

Comparison between Iraqi Jenoubi heifers and bulls fed on date palm pollen to accelerate puberty and sexual maturity

H H H AL-ABBASI¹✉, A S MAHDI¹, I H S ALKAFAJY¹ and A A M AL-WAZEER¹

University of Kufa, Najaf, Iraq

Received: 7 July 2022; Accepted: 26 September 2022

ABSTRACT

The objective of the current study was to compare puberty and sexual maturity in Iraqi Jenoubi bulls and heifers that were fed date palm pollen (DPP) in their diets. Ten Jenoubi heifers and ten bulls, 6-month-old were divided randomly into two treatments: DPP treatment (T1) supplemented with 2 g of DPP/ head/day and control treatment (T2) was not supplemented with DPP. In heifers, the overall mean value of control group was significantly lower (27.53, 0.96 ng/ml and 9.44 pg/ml) than treatment group (70.42, 18.27 ng/ml and 19.16 pg/ml) for FSH, LH and estrogen, respectively in puberty period. Similar trend was noticed for hormone concentrations (FSH, LH, Estrogen and Testosterone) for bulls at puberty and maturity age. In bulls, T1 was significantly higher (155 and 212.5 kg) than control group (139.5 and 196 kg) for body weight in puberty and maturity age respectively, and in heifers T1 was significantly higher (150.33 and 206.66 kg) than control group (142 and 198.66 kg) for body weight in puberty and maturity age respectively, and same trend was observed for body condition score (BCS). It can be concluded that adding date palm pollen to ration of heifers and bulls accelerates puberty and maturity in heifers and bulls. But the acceleration in heifers was more than in bulls.

Keywords: Date palm pollen, Jenoubi heifers, Hormones, Maturity, Puberty

Date palm (*Phoenix dactylifera*) is a monocotyledon, belonging to the palm tree family Arecaceae (Coryphiodeae). Date palm is traced to the gender Phoenix which includes many species (Daoud *et al.* 2019). Date palm pollen (DPP) is one of the most important by-products of date palm trees (Araújo de Oliveira *et al.* 2021). Several reports revealed that DPP showed anti-inflammatory activity and increases immunity (Elberry *et al.* 2011). DPP contains estradiol which activates the pituitary (Hassan *et al.* 2021, Bruinje *et al.* 2021) and is used as feed additive as it contains nutrients including essential and non-essential amino acids, fatty acids, proteins, carbohydrates, vitamins and minerals (Hassan 2011). DPP has positive effects on infertility in males and females (Mohamed *et al.* 2018).

After the birth of a heifer, the ovary produces enough ova and sex hormones to allow the heifer to get pregnant. It wasn't a sudden occurrence. Instead, the endocrine and reproductive systems of the female were gradually preparing for reproduction (Severino-Lendecky *et al.* 2020). Heifers are typically pubertal around 12 months (Cardoso *et al.* 2018) and reach maturation when their weight is about 65% of adult weight and mature at 65% of adult weight, while mature cows, whose maturation was discovered during the first pregnancy, are around

18 months old (Stockton *et al.* 2012). The age and weights of different breeds varied enormously (Gasser *et al.* 2006). Due to the economic impact, maturity of heifers was a major consideration (Parkinson 2019). Feed intake and sex development were strongly linked (Bachtiar and Udin 2019). In beef cattle, the heifer is not reaching the complete adult weight unless after 30 months of first breeding cycle, at about 4-5 years old age (Brickell *et al.* 2009). In the absence of an accurate weight for mature heifers, researchers make educated guesses about a cow's subcutaneous fat content based on herd average weight, maturity index, and BCS scores (Dickinson *et al.* 2019). Hence, an attempt was made to investigate the potential to accelerate puberty and maturity age of Iraqi Jenoubi heifers and bulls by palm date pollen addition.

MATERIALS AND METHODS

Study site and ethical issue: This study was conducted in private small holder farm at Al-Abassiya district, Najaf Governorate, Iraq. Al-Abassiyawassituatedat32°05'21.8"N latitude and 44°30'08.5" E longitude. The laboratory tests were conducted at laboratories at University of Kufa, Najaf, Iraq. All the protocol and analysis in this study was approved by Bioethics Committee at University of Kufa, Iraq (No. 2940/2022).

