInsight®, GeoInsight® and EnviroInsight®) were obtained. These data were interpreted in conjunction with author pre-trial onsite investigations to ensure intra and inter-site comparability of *F. japonica* control results and appropriate siting of field trial plots and treatment groups (TG), i.e. only biactive formulations of glyphosate may be used near water and excavation should not be undertaken near watercourses due to the potential for dispersal of vegetative propagules.

Site 1: Lower Swansea Valley Woods
Location: WGS 84: 51.6468.92, -3.914362
Ordnance Survey (OS) Grid Reference: SS 676 959
No artificial ground, landslips, ground workings or natural ground subsidence was reported at site 1 (Table S2.1). Measures of natural ground subsidence are rated very low to moderate at the site. Substratum at the site consists of glacial till, with high permeability, overlying permeable mudstone, siltstone and sandstone bedrock. Estimated soil chemistry reported: arsenic (25 to 45 mg kg^{-1}), cadmium (<1.8 mg kg^{-1}), chromium (60 to 90 mg kg^{-1}), nickel (15 to 45 mg kg^{-1}) and lead (<150 mg kg^{-1}).

Supplementary Table S2.1 Summary of site 1 GroundSure GeoInsight® report.

Report Section		Description
Artificial Ground	Is there any Artificial Ground /Made Ground present beneath the study site?	No
	Are there any records relating to permeability of artificial ground within the study site boundary?	No
Superficial Geology & Landslips	Is there any Superficial Ground/Drift Geology present beneath the study site?	Yes
	Are there any records relating to permeability of superficial geology within the study site boundary?	Yes
	Are there any records of landslip within 500m of the study site boundary?	No
	Are there any records relating to permeability of landslips within the study site boundary?	No
Ground Workings	Historical Surface Ground Working Features from Small Scale Mapping	0
	Historical Underground Workings Features from Small Scale Mapping	0
	Current Ground Workings	0
Natural Ground Subsidence	Shrink-Swell Clay	Very Low
	Landslides	Moderate
	Ground Dissolution of Soluble Rocks	Negligible
	Compressible Deposits	Moderate

	Collapsible Deposits	Very Low
	Running Sand	Low
Records of Superficial Deposits/Drift Geology	Lex Code	TILLD-DMTN
	Description	TILL, DEVENSIAN
	Rock Description	DIAMICTON
Records of Permeability of Superficial Ground	Flow type	Intergranular & Mixed
	Maximum Permeability	High—Very High
	Minimum Permeability	Very Low—High
Records of Bedrock & Solid Geology	Lex Code	SW-SDST; SW-MDSS
	Rock Description	Swansea Member - Sandstone; Swansea Member - Mudstone, Siltstone And Sandstone
	Rock Age	Westphalian D
Records of Permeability of Bedrock Ground	Flow type	Fracture
	Maximum Permeability	Moderate—High
	Minimum Permeability	Low—Moderate
Faults	Category description	FAULT
	Feature description	Normal fault, inferred
Estimated Background Soil Chemistry	Sample Type	Sediment
	Arsenic (As) soil concentration range	25-45 mg/kg
	Cadmium (Cd)	<1.8 mg/kg
	Chromium (Cr)	60-90 mg/kg
	Nickel (Ni)	15-45 mg/kg
	Lead (Pb)	<150 mg/kg

No environmental permits, incidents or registers were reported at site 1 (Table S2.2). There are no abstraction licenses or Source Protection Zones (SPZs) within 2000 and 500 m of the site, respectively, although there are aquifers present within superficial and bedrock deposits at the site. No rivers are present at the site, though Environment Agency indicative zone 2 and 3 floodplains are present within 250 m of the site. British Geological Society (BGS) ground water flooding susceptibility is very high near the site, though the field trial plots are located on gently sloping high ground: therefore, ground water flood risk is low. There are no environmentally sensitive site designations for site1. Ground subsidence risk is moderate.

Supplementary Table S2.2 Summary of site 1 GroundSure EnviroInsight® report.

Report Section		Description
Environmental Permits, Incidents and Registers	Environmental Permits, Incidents and Registers at study site	0
Hydrogeology and Hydrology	Aquifer present within Superficial Deposits	Unproductive
	Aquifer present within Bedrock Deposits	Secondary A
	Groundwater Abstraction Licences (within 2000m of the study site)	0
	Surface Water Abstraction Licences (within 2000m of the study site)	0
	Potable Water Abstraction Licences (within 2000m of the study site)	0
	Are there any Source Protection Zones within 500m of the study site?	No
Hydrology – Detailed River Network and River Quality	Rivers present at study site?	0
Flooding	Are there any Environment Agency indicative Zone 2 floodplains within 250m of the study site?	Yes
	Are there any Environment Agency indicative Zone 3 floodplains within 250m of the study site?	Yes
	Are there any Flood Defences within 250m of the study site?	No
	Are there any areas benefiting from Flood Defences within 250m of the study site?	No
	Are there any areas used for Flood Storage within 250m of the study site?	No
	What is the maximum BGS Groundwater Flooding susceptibility within 50m of the study site?	Very high
	What is the BGS confidence rating for the Groundwater Flooding susceptibility areas?	High
Designated Environmentally Sensitive Sites	Records of Sites of Special Scientific Interest (SSSI)	0
	Records of National Nature Reserves (NNR)	0
	Records of Sites of Special Scientific Interest (SSSI)	0
	Records of Local Nature Reserves (LNR)	0
	Records of Special Areas of Conservation (SAC)	0
	Records of Special Protection Areas (SPA)	0
	Records of Ramsar sites	0
	Records of World Heritage Sites	0
	Records of Environmentally Sensitive Areas	0
	Records of Areas of Outstanding Natural Beauty (AONB)	0
	Records of National Parks	0
	Records of Nitrate Sensitive Areas	0
	Records of Nitrate Vulnerable Zones	0
Natural Hazards	What is the maximum risk of natural ground subsidence?	Moderate

Site 2: Swansea Vale Nature Reserve
Location: WGS 84: 51.666021, -3.901445
Ordnance Survey (OS) Grid Reference: SS 658 981

No artificial ground, landslips, or natural ground subsidence was reported at site 2 (Table S2.3). Historic ground workings are recorded near the site, though they do not impact upon the field trial plots. Measures of natural ground subsidence are rated very low to moderate at the site. Substratum at the site consists of alluvium, glacial till and glaciofluvial deposits with high permeability, overlying permeable mudstone, siltstone and sandstone bedrock. Estimated soil chemistry reported: arsenic (25 to 45 mg kg^{-1}), cadmium (<1.8 mg kg^{-1}), chromium (60 to 90 mg kg^{-1}), nickel (15 to 45 mg kg^{-1}) and lead (<150 mg kg^{-1}).

Supplementary Table S2.3 Summary of site 2 GroundSure GeoInsight® report.

