Variation in ripening years of seed cones of *Juniperus communis* L.

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ABSTRACT

Many *Juniperus communis* populations are in decline, often with poor reproductive performance. Biological and phenological studies have shown that seed cone ripening time takes between two and three summers after pollination, and occasionally four. Individual junipers differ: in samples from northern England some were almost entirely three year, some were two year and others had mixed proportions. The limited evidence suggests that two year ripening is commoner in warmer areas such as southern England, and three years more frequent in northern and mountainous parts of Europe. Suggestions about the causes of these differences include the effects of climate on pollen tube growth rates, phasing of the seed cone crop production and genetic background.

KEYWORDS: Berries, Maturation, Phenology, Reproduction, Fixed-point photography.

INTRODUCTION

The failure of regeneration in *Juniperus communis* L. in various areas of lowland Europe has been of concern for many years (Ward 1973; Burny 1985; Lejeune *et al*. 1986; Landolt 1994; Clifton *et al.* 1997; García *et al.* 1999; García & Zamora 2003; Verheyen *et al.* 2005). Some of the declines can be attributed to modern land use which no longer provides the necessary open nutrient-poor habitat conditions for seedlings. This may be compounded by heavy grazing by domestic animals or rabbits (Ward & King 2006). Additionally seed production can be very poor (García *et al*. 2000; Verheyen *et al.* 2009). The ecological and biological background to seed production is not completely understood however, and as part of a detailed study, this paper shows how sites and individuals may differ in seed cone (berry) ripening times.

Juniperus communis L. is wind-pollinated in late spring, and seed cones (Farjon 2005) ripen over two or three years (Adams 2004; Thomas *et al*. 2007; Tutin *et al.* 1993). Three year cycles have been recorded more often, with seed cones remaining small until fertilized in the following spring (Ottley 1909; Stiles 1980; Roques 1983; Raatikainen & Tanska 1993; Chambers *et al*. 1999; García *et al*. 2002). These small green cones swell in their second summer and ripen to purple by the end of the third summer. However cones can ripen over two years (Tueller *et al*. 1975: Farjon 2005) and in southern England Ward (2007) found a correlation between abundant male pollen in spring the year before good cropping in females, but not two years before.

As this variation in ripening year was little understood, cones were counted on wood of different ages, contrasting two southern and one northern site in the UK. An earlier phenological study in southern England in 1988–1989 had utilized fixed point photography of twigs, but it is only now with the use of digital scanning that it has become practical to count the individual cones and to present relevant data about the cone life cycles.

The results will show whether the life cycles of seed cones lasted for two or three years (or more) and whether there were differences between individuals and sites. Some possible reasons for differences will be discussed.

MATERIALS AND METHODS

STUDY SITES

Samples for seed cone age studies were taken from (i) The Breck, Porton Ranges of the Defence Science Technology Laboratory (DSTL) on the Hampshire/Wiltshire border in southern England (Nat. Grid Ref. SU2103700) on 6/10/2006, a site where juniper populations have been monitored since 1983 (Ward 2007), (ii), Bulford Down SSSI (Wiltshire SU204443) on 16/8/2006, with junipers of the same age class as on the Breck (i.e. both colonized after myxomatosis destroyed browsing rabbits in 1954–1955), (iii) Mardale Banks in northern England in Cumbria (NY481128) on 17/10/2006.

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TABLE 1. SEED CONE TYPES RECOGNIZED IN THIS STUDY

Key	Colour	Size	Ripening stage
F	Green	very small	Conelets/'flowers' – receptive/just pollinated
GV	Green	small	Conelets after pollination but little swelling
G	Green	normal	immature unripe cones
PG	Purple/Green	normal	Colour change to purple ripe beginning
P	Purple	normal	Ripening/ripe (fully ripe when bloom appears)
Y	Yellow	normal	Abortion/dying (all stages except brown)
B	Brown	over-ripe	not dispersed or fallen
S	Shrivelling	all sizes	Shrivelling all ages, diseased or seeds not fertilized

The phenological study in 1988–1989 was of a young healthy population (modal age about 21 years) at Pepperbox Hill in the Brickworth Down and Dean Hill SSSI (Wiltshire SU212247).

On all the sampling sites there were more males than females, the sexes were growing close together, and the weather in spring was dry, so that pollination problems were unlikely to be limiting (Ward 2007).

WOOD AGE AND CONE DEVELOPMENT

At each of the three study sites, ten twigs of 15–25 cm with >75 green (immature) or purple (mature) cones were cut from ten individuals. The numbers of growing shoots over the last three years were counted, and cones assigned to the age of the wood to which they were attached, using the annual bud scars/scales and comparative leaf lengths (leaves are short near the annual scar). Some cone-bearing shoots stop growing, and their age was harder to assess, but could usually be determined by comparison to nearby similar shoots. Cones were assigned to different types (Table 1). Filled white seed, and empty and dead seeds and insect attack were also counted, and these results will appear elsewhere.