Experimental animals and diet: Ten Iraq Jenoubi heifers and ten bulls of 6 months age were randomly distributed into two groups (average body weight 98.66 and 101.33 kg

Present address: ¹Department of Animal Production, Faculty of Agriculture, University of Kufa, Najaf, Iraq. ✉Corresponding author email: hasanh.alabbasi@uokufa.edu.iq

for group 1 and 2, respectively). First group containing 5 heifers and 5 bulls were fed the basal ration rice straw 49%, green forage 25%, concentrated ration 25% and 1% salt and vitamins plus DPP supplementation (collected from local date palm trees) @ 2 g/head/day, this ration may be changed as necessary weekly till puberty. Second group containing 5 heifers and 5 bulls were fed with same ration but without supplementation of DPP and was regarded as control group.

Blood collection and hormones assay: Blood samples were collected three times [at first day of study (prepubertal period), pubertal and at puberty periods] to determine the effect of DPP on concentrations of FSH, LH, Estrogen and Testosterone from the jugular vein of all animals in non-EDTA tubes. Sera samples were harvested by centrifugation 3000 rpm for 15 min at 4°C and transported under cooling to the laboratory. The quantitative measurement of FSH, LH, Testosterone and Estrogen in serum using mini-VIDS® Analyzer which was used for the enzyme linked fluorescent assay (ELFA) technique, following the instruction in the mini-VIDS® fertility kit (BioMerieux-France).

Body weight and body condition score: All heifers and bulls were weighed three times at the time of blood collection. Body condition score was determined before feeding and drinking of water, heifers scoring from 1-5 score by inspection, and regarded the grade 1 for the thinner heifers and grade 5 for the fitter heifers, the scoring was performed at the same three times of weight measurement.

Statistical analysis: Data were analyzed statistically as a completely randomized design with repeated measures using the PROC MIXED procedure of SAS (2012) to compared the effect of dietary addition of DPP on advance of puberty and sexual maturity in Iraqi Jenoubi heifers and bulls. The pair-wise comparison of means was carried out using post-hoc Duncan multiple comparison test (Duncan 1955). Means expressed as means±SD. Significance declared at the level of probability ($P<0.01$ and $P<0.05$).

RESULTS AND DISCUSSION

Hormone concentrations (FSH, LH estrogen and testosterone): In heifers, the overall mean value of control group was significantly ($P<0.05$) lower (27.53, 0.96 ng/ml and 9.44 pg/ml) than treatment group (70.42, 18.27 ng/ml and 19.16 pg/ml) for FSH, LH and estrogen respectively in puberty period (Table 1). This may be due to the fact that DPP contains cholesterol, carotene and estrone, which enhanced the gonads and caused an increase in the FSH and LH concentrations (Karl *et al.* 2021). Messman *et al.* (2020) reported that presence of cytochrome enzyme and estrogen-like compounds in the blood led to increased estrogen level, these compounds were found in the DPP, for which T1 mean values of FSH, LH and estrogen were higher than T2. Melvin *et al.* (2020) reported that increasing of estrogen leads to advance puberty in heifers. Our results are in agreement with results reported by Gasser *et al.* (2006) who found that ration with high concentrate diet

Table 1. Effect of date palm pollen on hormone concentrations of heifers and bulls' serum at different ages (Means ± S.D.)

Treatments	FSH (ng/ml)			LH (ng/ml)			Estrogen (pg/ml)			Testosterone (ng/ml)		
	Prepuberty	Puberty	Maturity	Prepuberty	Puberty	Maturity	Prepuberty	Puberty	Maturity	Prepuberty	Puberty	Maturity
<i>Heifers</i>												
T1 (DPP)	16.72±1.2	70.42±3.24 ^a	70.42±3.24 ^a	0.83±0.08	18.27±2.14 ^a	0.82±0.12	6.18±0.6	19.16±3.09 ^a	27.03±3.14 ^a	0.12±0.06	0.13±0.04	0.12±0.08
T2 (Control)	17.40±1.3	27.53±1.58 ^b	27.53±1.58 ^b	0.81±0.09	0.96±0.08 ^b	0.77±0.09	7.21±0.54	9.44±1.52 ^b	12.55±2.21 ^b	0.14±0.01	0.11±0.01	0.11±0.05
Significance	N.S.	**	**	N.S.	**	N.S.	N.S.	**	*	N.S.	N.S.	N.S.
<i>Bulls</i>												
T1 (DPP)	0.44±0.09	5.75±0.99 ^a	11.93±2.14 ^a	8.92±1.4	15.97±1.66 ^a	29.70±2.11 ^a	3.65±0.38	8.39±1.10 ^a	11.52±1.64 ^a	0.32±0.05	1.67±0.08 ^a	3.19±0.91 ^a
T2 (Control)	0.4±0.04	1.19±0.18 ^b	2.76±0.33 ^b	8.27±1.1	8.81±0.95 ^b	20.76±2.01 ^b	3.76±0.41	4.26±0.62 ^b	5.44±0.82 ^b	0.29±0.04	0.42±0.03 ^b	0.78±0.04 ^b
Significance	N.S.	**	**	N.S.	*	*	N.S.	*	*	N.S.	*	*