Report Section		Description
Artificial Ground	Is there any Artificial Ground /Made Ground present beneath the study site?	No
	Are there any records relating to permeability of artificial ground within the study site boundary?	No
Superficial Geology & Landslips	Is there any Superficial Ground/Drift Geology present beneath the study site?	Yes
	Are there any records relating to permeability of superficial geology within the study site boundary?	Yes
	Are there any records of landslip within 500m of the study site boundary?	No
	Are there any records relating to permeability of landslips within the study site boundary?	No
Ground Workings	Historical Surface Ground Working Features from Small Scale Mapping	2
	Historical Underground Workings Features from Small Scale Mapping	0
	Current Ground Workings	0
Natural Ground Subsidence	Shrink-Swell Clay	Very Low
	Landslides	Very Low
	Ground Dissolution of Soluble Rocks	Negligible
	Compressible Deposits	Moderate
	Collapsible Deposits	Very Low
	Running Sand	Low
Records of Superficial Deposits/Drift Geology	Lex Code	GFSDD-SAGR; ALV-CSSG; TILLD-DMTN; GFDUD-SAGR
	Description	GLACIOFLUVIAL SHEET DEPOSITS, DEVENSIAN; ALLUVIUM; TILL, DEVENSIAN; GLACIOFLUVIAL DEPOSITS, DEVENSIAN
	Rock Description	SAND AND GRAVEL; CLAY, SILT, SAND AND GRAVEL; SAND AND GRAVEL
Records of Permeability of Superficial Ground	Flow type	Intergranular & Mixed
	Maximum Permeability	High—Very High

	Minimum Permeability	Very Low—High
Records of Bedrock & Solid Geology	Lex Code	GDB-MDSS
	Rock Description	Grovesend Formation - Mudstone, Siltstone And Sandstone
	Rock Age	Westphalian D
Records of Permeability of Bedrock Ground	Flow type	Fracture
	Maximum Permeability	Moderate
	Minimum Permeability	Low
Faults	Are there any records of Faults within 500m of the study site boundary?	Yes
	Category description	FAULT; ROCK
	Feature description	Normal fault, inferred; Coal seam, inferred
Estimated Background Soil Chemistry	Sample Type	Sediment
	Arsenic (As) soil concentration range	25-35 mg/kg
	Cadmium (Cd)	<1.8-2.2 mg/kg
	Chromium (Cr)	60-90 mg/kg
	Nickel (Ni)	15-45 mg/kg
	Lead (Pb)	<150 mg/kg

Two environmental permits, incidents or registers were reported at site 2 (Table S2.4). There are no abstraction licenses within 2 000 m of the site and there are no Source Protection Zones (SPZs) within 500 m of the site, although there are aquifers present within superficial and bedrock deposits at the site. One river is present at the site (Nant-bran, a tributary of the River Tawe) and field trial plots are located within Environment Agency indicative zone 2 and 3 floodplains. BGS ground water flooding susceptibility is very high for the site. There are no environmentally sensitive site designations shown in the GroundSure EnviroInsight® report, though the site is a Site of Importance for Nature Conservation (SINC 211). Ground subsidence risk is moderate.

Supplementary Table S2.4 Summary of site 2 GroundSure EnviroInsight® report.

Report Section		Description
Environmental Permits, Incidents and Registers	Environmental Permits, Incidents and Registers at study site	2
Hydrogeology and Hydrology	Aquifer present within Superficial Deposits	Secondary A
	Aquifer present within Bedrock Deposits	Secondary A
	Groundwater Abstraction Licences (within 2000m of the study site)	0
	Surface Water Abstraction Licences (within 2000m of the study site)	0
	Potable Water Abstraction Licences (within 2000m of the study site)	0
	Are there any Source Protection Zones within 500m of the study site?	No
Hydrology – Detailed River Network and River Quality	Rivers present at study site?	1
Flooding	Are there any Environment Agency indicative Zone 2 floodplains within 250m of the study site?	Yes
	Are there any Environment Agency indicative Zone 3 floodplains within 250m of the study site?	Yes
	Are there any Flood Defences within 250m of the study site?	No
	Are there any areas benefiting from Flood Defences within 250m of the study site?	No
	Are there any areas used for Flood Storage within 250m of the study site?	No
	What is the maximum BGS Groundwater Flooding susceptibility within 50m of the study site?	Very high
	What is the BGS confidence rating for the Groundwater Flooding susceptibility areas?	High
Designated Environmentally Sensitive Sites	Records of Sites of Special Scientific Interest (SSSI)	0
	Records of National Nature Reserves (NNR)	0
	Records of Sites of Special Scientific Interest (SSSI)	0
	Records of Local Nature Reserves (LNR)	0
	Records of Special Areas of Conservation (SAC)	0
	Records of Special Protection Areas (SPA)	0
	Records of Ramsar sites	0
	Records of World Heritage Sites	0
	Records of Environmentally Sensitive Areas	0
	Records of Areas of Outstanding Natural Beauty (AONB)	0
	Records of National Parks	0
	Records of Nitrate Sensitive Areas	0
	Records of Nitrate Vulnerable Zones	0
Natural Hazards	What is the maximum risk of natural ground subsidence?	Moderate

Site 3: Taffs Well
Location: WGS 84: 51.534124, -3.259120
Ordnance Survey (OS) Grid Reference: ST 127 824
No artificial ground, landslips, ground workings or natural ground subsidence was reported at site 3 (Table S2.5). Measures of natural ground subsidence are rated very low to moderate at the site. Substratum at the site consists of alluvium, river terrace and glaciofluvial deposits with high permeability, overlying permeable limestone. Estimated soil chemistry reported: arsenic (25 to 45 mg kg^{-1}), cadmium (<1.8 mg kg^{-1}), chromium (60 to 90 mg kg^{-1}), nickel (15 to 45 mg kg^{-1}) and lead (<150 mg kg^{-1}).

Supplementary Table S2.5 Summary of site 3 GroundSure GeoInsight® report.

Report Section		Description
Artificial Ground	Is there any Artificial Ground /Made Ground present beneath the study site?	No
	Are there any records relating to permeability of artificial ground within the study site boundary?	No
Superficial Geology & Landslips	Is there any Superficial Ground/Drift Geology present beneath the study site?	Yes
	Are there any records relating to permeability of superficial geology within the study site boundary?	Yes
	Are there any records of landslip within 500m of the study site boundary?	No
	Are there any records relating to permeability of landslips within the study site boundary?	No
Ground Workings	Historical Surface Ground Working Features from Small Scale Mapping	0
	Historical Underground Workings Features from Small Scale Mapping	0
	Current Ground Workings	0
Natural Ground Subsidence	Shrink-Swell Clay	Very Low
	Landslides	Moderate
	Ground Dissolution of Soluble Rocks	Low
	Compressible Deposits	Moderate
	Collapsible Deposits	Very Low
	Running Sand	Low
Records of Superficial Deposits/Drift Geology	Lex Code	GFSDD-SAGR; RTDU-SAGR; ALV-CSSG
	Description	GLACIOFLUVIAL SHEET DEPOSITS, DEVENSIAN; RIVER TERRACE DEPOSITS (UNDIFFERENTIATED); ALLUVIUM
	Rock Description	SAND AND GRAVEL; SAND AND GRAVEL; CLAY, SILT, SAND AND GRAVEL
Records of Permeability of Superficial Ground	Flow type	Intergranular
	Maximum Permeability	High—Very High
	Minimum Permeability	Very Low—High

Records of Bedrock & Solid Geology	Lex Code	CCL-LMST; PEMB-DOLM
	Rock Description	Castell Coch Limestone Formation - Limestone; Pembroke Limestone Group - Dolomitic Limestone
	Rock Age	Courceyan; Brigantian / Courceyan
Records of Permeability of Bedrock Ground	Flow type	Fracture
	Maximum Permeability	Very High
	Minimum Permeability	High
Faults	Category description	FAULT
	Feature description	Normal fault, inferred
Estimated Background Soil Chemistry	Sample Type	Sediment
	Arsenic (As) soil concentration range	25-35 mg/kg
	Cadmium (Cd)	<1.8-2.2 mg/kg
	Chromium (Cr)	60-90 mg/kg
	Nickel (Ni)	15-45 mg/kg
	Lead (Pb)	<150 mg/kg

One environmental permit, incident or register was reported at site 3, though it was not stated what the report related to (Table S2.6). There are no abstraction licenses within 2 000 m of the site and there are no Source Protection Zones (SPZs) within 500 m of the site, although there are aquifers present within superficial and bedrock deposits at the site. One river (River Taff) is present at the site and field trial plots are located within Environment Agency indicative zone 2 and 3 floodplains. BGS ground water flooding susceptibility is very high for the site. There are no environmentally sensitive site designations for site 1. Ground subsidence risk is moderate.