FIXED POINT PHOTOGRAPHY OF SMALL BRANCHES

At Pepperbox Hill a branch (c. 20–40 cm) with some purple cones was selected on each of 12 individuals, and these were subsequently photographed on each of 14 monthly visits from August 1988 to November 1989. 1988: 8/8, 14/9 (Oct missed), 11/11, 14/12. 1989: 18/1, 14/2, 29/3 (April missed), 5/5, 8/6, 3/7, 16/8, 12/9, 19/10, 28/11. Owing to the growth of foliage in the second year, only seven branches provided photographs clear enough to be scanned for counting the same cones over all sampling dates. On some months cones were obscured because of slightly different photographic angles, but were visible in later scans and could be added back into the data. Similarly, more of the small undeveloped conelets were not seen until they started to swell, and these counts were therefore underestimated.

RESULTS

PHENOLOGY OF SEED CONE DEVELOPMENT

The detailed study at Pepperbox Hill shows the differing phenology during the ripening of two and three year seed cones (Fig. 1), using total counts of the numbers in the sampled seven branches (Fig. 2).

The very small seed conelets became receptive in late April and May when a small liquid drop (pollination droplet) appeared at the tip of the cone. Cones that will last for only two years swelled in June but many of these small conelets aborted and were shed.

FIGURE 1. (opposite) Seed cone ripening variation in *Juniperus communis* at Pepperbox Hill (Seed cone types Table 1). 1–4 in left column on two year cycle. 1. May – receptive conelets (F) on last year's wood. 2. June – conelets swelling (GV), some aborting/falling. 3. July – normal green cones (G) in first year, others shrivelling (S). 4. November – green cones (G) typical size,and a two year purple cone (P). 5–6 in right column mixed two and three year cycles. 5. May – conelets/small cones (F and GV) from last spring swelling on last year's wood, new current year conelets visible. 6. June – two year green cones (G) slightly larger and darker than the one year, many aborting cones (GYS). 7. July – some one year cones swollen (GV) others remain as conelets (F). 8. November – ripe cones purple (P-second and ?third years), green cones (G-first and second years) and a few conelets (GV).