NS: not significant, **: ($P<0.01$), *: ($P<0.05$); FSH, Follicle-stimulating hormone; LH, Luteinizing hormone. T1, date palm pollen (DPP) treatment; T2, control treatment.

Table 2. Effect of date palm pollen on body weight and body condition scoring of heifers and bulls at different ages (Means±S.D.)

Treatment	Body weight (kg)			Body condition score (BCS)		
	Prepuberty	Puberty	Maturity	Prepuberty	Puberty	Maturity
<i>Heifers</i>						
T1 (DPP)	98.66±4.21	150.33±3.19 ^a	206.66±1.64 ^a	1.00±0.0	3.40±0.19	4.60± 0.64 ^a
T2 (Control)	101.33±4.17	142.66±2.12 ^b	198.66±1.19 ^b	1.00 ±0.0	3.16±0.12	3.96± 0.29 ^b
Significance	N.S.	*	*	N.S.	N.S.	*
<i>Bulls</i>						
T1 (DPP)	105.5 ±6.86	155±3.30 ^a	212.5±2.11 ^a	1.00±0.0	3.25±1.30	4.70± 0.50 ^a
T2 (Control)	99.5 ±5.22	139.5±3.17 ^b	196±2.42 ^b	1.00 ±0.0	3.00±0.17	3.65± 0.22 ^b
Significance	N.S.	*	*	N.S.	N.S.	*

NS: Not significant; **: ($P<0.01$), *: ($P<0.05$); T1, date palm pollen (DPP) treatment; T2, control treatment.

accelerates puberty and sexual maturity in heifers.

In bulls, the overall mean value of treatment group was significantly ($P<0.05$) higher (5.75, 15.97 ng/ml and 8.39 pg/ml) than control group (1.19, 8.81 ng/ml and 4.26 pg/ml) for FSH, LH and estrogen respectively in puberty period (Table 1). Amirah *et al.* (2020) reported DPP supplementation increased testosterone level in the blood of bulls which leads to puberty and sexual maturity advancement. Thus, significant differences in hormone concentrations of bulls at puberty and maturity age may be due to presence of DPP which improved the growth of pituitary gland and contains gonadotropin-like compounds which increase the concentration of FSH, LH and testosterone consequently leading to advance male puberty and sexual maturity, as these hormones play a major role in growing of testes, controlling sperm formation and improving male fertility (Bahmanpour *et al.* 2006, Abedi *et al.* 2013). The results showed that there were no significant differences ($P>0.05$) between T1 and T2 on hormone concentrations at prepubertal age in heifers and in bulls (Table 1).

Body weight and body condition score (BCS): In bulls, the overall mean value of treatment group was significantly ($P<0.05$) higher (155 and 212.5 kg) than control group (139.5 and 196 kg) for body weight in puberty and maturity age respectively, while in heifers, the overall mean value of treatment group was significantly ($P<0.05$) higher (150.33 and 206.66 kg) than control group (142 and 198.66 kg) for body weight in puberty and maturity age respectively (Table 2). This increase could be due to the DPP which contains chlorine and sodium, which increases appetite of

animal and body weight. Pei *et al.* (2016) and Hassan (2011) noted that the presence of mineral elements in diet leads to increase in body weight. The DPP also contains zinc, essential element for hormone production. Underwood and Suttle (1999) reported that zinc was involved in a wide range of metabolic activities, reproductive performances and humoral immune system. Satapathy *et al.* (2018) reported that cross-bred cattle raised in smallholder dairy production in the Kakatpur (India) exhibited delayed maturation and reproductive difficulties associated to mineral deficits.