Supplementary Table S2.6 Summary of site 3 GroundSure EnviroInsight® report.

Report Section		Description
Environmental Permits, Incidents and Registers	Environmental Permits, Incidents and Registers at study site	1
Hydrogeology and Hydrology	Aquifer present within Superficial Deposits	Secondary A
	Aquifer present within Bedrock Deposits	Principal Aquifer
	Groundwater Abstraction Licences (within 2000m of the study site)	0
	Surface Water Abstraction Licences (within 2000m of the study site)	0
	Potable Water Abstraction Licences (within 2000m of the study site)	0
	Are there any Source Protection Zones within 500m of the study site?	No
Hydrology – Detailed River Network and River Quality	Rivers present at study site?	1
Flooding	Are there any Environment Agency indicative Zone 2 floodplains within 250m of the study site?	Yes
	Are there any Environment Agency indicative Zone 3 floodplains within 250m of the study site?	Yes
	Are there any Flood Defences within 250m of the study site?	No
	Are there any areas benefiting from Flood Defences within 250m of the study site?	No
	Are there any areas used for Flood Storage within 250m of the study site?	No
	What is the maximum BGS Groundwater Flooding susceptibility within 50m of the study site?	Very high
	What is the BGS confidence rating for the Groundwater Flooding susceptibility areas?	High
Designated Environmentally Sensitive Sites	Records of Sites of Special Scientific Interest (SSSI)	0
	Records of National Nature Reserves (NNR)	0
	Records of Sites of Special Scientific Interest (SSSI)	0
	Records of Local Nature Reserves (LNR)	0
	Records of Special Areas of Conservation (SAC)	0
	Records of Special Protection Areas (SPA)	0
	Records of Ramsar sites	0
	Records of World Heritage Sites	0
	Records of Environmentally Sensitive Areas	0
	Records of Areas of Outstanding Natural Beauty (AONB)	0
	Records of National Parks	0
	Records of Nitrate Sensitive Areas	0
	Records of Nitrate Vulnerable Zones	0
Natural Hazards	What is the maximum risk of natural ground subsidence?	Moderate

National Flood Risk Assessment Flood Rating (NaFRA) was reported as moderate for site 3 (Table S2.7). Further, the Environment Agency and BGS have recorded historic pluvial and ground water flooding at the site.

Supplementary Table S2.7 Summary of site 3 GroundSure FloodInsight® report.

Report Section		Description
National Flood Risk Assessment (NaFRA)	What is the National Flood Risk Assessment (NaFRA) Flood Rating for the study site?	Moderate
Historic Flood Events	Has the site been subject to past flooding as recorded by the Environment Agency?	Yes
Surface Water Floods	Is the site or any area within 50m at risk of Surface Water (Pluvial) Flooding?	Yes
Groundwater Flooding	What is the maximum BGS Groundwater Flooding susceptibility within 50m of the study site?	Very High
	What is the BGS confidence rating for the Groundwater Flooding susceptibility areas?	High
BGS Geological Indicators of historic flooding	Are there any geological indicators of historic flooding within 250m of the study site?	Yes

Overview of geological and hydrological conditions at sites 1 to 3

Geological and hydrological conditions at sites 1 to 3 are comparable, with substrata exhibiting high permeability, characteristic of south Wales river valleys (Humpage and Bide 2010) and heavy metal concentrations within UK central estimates (Vincent and Passant 2006).

Demarcating field trial plots and TG assignment

Following desk-based assessment and pre-experiment onsite investigations, field trial plots were delineated and TGs assigned, based upon proximity to water, conservation designations and accessibility. Site 1 was located away from any watercourses and consequently was assigned TG d2 and d3 that required excavation followed by application of herbicides, including picloram (Tordon 22K: excavation is not recommended near watercourses and picloram could not legally be used within 50 m of a watercourse, or SPZ. Site 2 was located adjacent to a watercourse and is designated as a Site of Importance for Nature Conservation (SINC 211). Given that herbicide use at this site may have caused public concern TG d4 was assigned, that required continuous covering and no use of herbicides. Site 3 is bordered by the River Taff to the west and south and therefore, different TG were sited in different areas of the knotweed monoculture. For example, TG incorporating glyphosate were sited toward the periphery of the site near the river and TG incorporating picloram were sited toward the centre (east) of the site. Therefore TG assignment was random where possible, but constrained by practical and legal restrictions.

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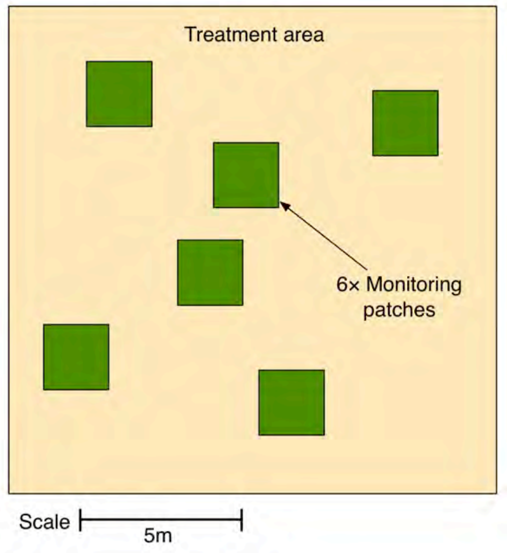
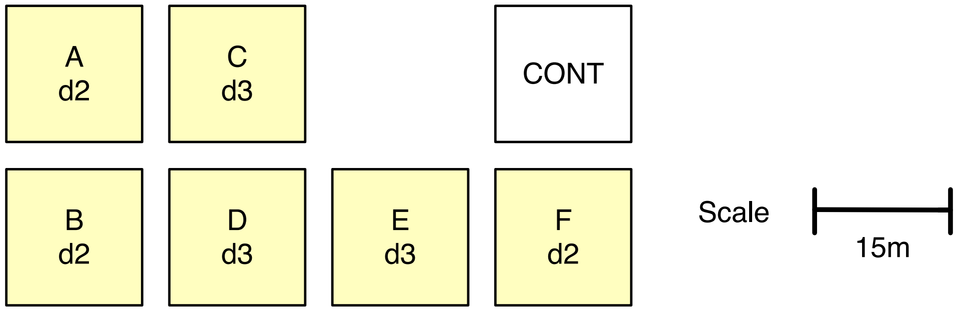
[3]Complete Weed Control Ltd., Newton Aycliffe, UK
[4]Complete Weed Control Ltd., Cardiff, UK
[5]Department of Geography, Swansea University, UK
Corresponding author email address: daniel.ll.jones@gmail.com

Online Resource 3: Field trial plot design and site treatment group assignment

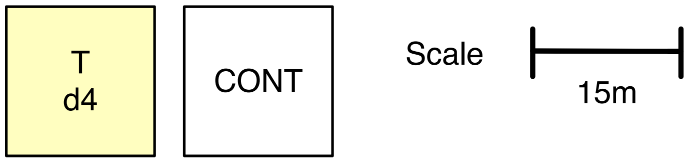


Supplementary Fig. S3.1 Scale drawing of treatment/control plot. Shows spatial arrangement of treatment area (yellow shading) and monitoring patches (green shading). Treatment plots consisted of a 225 m^2 treatment area, 1 m buffer zone surrounding the treatment area and six 4 m^2 monitoring patches assigned at random within each treatment area. The 225 m^2 treatment area and 1 m buffer zone adopted is a compromise between maximal rhizome extension (potentially resulting in treatment interference), treatment replication requirements and excavator costs.