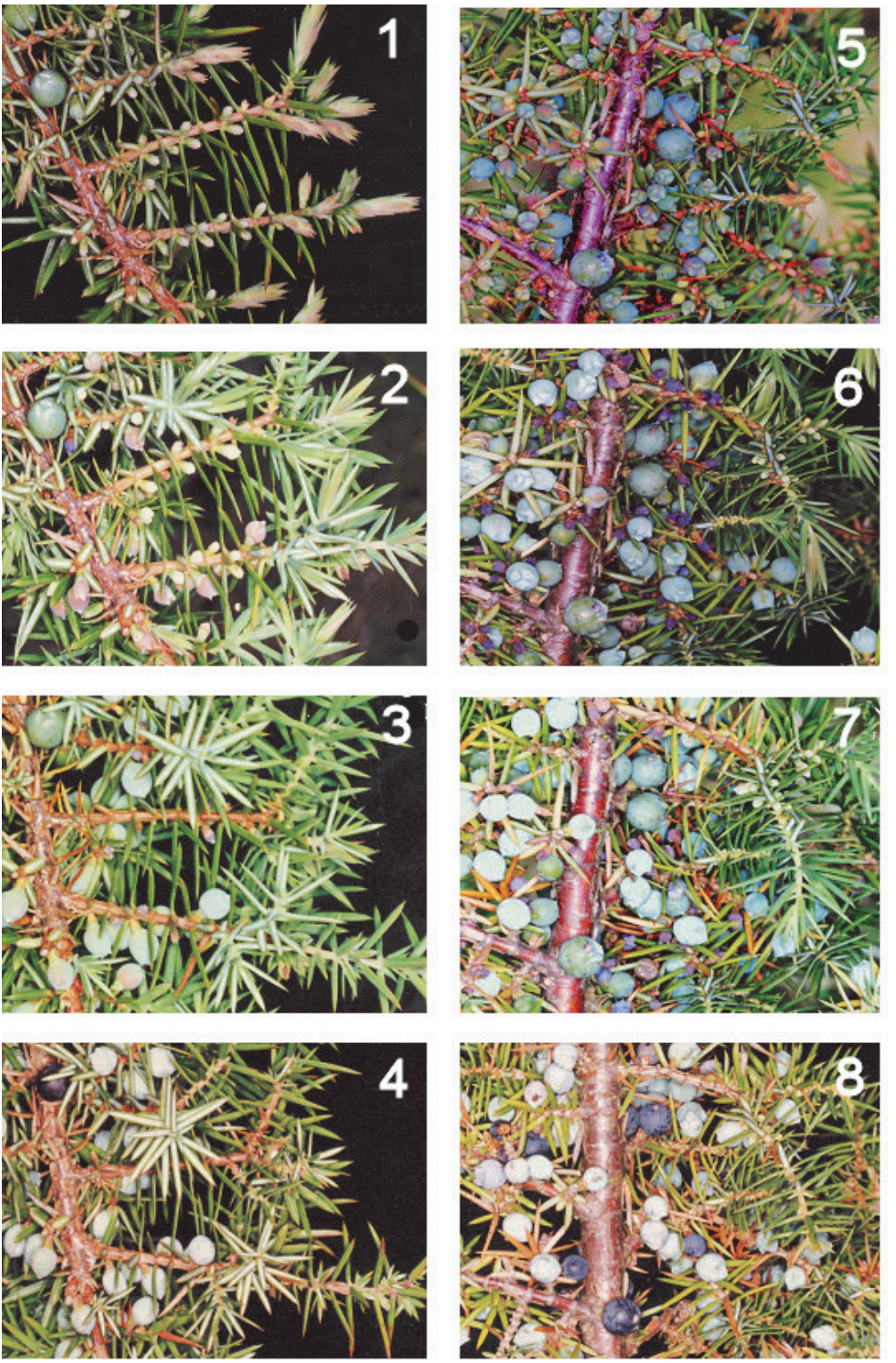

FIGURE 1.

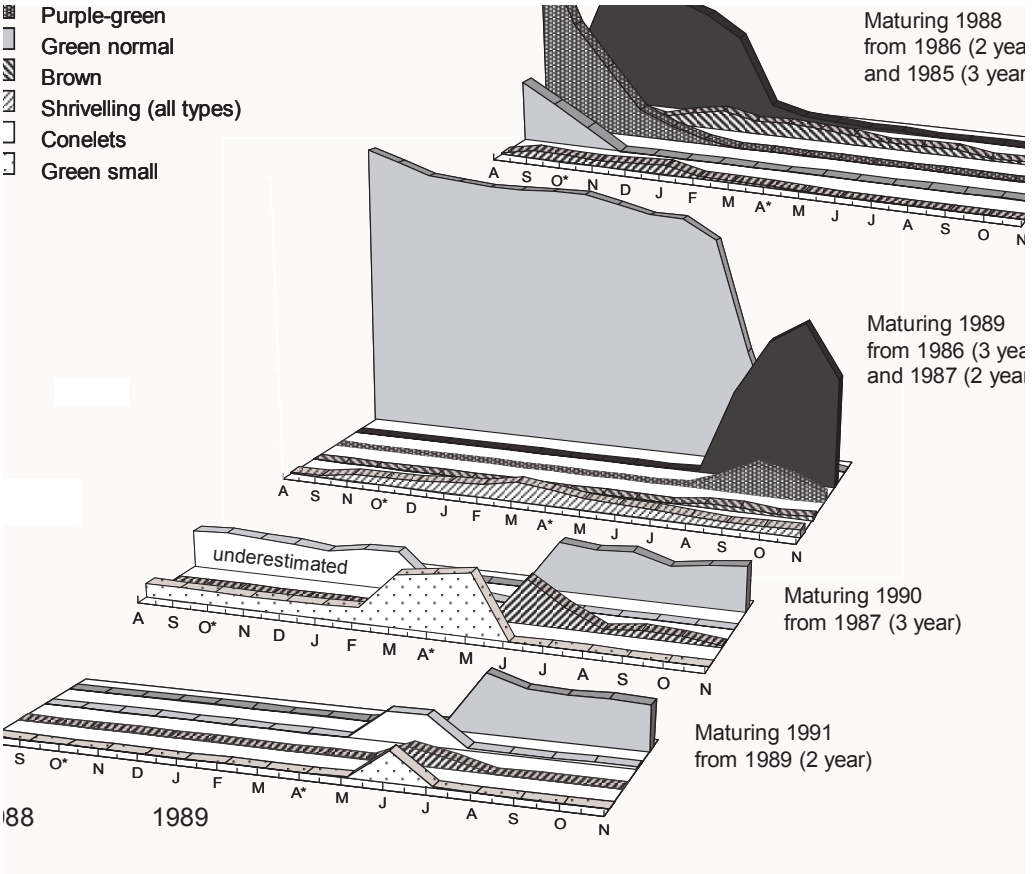

FIGURE 2 Monthly total counts of all seed cone types with years of pollination and ripening at Pepperbox Hill from fixed point photographs (Counts on a scale 0–80 cones on seven twigs from different individuals: no three year old cones to ripen in 1991: actual dates 1988 – 8/8, 14/9 (Oct missing) 11/11, 14/12; 1989 –18/1, 14/2, 29/3, (April missing) 5/5, 8/6, 3/7, 16/8, 12/9, 19/10, 28/11): * missing values interpolated).