In heifers, the overall mean value of treatment group was significantly ($P<0.05$) higher (4.6) than control group (3.9), while in bulls, the overall mean value of treatment group was significantly ($P<0.05$) higher (4.7) than control group (3.65) for BCS in maturity age (Table 2). The significant improvement of BCS of heifers and bulls in the current study may be due to DPP containing antioxidants which improves cell growth, immunity, body weight, body condition score, stress of heat and fertility, which leads to improved health of heifers and bulls and finally enhancement of sexual maturity. Hassan (2011) reported the presence of carbohydrates, proteins, saponins, alkaloids, minerals in DPP leads to improved body weight. This improvement in BCS may be due to the androgenic effects that have a major role in the growth and differentiation of many tissues in addition to the reproductive organs. Results revealed no significant differences ($P>0.05$) between T1 and T2 on body weight and BCS of heifers and bulls at prepuberty age (Table 2).

Advanced puberty and maturity: In heifers the overall

Table 3. Effect of date palm pollen on advanced puberty and maturity of heifers and bulls at different ages (months ± S.D.)

Treatment	Puberty	Advanced Puberty	Maturity	Advanced Maturity
<i>Heifers</i>				
T1 (DPP)	7.3 ±2.21 ^a	4.66±0.44 ^a	9.33±1.18 ^a	8.66±1.20 ^a
T2 (Control)	12.33±3.17 ^b	-0.33±0.01 ^b	19.0±2.09 ^b	-1.33± 0.10 ^b
Significance	*	**	**	**
<i>Bulls</i>				
T1 (DPP)	8.5±2.80 ^b	3.5± 0.11 ^a	9.5±0.30 ^b	8.5 ±1.10 ^a
T2 (Control)	13.0±3.11 ^a	-1.0± 0.02 ^b	18.0± 2.17 ^a	0.00±0.00 ^b
Significance	*	**	**	**

NS: Not significant; **: ($P<0.01$), *: ($P<0.05$); T1, date palm pollen (DPP) treatment; T2, control treatment.

mean value of treatment group was significantly ($P < 0.05$) higher (4.66 and 8.66 month) than control group (- 0.33 and - 1.33 month), and in bulls, the overall mean value of treatment group was significantly ($P < 0.05$) higher (3.5 and 8.3 month) than control group (-1 and 0.0 month) for advance in puberty and maturity age respectively (Table 3). This may be due to the fact that DPP led to accelerated puberty and sexual maturity, as DPP contains carbohydrates, proteins, saponins, alkaloids, minerals. Saha and Bhattacharya (2017) reported DPP is a rich source of antioxidants, energy and hydrophobic minerals, so it is suitable as a regular component in the animal diets which leads to advance puberty and maturity. Al-Samarai *et al.* (2018) obtained similar results. Gasser *et al.* (2006) mentioned high-concentrate diet (60% corn) begin in puberty, precocious puberty and sexual maturity, showing that calves can be continuously stimulated by starting to feed a high-energy diet such as carbohydrates in beef veal at about 3 months of age. Bachtiar and Udin (2019) reported that iron increases immunity, improves the health of calves and reduces anemia, which leads to early puberty and sexual maturity.

Dramatic increase in the absorption of trace minerals in the liver, heart, kidneys, spleen and lungs led to advance puberty and sexual maturity. Gomez-León *et al.* (2020), Handcock *et al.* (2020) mentioned increase in the hormone concentrations in the blood plasma and a high rate of BCS and weight are all factors that advance the age of puberty and sexual maturity in cows. Li *et al.* (2019) reported the biofeedback episode causes the hypothalamus and thyroid gland to release a series of youth-promoting hormones such as growth hormone, estrogen and testosterone, as well as hormones that stimulate the adrenal gland and thyroid gland, leading lead to precocious puberty and sexual maturity.

Adding date palm pollen to ration of Iraqi Jenoubi heifers and bulls accelerates puberty and maturity in female more than in male and increases concentrations of sexual hormones, body condition score and body weight. Puberty and maturity in heifers and bulls accelerates about 3.5 months, which provide 105\$ for an animal, about 25% benefit ratio.