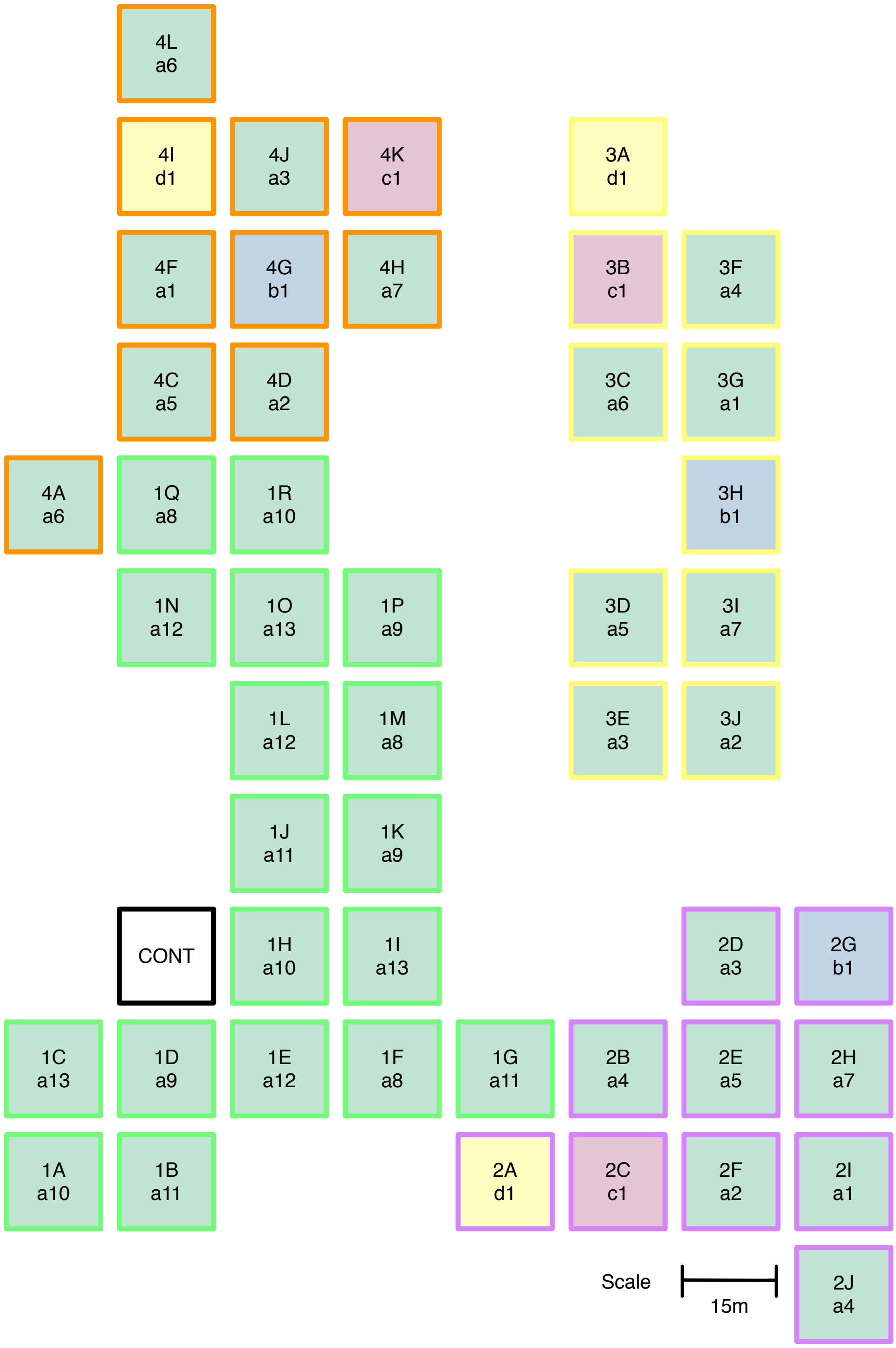
Supplementary Fig. S3.2 Schematic of field trial site 1 (Lower Swansea Valley Woods) treatment and control plot assignment. Yellow shading = integrated physiochemical control treatments (TG code d).

Supplementary Fig. S3.3 Aerial photograph of field trial site 1 (Lower Swansea Valley Woods), showing arrangement of 6 field trial plots (green outline) and one control plot (white outline) (Google Earth 2016).



Supplementary Fig. S3.4 Schematic of field trial site 2 (Swansea Vale Nature Reserve) treatment and control plot assignment. Yellow shading = integrated physiochemical control treatments (TG code d).

Supplementary Fig. S3.5 Aerial photograph of field trial site 2 (Swansea Vale Nature Reserve), showing arrangement of field trial plot (green outline) and control plot (white outline) (Google Earth 2016).



Supplementary Fig. S3.6 Schematic of field trial site 3 (Taffs Well) treatment and control plot assignment across experimental blocks 1-4. Green outline = block 1; magenta outline = block 2; yellow outline = block 3; orange outline = block 4. Green shading = soil and foliar spray herbicide application methods (TG code a); blue shading = cut and fill herbicide application method (TG code b); red shading = stem injection herbicide application method (TG code c); yellow shading = integrated physiochemical control treatments (TG code d).

Supplementary Fig. S3.7 Aerial photograph of field trial site 3 (Taffs Well), showing arrangement of field trial and control plots (Google Earth 2016). Green outline = block 1; magenta outline = block 2; yellow outline = block 3; orange outline = block 4; white outline = control plot.

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[4]Complete Weed Control Ltd., Cardiff, UK
[5]Department of Geography, Swansea University, UK
Corresponding author email address: daniel.ll.jones@gmail.com

Online Resource 4: *F. japonica* field trial herbicide properties, manufacturers and suppliers

Supplementary Table S4.1 Herbicide products selected for use in the *F. japonica* field trial, including physical properties, fields of use, legal designations and UK inclusion date. Herbicide products are organised according to the Herbicide Resistance Action Committee (HRAC) classification system. Note that from August 2014 only specific (biactive) formulations of glyphosate are approved for use in or near water in the UK and from June 2015 picloram was withdrawn from use in the UK (Mallory-Smith and Retzinger 2009). Where: PRE, product pre emergent herbicidal activity and POST product post emergent herbicidal activity.