The surviving green cones swelled to a normal size by July/August. Cones that will last for three years developed slower, the tiny conelets hardly changed or they swelled slightly and remained much smaller than the normal larger green cones until about March of the following year. These one year old conelets began to swell just before the current year's conelets became receptive for pollination. In May and June quite a lot of these one year old conelets aborted, possibly due to lack of fertilization, and these shrivelled conelets were more conspicuous than the aborting conelets of the current year. By July of the second year these one year old cones looked like those that had swollen in the same year, but they were located on two year old wood. There were usually more second and third year green cones than purple, but some of the green cones shrivelled, mainly in spring and early summer, because the seeds were unfertilized, aborted or were attacked by the juniper berry gall mite, *Trisetacus quadrisetus* (Thomas). Green cones enlarged slightly more at the beginning of their last season (observed also by Raatikainen & Tanska 1993), and in late summer or autumn usually became purple-green blotched, before turning to dark blue/purple, at times varying between individuals (Fig. 3). Cones were progressively shed or eaten by birds etc from September onwards, although a few persisted through the winter, eventually turning brown and shrivelling. A small proportion of these were serotinous with some filled white seeds (presumed viable).

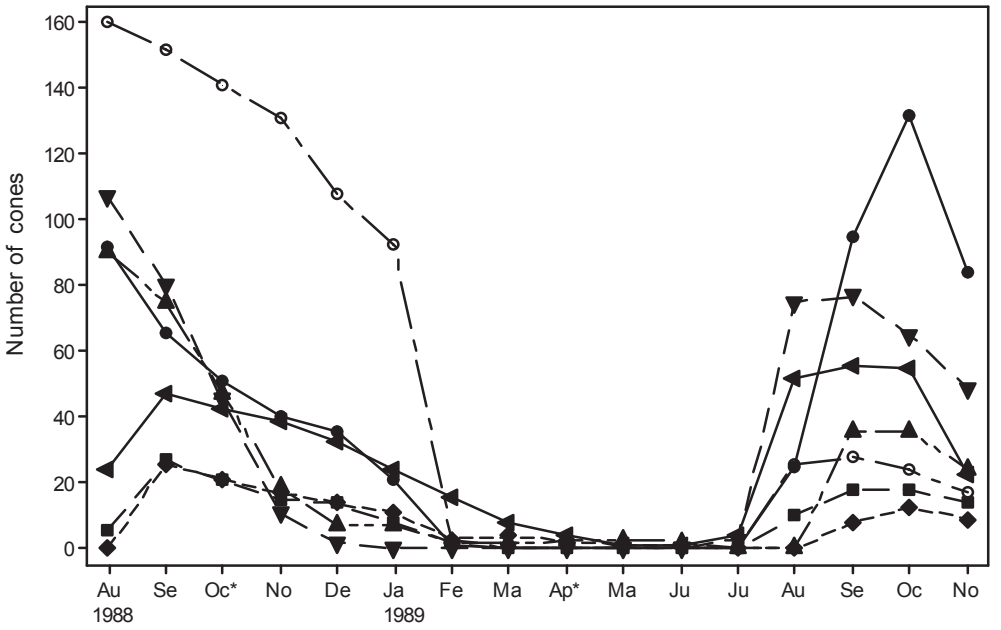

FIGURE 3. Timing of ripening and seed cone disappearance on seven individuals, with differences in successive years at Pepperbox Hill 1988–1989 (* = missing values interpolated).