REFERENCES

- Abedi A, Parviz M, Karimian, S M and Rodsari H R S. 2013. Aphrodisiac activity of aqueous extract of *Phoenix dactylifera* pollen in male rats. *Advances in Sexual Medicine* 3: 28–34.
- Al-Samarai A H, Al-Salihi F G and Al-Samarai R R. 2018. Phytochemical constituents and nutrient evaluation of date palm (*Phoenix dactylifera* L.) pollen grains. *Tikrit Journal of Pure Science* 21(1): 56–62.
- Amirah A, Raja I A R, Jakkhaphan P, Nor D R, Khairiyah M and Mohammad M R. 2020. Effect of extender supplemented with date palm pollen grain on bovine semen qualities. *Journal of TRASS* 8(2): 103–07.
- Araújo de Oliveira A C, da Silva Lédo A, Polek M, Krueger R, Shepherd A and Volk G M. 2021. Optimization of *in vitro* germination and cryopreservation conditions for preserving date palm pollen in the USDA National Plant Germplasm System. *Plant Cell, Tissue and Organ Culture* 144(1): 223–32.
- Bachtiar A and Udin Z. 2019. Effects of mineral supplementation on reproductive efficiency of Simmental heifers. *Asian Journal of Agriculture and Biology* 7: 396–403.
- Bahmanpour S, Panjeh S M, Talaei T, Vojdani Z, Poust P A, Zareei S and Ghaemian M. 2006. Effect of *Phoenix dactylifera* pollen on sperm parameters and reproductive system of adult male rats. *Iranian Journal of Medical Sciences* 31(4): 208–12.
- Brickell J S, McGowan M M, Pfeiffer D U and Wathes D C. 2009. Mortality in Holstein-Friesian calves and replacement heifers, in relation to body weight and IGF-I concentration, on 19 farms in England. *Animal* 3(8): 1175–82.
- Bruinjé T C, Rosadiuk J P, Moslemipur F, Sauerwein H, Steele M A and Ambrose D J. 2021. Differing planes of pre- and postweaning phase nutrition in Holstein heifers: II. Effects on circulating leptin, luteinizing hormone, and age at puberty. *Journal of Dairy Science* 104(1): 1153–63.
- Cardoso R C, Alves B R and Williams G L. 2018. Neuroendocrine signaling pathways and the nutritional control of puberty in heifers. *Animal Reproduction* 15(Supplement 1): 868–78.
- Daoud A, Malika D, Bakari S, Hfaiedh N, Mnafigui K, Kadri A and Gharsallah N. 2019. Assessment of polyphenol composition, antioxidant and antimicrobial properties of various extracts of date palm pollen (DPP) from two Tunisian cultivars. *Arabian Journal of Chemistry* 12(8): 3075–86. <https://doi.org/10.1016/j.arabjc.2015.07.014>
- Dickinson S E, Elmore M F, Kriese-Anderson L, Elmore J B, Walker B N, Dyce P W, Rodning S P and Biase F H. 2019. Evaluation of age, weaning weight, body condition score, and reproductive tract score in pre-selected beef heifers relative to reproductive potential. *Journal of Animal Science and Biotechnology* 10(1): 1–7. <https://doi.org/10.1186/s40104-019-0329-6>
- Duncan D B. 1955. Multiple range and multiple F tests. *Biometrics* 11(1): 1–42.
- Elberry AA, Mufti S T, Al-Maghrabi J A, Abdel-Sattar E A, Ashour O M, Ghareib S A and Mosli H A. 2011. Anti-inflammatory and antiproliferative activities of date palm pollen (*Phoenix dactylifera*) on experimentally-induced atypical prostatic hyperplasia in rats. *Journal of Inflammation* 8(1): 1–13.
- Gasser C L, Behlke E J, Grum D E and Day M L. 2006. Effect of timing of feeding a high-concentrate diet on growth and attainment of puberty in early-weaned heifers. *Journal of Animal Science* 84: 3118–22.
- Gomez-León V E, Ginther OJ, Domingues R R, Guimarães J D and Wiltbank M C. 2020. Necessity for LH in selection and continued growth of the bovine dominant follicle. *Reproduction* 159(5): 559–69.
- Handcock R C, Lopez-Villalobos N, McNaughton L R, Back P J, Edwards G R and Hickson R E. 2020. Body weight of dairy heifers is positively associated with reproduction and stayability. *Journal of Dairy Science* 103(5): 4466–74.
- Hassan H M. 2011. Chemical composition and nutritional value of palm pollen grains. *Global Journal of Biotechnology and Biochemistry* 6(1): 1–7.
- Hassan M, Arfat M Y, Arshad U and Ahmad N. 2021. Ovarian dynamics, hormone profiles, and characterization of ovarian and uterine blood flow in cycling Sahiwal cows. *South African Journal of Animal Science* 51(2): 194–204.
- Karl K R, Jimenez-Krassel F, Gibbings E, Ireland J L, Clark Z L, Tempelman R J, Latham K E and Ireland J J. 2021. Negative