Herbicide product	Active ingredients	Site of action	Mechanism of action	HRAC Group	WSSA Group	Chemical family	Properties	Apply	Mode of uptake	Plant mobility	Residual	Use near water	Adjuvant	CAS number	MAPP number	Hazard code	Risk phrases	Concentration (g/L)	Inclusion date
Glyfos Proactive®	Glyphosate - isopropylamine salt	Aromatic amino acid synthesis inhibitor - glycine	Inhibition of the enzyme 5-enolpyruvylshikimimate-3-phosphate synthase (EPSPS) - inhibits amino acid formation	G	9	Glycine	Non-selective; systemic	POST	Leaves; roots	Phloem	−	+	+	038641-94-0	11976	N	R51, 53	480	2018
Chikara®	Flazsulfuron	ALS/AHAS inhibitor - sufonylurea	Inhibition of acetolactate (ALS) and actohydroxy acid (AHAS) - inhibits amino acid formation	B	2	Sulfonylurea	Selective; systemic	POST; PRE	Leaves	Phloem; xylem	+	−	+	104040-78-0	14189	N	R50/53	N/A - water dispersible granule containing 25% w/w flazasulfuron	2014
Digital®	Flumioxazine	PPO inhibitor - triazolinone	Inhibition of protoporphyrinogen oxidase (PPO) - inhibits growth; tissue bleaching; necrosis	E	14	N-phenylphthalamide	Non-selective; non-systemic (contact)	POST; PRE	Leaves; roots	Phloem	+	−	−	103361-09-7	13561	T, N	R61, R50/53	300	2018
Depitox®	2,4-D - dimethlamine salt	Synthetic auxin - phenoxy	Synthetic auxin growth regulator - disrupts normal cell and tissue formation	O	4	Phenoxy carboxylic acid	Selective; systemic	POST	Leaves; roots	Phloem; xylem	−	−	+	002008-39-1	13258	Xn;N	R22, 41, 43, 51-53	500	2018
Tordon®	Picloram - potassium salt	Synthetic auxin-pyridine carboxylic acid	Synthetic auxin growth regulator - disrupts normal cell and tissue formation	O	4	Pyridine carboxylic acid	Non-selective; systemic	POST; PRE	Leaves; roots	Phloem; xylem	+	−	+	002545-60-0	15682	Xi	R43	240	2015
Synero®	Aminopyralid potassium	Synthetic auxin-pyridine carboxylic acid	Synthetic auxin growth regulator - disrupts normal cell and tissue formation	O	4	Pyridine carboxylic acid	Selective; systemic	POST; PRE	Leaves; roots	Phloem; xylem	+	−	+	566191-87-5	14708	Xi	R41-52/53	36	2015
	Fluroxypyr - methylheptyl ester	Synthetic auxin-pyridine carboxylic acid	Synthetic auxin growth regulator - disrupts normal cell and tissue formation	O	4	Pyridine carboxylic acid	Selective; systemic	POST	Leaves; roots	Phloem; xylem	−			081406-37-3		N	R50/53	144	

Supplementary Table S4.2 Herbicide product and spray adjuvant manufacturers and suppliers. Where trade names appear, no discrimination is intended and no endorsement by Swansea University College of Science is implied.

Product	Manufacturer	Supplier
Glyfos Proactive®	Headland Agrochemicals Ltd.	Nomix Enviro Ltd.
Chikara®	Belchim Crop Protection	Nomix Enviro Ltd.
Digital®	Sumitomo Chemical Company Ltd.	Interfarm (UK) Ltd.
Depitox®	Nufarm UK Ltd.	Nomix Enviro Ltd.
Tordon®	DowAgrosciences	Nomix Enviro Ltd.
Synero®	DowAgrosciences	Nomix Enviro Ltd.
Topfilm®	Biosorb Inc.	Waterland Management Ltd.
Foam Fighter®	Miller Chemical & Fertilizer Corporation	Nomix Enviro Ltd.

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Authors: Daniel Jones[1,2], Gareth Bruce[1,2], Mike S. Fowler[1], Rhyan Law-Cooper[1,2], Ian Graham[3], Alan Abel[4], F. Alayne Street-Perrott[5] and Daniel Eastwood[1]
[1]Department of Biosciences, Swansea University, UK
[2]Advanced Invasives Ltd., Swansea University, UK
[3]Complete Weed Control Ltd., Newton Aycliffe, UK
[4]Complete Weed Control Ltd., Cardiff, UK
[5]Department of Geography, Swansea University, UK
Corresponding author email address: daniel.ll.jones@gmail.com

Online Resource 5: Tables of statistical results and comparison of treatment groups across sites

Supplementary Table S5.1 AIC comparison for model selection for arcsine transformed *F. japonica* % basal cover across sites 1-3. The model including the interaction term (DAT * TG) also includes all main effects.

Site	Model	Model deviance	d.f.	AIC	ΔAIC
1	DAT	48184.71	3	889.8	117.0
	TG	75224.39	4	918.7	36.9
	DAT + TG	38708.36	5	838.6	88.1
	DAT * TG	32263.73	7	801.7	0
2	DAT	1491.45	3	214.1	4.7
	TG	1350.54	3	211.0	1.6
	DAT + TG	1330.84	4	212.5	3.1
	DAT * TG	1227.40	5	209.4	0
3	DAT	48184.71	3	6867.1	347.1
	TG	75224.39	18	7354.1	834.1
	DAT + TG	38708.36	19	6674.4	154.4
	DAT * TG	32263.73	35	6520.0	0

Supplementary Table S5.2 ANCOVA linear regression results for arcsine transformed *F. japonica* % basal cover at site 1: comparison of individual factor level estimates with TG d3. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimates represent the change in slope for that TG compared to DAT estimate given for TG d3.

Parameter	Estimate	SE	t value	Pr(>|t|)
Intercept	19.7	1.22	16.2	<0.001
DAT	-0.0322	0.00269	-12	<0.001
TG CTRL	1.64	2.45	0.668	0.506
TG d2	-0.851	1.69	-0.505	0.614
DAT : TG CTRL	0.0349	0.00515	6.78	<0.001
DAT : TG d2	0.0090	0.00414	2.17	0.032

Supplementary Table S5.3 ANCOVA linear regression results for arcsine transformed *F. japonica* % basal cover at site 2: comparison of individual factor levels with TG d4. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimate represents the change in slope for that TG compared to DAT estimate given for the control TG.

Parameter	Estimate	SE	t value	Pr(>|t|)
Intercept	14	3.37	4.15	<0.001
DAT	0.0124	0.00586	-2.11	0.044
TG d4	2.91	4.19	0.695	0.493
DAT : TG d4	-0.0172	0.0078	2.21	0.036

Supplementary Table S5.4 ANCOVA linear regression results for arcsine transformed *F. japonica* % basal cover at site 3: comparison of individual factor levels with TG a3. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimates represent the change in slope for that TG compared to DAT estimate given for TG a3.

Parameter	Estimate	SE	t value	Pr(>|t|)
Intercept	22.1	1.21	18.3	<0.001
DAT	-0.0338	0.00269	-12.6	<0.001
TG CTRL	-0.261	1.61	-0.162	0.871
TG a1	-2.18	1.68	-1.3	0.195
TG a2	-2.87	1.68	-1.71	0.0876
TG a4	-0.132	1.72	-0.077	0.939
TG a5	-1.94	1.71	-1.13	0.257
TG a6	-3.76	1.72	-2.19	0.0289
TG a7	-1.28	1.72	-0.748	0.455
TG a8	-5.61	1.59	-3.53	<0.001
TG a9	-2.51	1.56	-1.6	0.109
TG a10	-0.382	1.56	-0.244	0.807
TG a11	-1.7	1.56	-1.08	0.278
TG a12	0.332	1.59	0.209	0.835
TG a13	-1.84	1.56	-1.17	0.24
TG b1	0.444	1.72	0.258	0.796
TG c1	-1.22	1.68	-0.725	0.469
TG d1	-1.08	1.68	-0.644	0.52
DAT : TG CTRL	0.0338	0.00337	10	<0.001
DAT : a1	0.0074	0.00407	1.82	0.0696
DAT : a2	0.0105	0.00407	2.58	0.01
DAT : a4	0.0109	0.00376	2.91	0.0037
DAT : a5	0.00795	0.0038	2.09	0.0368
DAT : a6	0.0203	0.00378	5.37	<0.001
DAT : a7	0.00949	0.00378	2.51	0.0123
DAT : a8	0.0169	0.0033	5.13	<0.001
DAT : a9	0.0135	0.00336	4.02	<0.001
DAT : a10	0.0131	0.00336	3.91	<0.001
DAT : a11	0.0128	0.00336	3.82	<0.001
DAT : a12	0.0249	0.0033	7.56	<0.001
DAT : a13	0.0134	0.00336	4	<0.001
DAT : b1	0.00802	0.00407	1.97	0.0494
DAT : c1	0.00292	0.00412	0.71	0.478
DAT : d1	0.00812	0.00407	1.99	0.0464

Supplementary Table S5.5 Initial and final mean % basal coverage (± S.E.) values across all replicate plots for each treatment group (TG) at each field trial site. Total length of treatment period in days is given in the Final DAT (days after treatment) column.