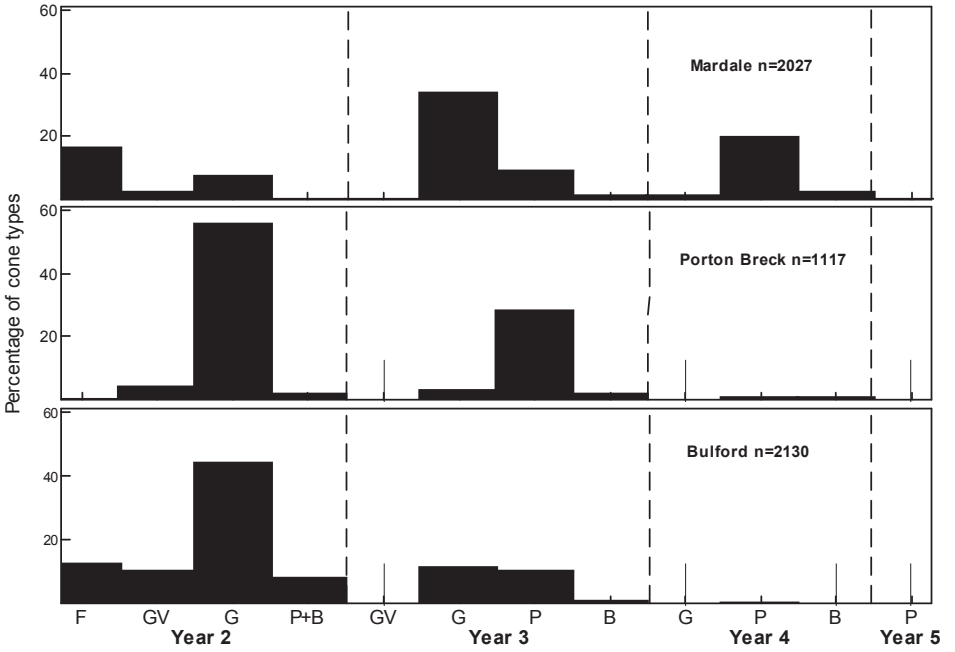

FIGURE 4. Percentage of total numbers of seed cone types on the years of growth of 10 sample twigs from each of Mardale, Porton Breck and Bulford in 2006 (see Table 1 for key to cone types).

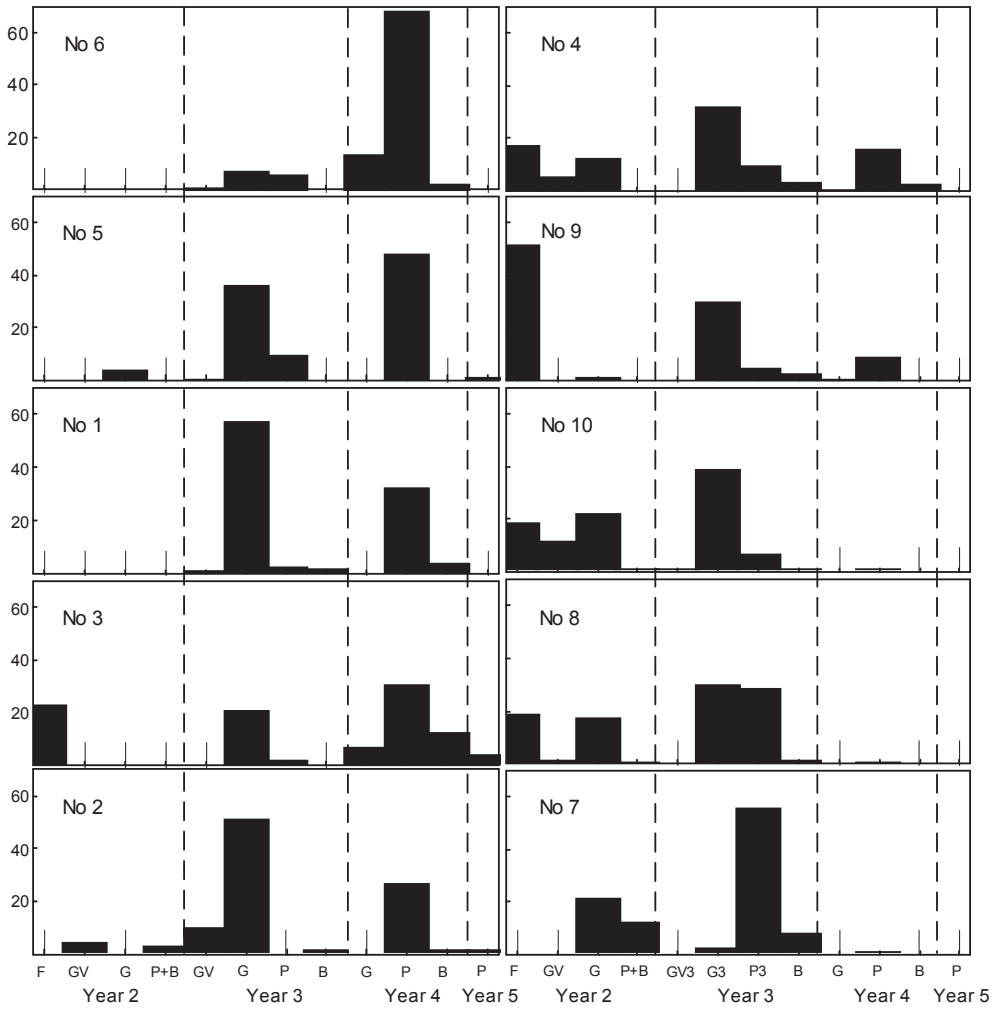

FIGURE 5. Percentage of seed cone types on the last five years of growth on 10 individual junipers - arranged from three year ripening top left to two year at bottom right (for cone types see Table 1).see

As the study progressed it was realized that the phenology of the year classes would be incompletely represented in Fig. 2 because the green and purple cones present at the start in 1988 could not be divided into their year classes. There were more two year cones than three in those that could be distinguished by the first year, and in the last summer of the recording there were apparently no three year cones to mature as the smaller numbers of green cones had swollen in their first year.