- impact of high doses of follicle-stimulating hormone during superovulation on the ovulatory follicle function in small ovarian reserve dairy heifers. *Biology of Reproduction* **104**(3): 695–705.
- Li R L, Russ J, Paschalides C, Ferrari G, Waisman H, Kysar J W and Kalfa D. 2019. Mechanical considerations for polymeric heart valve development: Biomechanics, materials, design and manufacturing. *Biomaterials* **225**: 119493.
- Melvin E J, B R Lindsey, J Quintal-Franco, E Zanella, K E Fike, C P Van Tassell and J E Kinder. 1999. Estradiol, luteinizing hormone, and follicle stimulating hormone during waves of ovarian follicular development in prepubertal cattle. *Biology of Reproduction* **60**: 405–12.
- Messman R D, Contreras-Correa Z E, Paz H A, Perry G and Lemley C O. 2020. Vaginal bacterial community composition and concentrations of estradiol at the time of artificial insemination in Brangus heifers. *Journal of Animal Science* **98**(6): 1–9.
- Mohamed N A, Ahmed O M, Hozayen W G and Ahmed M A. 2018. Ameliorative effects of bee pollen and date palm pollen on the glycemic state and male sexual dysfunctions in streptozotocin-Induced diabetic Wister rats. *Biomedicine and Pharmacotherapy* **97**: 9–18.
- Parkinson T J. 2019. Infertility and subfertility in the cow: structural and functional abnormalities, management deficiencies and non-specific infections, pp 372–81. *Veterinary Reproduction and Obstetrics*, 10th Edition. (Eds) Noakes D E, Parkinson T J and England G C W. Elsevier, Amsterdam, Netherlands.
- Pathak R, Prasad S, Kumaresan A, Patbandha T K, Kumari S, Boro P, Sreela L and Manimaran A. 2020. Association of peripartum progesterone, estradiol, cortisol, PGFM and relaxin concentrations with retention of fetal membranes in crossbred dairy cows. *Indian Journal of Animal Sciences* **90**(7): 993–97.
- Pei K, Ou J, Huang J and Ou S. 2016. Coumaric acid and its conjugates: Dietary sources, pharmacokinetic properties and biological activities. *Journal of the Science of Food and Agriculture* **96**(9): 2952–62.
- Saha B and Bhattacharya S G. 2017. Charting novel allergens from date palm pollen (*Phoenix sylvestris*) using homology driven proteomics. *Journal of Proteomics* **165**: 1–10.
- SAS. 2012. *Statistical Analysis System*, Users Guide. Statistical. Version 9th ed. SAS. Inst. Inc. Cary, NC, USA.
- Satapathy D, Mishra S K, Swain R K, Sethy K, Barik S, Sahoo J K and Boitai S S. 2018. Incidence of reproductive problems and blood mineral status of crossbred cattle in Kakatpur Block of Odisha, India. *International Journal of Current Microbiology and Applied Sciences* **7**: 471–48.
- Severino-Lendechey V H, Montiel-Palacios F, Ahuja-Aguirre C, Peralta-Torres, J A and Segura-Correa J C. 2020. Feed supplementation affect age and weight at puberty in Girolando (*Bos taurus* × *Bos indicus*) heifers in the tropics. *Livestock Science* **240**: 104154.
- Stockton M C, Wilson R K, Funston R, Feuz D and Stalker L A. 2012. Heifer breeding maturity and its effects on profitability: *Nebraska Sandhills Beef Cattle. Research Bulletin*, 350.
- Underwood E J and Suttle N F. 1999. *The Mineral Nutrition Of Livestock*, 3rd ed. CABI Publishing, CAB International, Wallingford, Oxon, UK. *Technology* **93**: 193–203.