Field Site	TG	Initial value	Final value	Final DAT
1	Control	12.17 ± 2.24	14.16 ± 2.30	741
	d2	12.67 ± 1.84	1.40 ± 0.28	593
	d3	15.94 ± 1.52	0.04 ± 0.03	701
2	Control	6.00 ± 1.15	16.17 ± 1.54	722
	d4	6.67 ± 0.89	5.33 ± 2.44	722
3	Control	15.08 ± 0.96	15.67 ± 1.69	1099
	a1	17.33 ± 1.11	2.33 ± 0.42	620
	a2	15.39 ± 0.98	1.58 ± 0.42	620
	a3	19.06 ± 1.07	0.17 ± 0.17	709
	a4	16.83 ± 1.18	2.58 ± 1.12	735
	a5	17.61 ± 0.95	2.00 ± 0.45	709
	a6	12.33 ± 1.20	3.00 ± 1.18	715
	a7	17.11 ± 1.39	1.46 ± 0.18	715
	a8	16.39 ± 0.89	1.25 ± 0.21	921
	a9	17.39 ± 0.59	0.70 ± 0.19	859
	a10	19.44 ± 1.06	1.25 ± 0.42	859
	a11	18.89 ± 0.86	1.65 ± 0.30	859
	a12	17.33 ± 0.71	4.17 ± 0.48	921
	a13	19.67 ± 1.02	0.70 ± 0.26	859
	b1	16.56 ± 0.71	0.83 ± 0.46	623
	c1	17.83 ± 0.90	1.42 ± 0.58	605
	d1	17.00 ± 1.36	3.25 ± 0.54	620

Supplementary Table S5.6 AIC comparison for model selection amongst *F. japonica* stem density (4 m^2) data across sites 1-3. The model including the interaction term (DAT * TG) also includes all main effects.

Site	Model	Model deviance	d.f.	AIC	ΔAIC
1	DAT	3304.46	2	3887.1	2019.1
	TG	2389.64	3	3809.0	1941.0
	DAT + TG	3384.57	4	2896.1	1028.1
	DAT * TG	1357.53	6	1868.0	0
2	DAT	651.15	2	856.3	78.9
	TG	670.77	2	888.2	110.8
	DAT + TG	703.29	3	857.9	80.5
	DAT * TG	588.31	4	777.4	0
3	DAT	13901	2	18380.0	5244.6
	TG	22600	17	27108.3	13972.9
	DAT + TG	12511	18	17021.8	3886.4
	DAT * TG	8592.7	34	13135.4	0

Supplementary Table S5.7 GLM with Negative Binomial error family results for *F. japonica* stem density (4 m^2) at site 1: comparison of individual factor levels with TG d3. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimates represent the change in slope for that TG compared to DAT estimate given for TG d3.

Parameter	Estimate	SE	z value	Pr(>|t|)
Intercept	3.87	0.033	114.25	<0.001
DAT	-0.008	0.0004	-21.86	<0.001
TG CTRL	0.364	0.057	6.42	<0.001
TG d2	0.148	0.044	3.32	<0.001
DAT : TG CTRL	0.008	0.0004	20.82	<0.001
DAT : d2	0.006	0.0004	16.17	<0.001

Supplementary Table S5.8 GLM with Negative Binomial error family results for *F. japonica* stem density (4 m^2) at site 2: comparison of individual factor levels with the control TG. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimate represents the change in slope for TG d4 compared to DAT value given for the control TG.

Parameter	Estimate	SE	z value	Pr(>|t|)
Intercept	3.55	0.0637	55.7	<0.001
DAT	0.0002	0.0001	2.01	0.0445
TG d4	0.514	0.0822	6.26	<0.001
DAT : d4	-0.0016	0.0002	8.99	<0.001

Supplementary Table S5.9 GLM with Negative Binomial error family results for *F. japonica* stem density (4 m^2) at site 3: comparison of individual factor levels with TG a3. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimates represent the change in slope for that TG compared to DAT value given for TG a3.

Parameter	Estimate	SE	z value	Pr(>|t|)
Intercept	3.97	0.0321	124	<0.001
DAT	-0.00601	0.000231	-26	<0.001
TG CTRL	-0.167	0.0424	-3.93	<0.001
TG a1	-0.185	0.0465	-3.98	<0.001
TG a2	-0.234	0.0467	-5.01	<0.001
TG a4	-0.188	0.0467	-4.03	<0.001
TG a5	-0.184	0.0469	-3.92	<0.001
TG a6	-0.272	0.0472	-5.77	<0.001
TG a7	-0.0746	0.0456	-1.64	0.102
TG a8	0.156	0.0432	3.62	<0.001
TG a9	-0.0429	0.0421	-1.02	0.308
TG a10	-0.00786	0.0419	-0.188	0.851
TG a11	0.0744	0.0419	1.78	0.0756
TG a12	0.102	0.041	2.48	0.0132
TG a13	0.037	0.0422	0.878	0.38
TG b1	0.0145	0.0443	0.326	0.744
TG c1	-0.0994	0.0461	-2.16	0.0309
TG d1	-0.177	0.0459	-3.86	<0.001
DAT : TG CTRL	0.00611	0.000237	25.8	<0.001
DAT : a1	0.00258	0.00028	9.23	<0.001
DAT : a2	0.00338	0.000269	12.6	<0.001
DAT : a4	0.00411	0.000253	16.2	<0.001
DAT : a5	0.00318	0.000264	12	<0.001
DAT : a6	0.00502	0.000248	20.3	<0.001
DAT : a7	0.00349	0.000257	13.6	<0.001
DAT : a8	-0.00062	0.000329	-1.89	0.0593
DAT : a9	0.00334	0.000253	13.2	<0.001
DAT : a10	0.00314	0.000255	12.3	<0.001
DAT : a11	0.00191	0.000272	7.02	<0.001
DAT : a12	0.0045	0.000241	18.7	<0.001
DAT : a13	0.00203	0.000271	7.49	<0.001
DAT : b1	0.00405	0.000253	16	<0.001
DAT : c1	0.000633	0.000321	1.97	0.0488
DAT : d1	0.00361	0.000264	13.7	<0.001

Supplementary Table S5.10 Initial and final mean stem density (± S.E.) values across all replicate plots for each treatment group (TG) at each field trial site. Total length of treatment period in days is given in the Final DAT (days after treatment) column.