SEED CONE RIPENING TIME DIFFERENCES – NORTH AND SOUTH ENGLAND

The results from Pepperbox Hill first showed that the ripening period for seed cones of *Juniperus communis* varied. In order to understand this cone development in relation to years more clearly, cones of different colours and stages of development were assessed by wood age from additional sites. At two southern sites, Porton and Bulford, almost all cones ripened over two years (Fig. 4) only 1·5% and 0·5% took three years. In contrast at the northern site, Mardale, 20·3% of cones ripened in their third year and 1·3% took four years to mature. All cones originated from the reproductive buds on second year wood, and at Mardale 16·9% of very small conelets for this first year appeared healthy and were expected to swell the following year reaching maturity in their third year. At Bulford there were 12·6%

of these small conelets, but nearly all were in poor condition (turning purple, shrivelling or damaged by Eriophyid mites), and so were expected to fall very soon. At Porton there were no small conelets, probably because the sampling date was slightly later (October) and these unhealthy conelets had already fallen. The ripening years of seed cones at Mardale varied in individual junipers (Fig. 5). Cones on No. 7 for example were two years and similar to those from the southern samples, while No. 1 had cones that nearly all matured over three years, as did No. 5 which also had a few purple cones of four years. It was not known how differences in ripening time affected survival of the seed cones, but the numbers of white, filled seed at Mardale was significantly higher in the green cones on the two year old wood (43·86%, n=579 seeds) than in those on the three year wood (30·29%., n=1875) (Chi-Sq = 17·334, DF = 1, P-Value = 0·000). There was no difference in seed numbers between ripe purple cones on three and four year old wood (19·6% and 19·3%).

DISCUSSION

The results show that the ripening period for seed cones of *Juniperus communis* varied between two and three years, with a few cones taking four years. Two southern sites in the UK were very strongly biased to two years, although a very young vigorous population at Pepperbox Hill had a slightly higher proportion of three years. At other southern sites the author has also noticed that the two year cycle is normal. The northern site (Mardale) had both two and three year cycle cones, but there were marked variations between individuals, some entirely following the two year pattern, while others were predominantly of three years. The limited evidence in this paper and in the literature (Lanzara & Pizzetti 1977; Raatikainen & Tanska 1993; García *et al*. 2002) suggests that three year cycle cones are more frequent in northern or mountainous locations in Europe than in warmer areas. After pollination, the pollen tube grows down to the ovule and fertilization can be delayed for up to a year (Ottley 1909; Schnarf 1933; Singh 1978). The growth of the pollen tube could therefore be a critical factor in ripening in relation to fertilization as this seems to trigger the swelling of the cone in either the first or second year. Indeed in various plants it is known that pollen tube growth increases with temperature (Bertin 1988; Murcia 1990; Hedhly *et al*. 2005). In *J. communis* also seed viability decreases in warmer latitudes and with higher nitrogen content (Verheyen *et al*. 2009) and these variables affect the physiological and nutritional state of the ovule which is also important in reproductive biology.

Genotypes within plant species affect pollination and fertilization and are important in adaptation to environmental differences. *J. communis* is a very variable species (Van der Merwe *et al*. 2000, Oostermeijer & de Knegt 2004, Michalczyk *et al*. 2006), and has two main sub-species in England (Tutin *et al*. 1993, Thomas *et al*. 2007) although recent DNA analyses indicate that these are not clearly distinguishable and should be varieties (Adams & Pandey 2003; Filipowicz *et al*. 2006). Foliage at the northern study site was variable and some individuals tended to have smaller and more appressed leaves and slower growth, perhaps more like that of *J. communis* L. ssp. *nana* and these were apparently more likely to bear three year ripening seed cones. Intermediates between the sub-species *J.communis communis* L and *J. communis nana* (Hook.) Syme are frequent (Stace 1977) and in Cumbria there could be an altitutindal cline, so that Mardale at an altitude of 459 m could be expected to show some variation. Relatively little is known about the phasing of seed cone production in *J. communis*, although good crops in females at Porton were more often followed by a poorer crop and greater growth of the foliage than in males, and females only cropped well on average for 3·1 years (Ward 2007). Discontinuous production of cones is suggested in the phenological study at Pepperbox Hill where the sample branches in fixed photographs were initially chosen to have many cones, but by the second year most had far fewer cones on the same branches and there were fewer three year cycle cones. Probably there is competition between growth and cone production for nutrient resources, and up to a point this affects the whole individual and not just branches. It is quite common to see an individual with mostly green immature cones (at the start of a productive cycle) while another nearby has only purple cones. Possibly if the initial phase is vigorous with very many cones, as in young plants, there is a higher probability of 3 year cycles due to resource competition. This might explain the greater numbers of smaller cones which swelled slightly but not to normal size.