Field Site	TG	Initial value	Final value	Final DAT
1	Control	64.33 ± 7.53	57.50 ± 9.77	741
	d2	66.00 ± 6.09	25.50 ± 5.36	593
	d3	48.67 ± 4.02	0.67 ± 0.50	701
2	Control	40.17 ± 10.33	48.50 ± 2.16	722
	d4	60.67 ± 21.18	25.00 ± 8.71	722
3	Control	49.21 ± 3.27	62.67 ± 3.14	1099
	a1	50.33 ± 1.90	11.33 ± 2.94	620
	a2	48.22 ± 1.78	9.00 ± 2.03	620
	a3	53.72 ± 2.09	0.33 ± 0.33	709
	a4	48.00 ± 2.03	10.50 ± 4.23	735
	a5	51.22 ± 1.84	15.17 ± 4.43	709
	a6	46.39 ± 1.95	19.50 ± 8.84	715
	a7	54.67 ± 2.65	14.50 ± 1.87	715
	a8	70.94 ± 2.82	2.50 ± 0.43	921
	a9	69.28 ± 1.74	11.17 ± 8.00	859
	a10	66.94 ± 2.61	4.67 ± 1.93	859
	a11	69.38 ± 1.22	4.17 ± 1.22	859
	a12	72.72 ± 3.22	10.17 ± 3.48	921
	a13	71.50 ± 2.57	6.17 ± 2.07	859
	b1	54.44 ± 2.42	3.83 ± 2.17	623
	c1	51.56 ± 1.44	3.17 ± 1.56	605
	d1	47.17 ± 1.94	20.67 ± 6.36	620

Supplementary Table S5.11 AIC comparison for model selection for *F. japonica* var. *japonica* whole plant maximum light utilisation efficiency of PSII (F_v/F_m) across sites 1-3. The model including the interaction term (DAT * TG) also includes all main effects.

Site	Model	Model deviance	d.f.	AIC	ΔAIC
1	DAT	48184.71	3	889.8	117.0
	TG	75224.39	4	918.7	36.9
	DAT + TG	38708.36	5	838.6	88.1
	DAT * TG	32263.73	7	801.7	0
2	DAT	0.99	3	-14.0	1.9
	TG	0.85	3	-11.2	4.7
	DAT + TG	0.84	4	-15.9	0
	DAT * TG	0.79	5	-13.9	2
3	DAT	48184.71	3	-2222.0	347.1
	TG	75224.39	18	-2111.0	834.1
	DAT + TG	38708.36	19	-2223.6	154.4
	DAT * TG	32263.73	35	-2274.9	0

Supplementary Table S5.12 ANCOVA linear regression results for *F. japonica* whole plant maximum light utilisation efficiency of PSII (F_v/F_m) at site 1: comparison of individual factor level estimates with TG d3. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimates represent the change in slope for that TG compared to DAT value given for TG d3.

Parameter	Estimate	SE	t value	Pr(>|t|)
Intercept	0.732	0.0157	46.7	<0.001
DAT	-0.0001	0.00005	-2.92	0.004
TG CTRL	-0.0060	0.0299	-0.201	0.841
TG d2	-0.0162	0.021	-0.771	0.442
DAT : TG CTRL	0.0001	0.00007	1.95	0.054
DAT : d2	0.000006	0.000006	0.098	0.922

Supplementary Table S5.13 ANCOVA linear regression results for *F. japonica* var. *japonica* whole plant maximum light utilisation efficiency of PSII (F_v/F_m) at site 2: comparison of individual factor levels with the control TG. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimates represent the change in slope for TG d4 compared to DAT value given for the control TG.

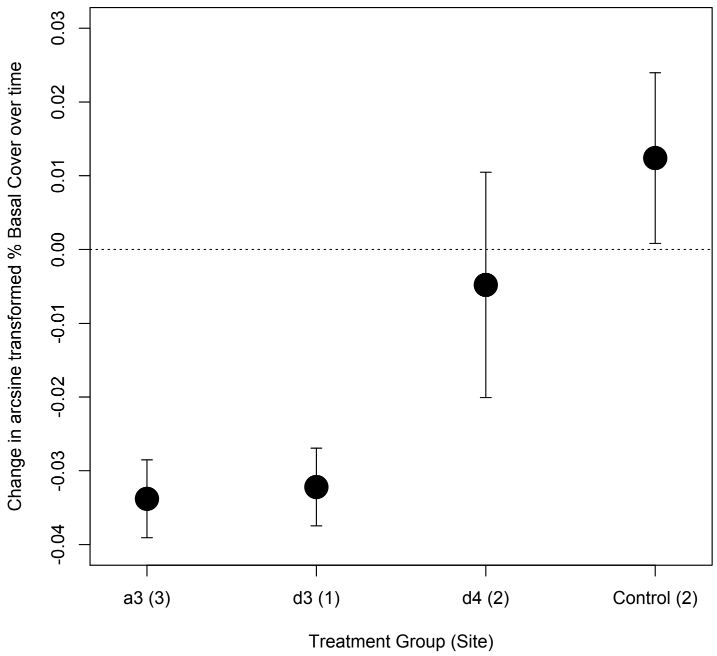
Parameter	Estimate	SE	t value	Pr(>|t|)
Intercept	0.723	0.088	8.224	<0.001
DAT	0.0001	0.0002	0.662	0.514
TG d4	-0.0023	0.1258	-0.018	0.985
DAT : TG d4	-0.0003	0.0002	1.278	0.212

Supplementary Table S5.14 ANCOVA linear regression results for *F. japonica* whole plant maximum light utilisation efficiency of PSII (F_v/F_m) at site 3: comparison of individual factor levels with TG a3. DAT = days after treatment; TG = treatment group; interaction terms are denoted with a colon between variables. Interaction term estimates represent the change in slope for that TG compared to DAT value given for TG a3.