The timing of cone ripening in autumn is important in seed dispersal by birds, and especially to the migrant flocks of fieldfares (*Turdus pilaris*) and other Turdidae attracted to larger populations of juniper (Garcia *et al*. 2001). The blue waxy bloom that appears on mature cones has UV reflectance in *Juniperus virginiana* L. and is thought to be a signal making the cones more visible to birds (Burkhardt 1982, Willson and Whelan 1989). Although most cones are ripe for dispersal in September individual junipers vary so some cones are available to birds throughout the winter months.

The complexity of seed cone ripening years in *J. communis* will need to be taken into account in studies of its ecological and reproductive biology, for example, it is not known how differences in ripening time affect the annual crop of seed cones. At Mardale there were more filled white seeds in the green seed cones on two year old wood than on three year old wood, but there was no difference in filled seed numbers in the ripe purple cones on three and four year old wood.

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REFERENCES

ADAMS, R. P. & PANDEY, R. N. (2003). Analysis of *Juniperus communis* and its varieties based on DNA fingerprinting. *Biochemistry Systematics and Ecology* **31**: 1271–1278.

ADAMS, R. P. (2004). *Junipers of the world: The genus* Juniperus. Trafford Publishing Co., Vancouver.

BERTIN, R. I. (1988). Paternity in plants. in LOVETT DOUST, J. & LOVETT DOUST, L., eds. *Plant Reproductive Ecology. Patterns and Strategies* pp. 30–59. Oxford University Press, Oxford.

BURKHARDT, D. (1982). Birds, berries and UV. A note on some consequences of UV vision in birds. *Naturwissenschaften* **69**: 153–157.

BURNY, J. (1985). Het vroeger en huidig voorkomen van de Jeneverbes *Juniperus communis* L. op de Hoge Kempen (Provincie Limburg, Belgie). *Wielewaal* **51**: 10–30.

CHAMBERS, J. C., VANDERWALL, S. B. & SCHUPP, E. W. (1999). Seed and seedling ecology of piñon and juniper species in the pygmy woodlands of western North America. *Botanical Review* **65**: 1–38.

CLIFTON, S. J., WARD, L. K.& RANNER, D. S. (1997). The status of juniper *Juniperus communis* L. in north-east England. *Bioloical Conservation* **79**: 67–77.

FARJON, A. (2005). *A monography of Cupressaceae and Sciadopitys*. Royal Botanic Gardens, Kew.

FILIPOWICZ, N., PIOTROWSKI, A, OCHOCKA, J. R. & ASZTEMBORSKA, M. **(**2006). The phytochemical and genetic survey of common and dwarf juniper (*Juniperus communis* and *Juniperus nana*) identifies chemical races and close taxonomic identity of the species. *Planta Medica* **72**: 850–853.

GARCÍA, D.(2001). Effects of seed dispersal on *Juniperus communis* recruitment on a Mediterranean mountain. *Jounal of Vegetation Science* **12**: 839–848.

GARCÍA, D. & ZAMORA, R. (2003). Persistence, multiple demographic strategies and conservation in long-lived Mediterranean plants. *Journal of Vegetation Science* **14**: 921–926.

GARCÍA, D., ZAMORA, R., GÓMEZ, J. M. & HÓDAR, J. A. (2001). *Juniperus communis* depends more on population characteristics than on individual attributes. *Journal of Ecology* **89:** 639–647.

GARCÍA, D., ZAMORA, R., GÓMEZ, J. M. & HÓDAR, J. A. (2002). Annual variability in reproduction of *Juniperus communis* L. in a Mediterranean mountain: Relationship to seed predation and weather. *Ecoscience* **9**: 251–255.

GARCÍA, D. *et al*. (2000). Geographical variation in seed production, predation and abortion in *Juniperus communis* throughout its range in Europe. *Journal of Ecology* **88**: 435–446.

GARCÍA, D., ZAMORA, R., HÓDAR, J. A., & GÓMEZ, J. M. (1999). Age structure of *Juniperus communis* L. in the Iberian peninsula: Conservation of remnant populations in Mediterranean mountains. *Biological Conservation* **87**: 215–220.

HEDHLY, A., HORMAZA, J. I. & HERRERO, M. (2005). The effect of temperature on pollen germination, pollen tube growth, and stigmatic receptivity in peach. *Plant Biology* **7**: 476–483.

LANDOLT, E. (1994). Beitrage zur der Flora der Stadt Zürich. I Einleitung-Beschreibung der neuen Flora, Pteridophyten und Gymnospermen. *Botanica Helvetica* **104**:157–170.

LANZARA, P. & PIZZETTI, M. (1977). *Simon and Schuster's Guide to Trees*. Simon and Schuster Inc., New York.

LEJEUNE, M., VERBEKE, W.& HEYVAERT, J. (1986). Jeneverbes (*Juniperus communis* L.) op de Sint-Pietersberg (Provincie Luik, België). *Natuurhistorisch Maandblad* **75**: 63–68.

MICHALCZYK I. M. *et al*. (2006). Characterization of highly polymorphic nuclear microsatellite loci in *Juniperus communis* L. *Molecular Ecology Notes* **6**: 346–348.
MURCIA, C. (1990). Effect of floral morphology and temperature on pollen receipt and removal in *Ipomoea trichocarpa*. *Ecology* **71**: 1098–1109.
OOSTERMEIJER J. G. B. & DE KNEGT B.(2004). Genetic population structure of the wind-pollinated, dioecious shrub *Juniperus communis* in fragmented Dutch heathlands. *Plant Species Biology* **19**: 175–184.
OTTLEY, A. M. (1909). The development of the gametophytes and fertilization in *Juniperus communis* and *Juniperus virginiana*. *Botanical Gazette* **48**: 31–46.
PRESTON, C. D., PEARMAN, D. A. & DINES, T. D. (2002). *New Atlas of the British and Irish flora.* Oxford University Press, Oxford.
RAATIKAINEN, N. & TANSKA, T. (1993). Cone and seed yields of the juniper (*Juniperus communis*) in southern and central Finland. *Acta botanica. Fennica* **149**: 27–39.
ROQUES, A. (1983). *Les insects ravageurs des cônes et graines de conifères en France* Institute National de laRecherche Agronomique (INRA), Paris.
SCHNARF, K. (1933). *Handbuch der Pflanzenanatomie*, Bd. X2: Embryologie der Gymnospermen. Gebrüder Borntraeger, Berlin.
SINGH, H., (1978). *Embryology of Gymnosperms*. Gebrüder Borntraeger, Berlin.
STACE, C. (1977). New Flora of the British Isles. Second Edition. Cambridge University Press, Cambridge.
STILES, E.W. (1980). Patterns of fruit presentation and seed dispersal in bird-disseminated woody plants in the eastern deciduous forest. *American Naturalist* **116**: 670–688.
THOMAS, P. A., EL-BARGHATHI, M. & POLWART, A. (2007). Biological Flora of the British Isles: *Juniperus communis* L. *Journal of Ecology*: **95**: 1404–1440.
TUELLER, P. T. & CLARK, J. E. (1975). Autecology of pinyon-juniper species of the Great Basin and Colorado Plateau, in GIFFORD, G. F.& BUSBY, F. E. eds. *The pinyon-juniper ecosystem: a symposium* pp. 27–40. Utah Agricultural Experimental State Publication, Logan.
TUTIN, T. G. *et al*., eds. (1993). *Flora Europaea* 1: Cambridge University Press, Cambridge.
VAN DER MERWE, M., WINFIELD, M. O., ARNOLD, G. M. & PARKER, J. S. (2000). Spatial and temporal aspects of the genetic structure of *Juniperus communis* populations. *Molecular Ecology* **9**: 379–386.
VERHEYEN, K., SCHREURS, K., VANHOLEN, B. & HERMY, M. (2005). Intensive management fails to promote recruitment in the last large population of *Juniperus communis* L. in Flanders (Belgium). *Biological Conservation* **124**: 113–121.
VERHEYEN, K. *et al*. (2009). *Juniperus communis*: victim of the combined action of climate change and nitrogen deposition? *Plant Biology*. Online publication date: 1-Jun-2009.
WILLSON, M. F. & WHELAN, C. J. (1989). Ultraviolet reflectance of fruits of vertebrate-dispersed plants. *Oikos* **55**: 341–348.
WARD, L. K. (1973). The conservation of Juniper I Status of juniper in southern England. *Journal of applied Ecology* **10**: 165–188.
WARD, L. K. (1977). The conservation of juniper; the associated fauna with special reference to southern England. *Journal of applied Ecology* **14**: 81‑120.
WARD L. K. & KING, M. (2006). Decline of juniper in Sussex. *Quarterly Journal of Forestry* **100**: 263–272.
WARD, L. K. (2007). Lifetime sexual dimorphism in *Juniperus communis* L. var. *communis*. Plant Species Biology **22**:11–21.

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