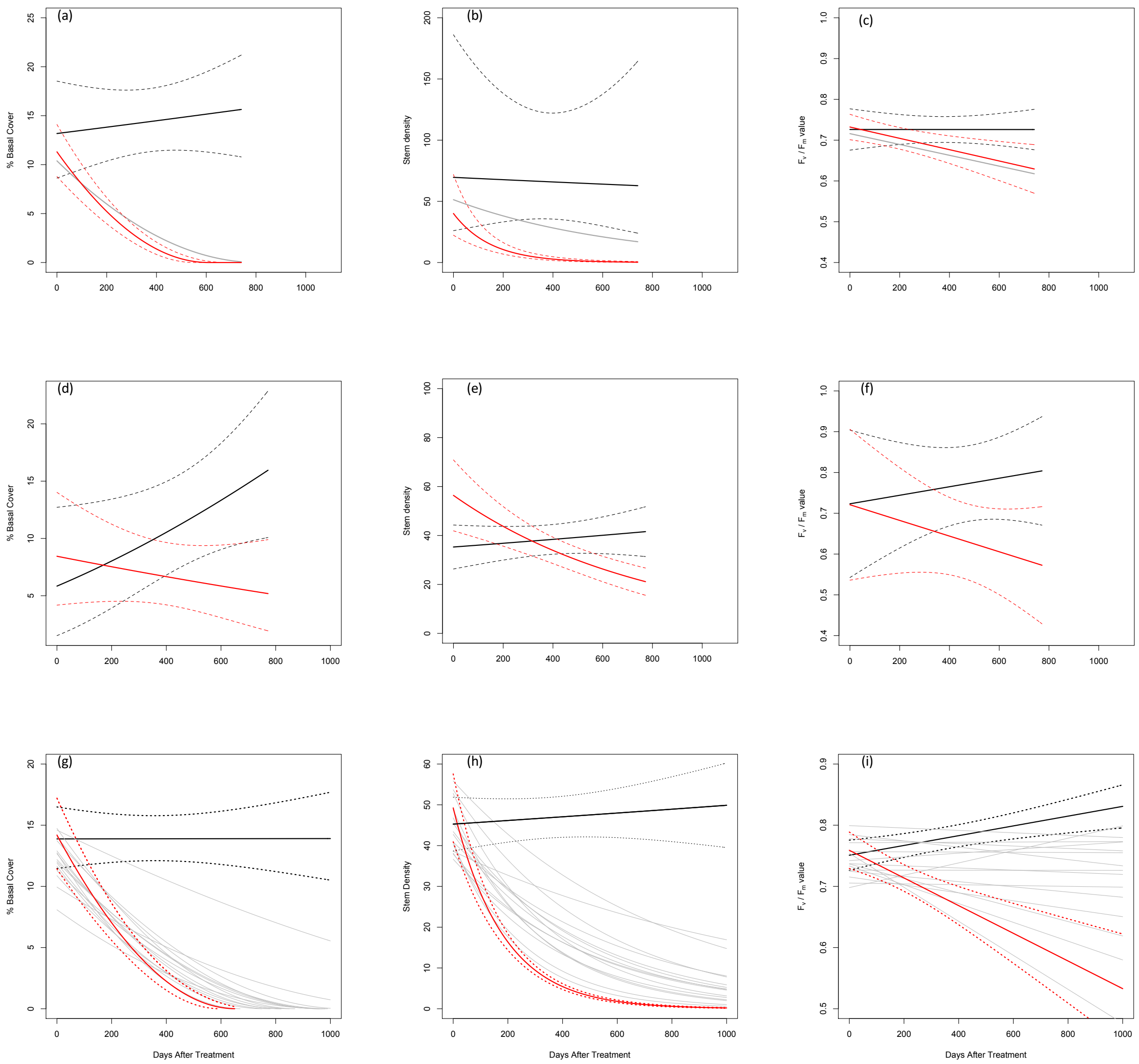
Parameter	Estimate	SE	t value	Pr(>|t|)
Intercept	0.759	0.0152	50	<0.001
DAT	-0.000226	5.46e-05	-4.15	<0.001
TG CTRL	-0.00809	0.0197	-0.411	0.681
TG a1	-0.0536	0.0206	-2.61	0.009
TG a2	-0.0343	0.0206	-1.67	0.096
TG a4	-0.0257	0.022	-1.17	0.242
TG a5	-0.0225	0.0213	-1.05	0.292
TG a6	-0.0436	0.022	-1.99	0.047
TG a7	-0.00494	0.022	-0.225	0.822
TG a8	-0.061	0.0195	-3.13	0.002
TG a9	0.0188	0.0192	0.983	0.326
TG a10	0.0403	0.0192	2.11	0.036
TG a11	0.0255	0.0192	1.33	0.183
TG a12	-0.00219	0.0194	-0.113	0.910
TG a13	0.0123	0.0192	0.644	0.519
TG b1	-0.0325	0.0211	-1.54	0.124
TG c1	-0.0169	0.0205	-0.823	0.411
TG d1	-0.0219	0.0206	-1.07	0.287
DAT : TG CTRL	0.000306	5.96e-05	5.13	<0.001
DAT : a1	0.00022	6.8e-05	3.23	0.001
DAT : a2	0.000184	6.86e-05	2.68	0.008
DAT : a4	7.27e-05	8.45e-05	0.861	0.390
DAT : a5	0.000109	7.42e-05	1.46	0.143
DAT : a6	0.000161	8.51e-05	1.9	0.058
DAT : a7	-5.17e-05	8.51e-05	-0.608	0.543
DAT : a8	0.000327	5.91e-05	5.54	<0.001
DAT : a9	0.000206	5.95e-05	3.47	<0.001
DAT : a10	0.000207	5.95e-05	3.47	<0.001
DAT : a11	0.000175	5.95e-05	2.94	0.003
DAT : a12	0.000224	5.91e-05	3.8	<0.001
DAT : a13	0.000228	5.95e-05	3.83	<0.001
DAT : b1	0.000226	7.05e-05	3.2	0.001
DAT : c1	0.000257	6.86e-05	3.74	<0.001
DAT : d1	0.000209	6.8e-05	3.07	0.002

Supplementary Table S5.15 Initial and final mean F_v/F_m values (± S.E.) values across all replicate plots for each treatment group (TG) at each field trial site. Total length of treatment period in days is given in the Final DAT (days after treatment) column.

Field Site	TG	Initial value	Final value	Final DAT
1	Control	0.73 ± 0.01	0.73 ± 0.01	741
	d2	0.73 ± 0.01	0.65 ± 0.02	593
	d3	0.73 ± 0.01	0.64 ± 0.03	701
2	Control	0.74 ± 0.01	0.81 ± 0.01	722
	d4	0.71 ± 0.02	0.58 ± 0.11	722
3	Control	0.76 ± 0.01	0.86 ± 0.00	1099
	a1	0.75 ± 0.01	0.75 ± 0.02	620
	a2	0.77 ± 0.01	0.80 ± 0.01	620
	a3	0.78 ± 0.01	0.75 ± 0.01	709
	a4	0.73 ± 0.01	0.72 ± 0.01	735
	a5	0.76 ± 0.01	0.79 ± 0.01	709
	a6	0.72 ± 0.01	0.74 ± 0.01	715
	a7	0.75 ± 0.01	0.65 ± 0.02	715
	a8	0.81 ± 0.00	0.83 ± 0.01	921
	a9	0.79 ± 0.01	0.76 ± 0.03	859
	a10	0.81 ± 0.00	0.77 ± 0.02	859
	a11	0.80 ± 0.01	0.77 ± 0.02	859
	a12	0.81 ± 0.01	0.79 ± 0.02	921
	a13	0.80 ± 0.01	0.78 ± 0.01	859
	b1	0.75 ± 0.01	0.80 ± 0.01	623
	c1	0.76 ± 0.01	0.78 ± 0.01	605
	d1	0.77 ± 0.01	0.76 ± 0.01	620



Supplementary Fig. S5.1 Partial estimates (± 95% CIs) from linear models of the change of arcsine transformed % basal cover over time for four treatment groups (a3, d3, d4, Control) in three different field trial sites (1: Lower Swansea Valley Woods; 2: Swansea Vale Nature Reserve; 3: Taffs Well). Treatment groups a3 (chemical control only) and d3 (combined physiochemical control) perform similarly well and both reduce basal coverage significantly more than d4 (physical control only – covering), which showed no change in basal coverage over time. The no-treatment Control at site 2 showed an increase in % cover over time. See Table 1 for specific details of each treatment applied and Tables S5.1, 5.6 and S5.11 for further details of the linear models.



Supplementary Fig. S5.2 Best performing *F. japonica* treatment group at each site (a-c: Lower Swansea Valley Woods; d-f: Swansea Vale Nature Reserve; g-i: Taffs Well), for each response variable (% basal cover, stem density and light utilisation efficiency). Lines show model predicted values for the effects of each different treatment group over time. Solid black lines show values from control plots (no treatment applied). Red lines show results from the best overall performing treatment group at each site (d3: Lower Swansea Valley Woods; d4: Swansea Vale Nature Reserve; a3: Taffs Well). Grey lines show all other treatment groups. Dashed lines indicate 95% confidence intervals (CIs) for control and the best overall performing treatment group at each site. Linear model predicted values for arcsine transformed % basal cover were back transformed for presentation in (a, d, and g), Negative Binomial GLM values were used in (b, e and h) and untransformed linear model values used in (c, f and i). See Table 1 for specific details of each treatment applied. Coefficient estimates for all treatments are given in Tables S5.2-5.4, S5.7-5.9 and S5.12-S5.14.