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Development of pollen substitutes for dearth period management of honeybee (*Apis mellifera*) colonies in foothills of Shivalik range of Himalayas

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ABSTRACT

Experiment was conducted to develop efficient and cheap pollen substitute for dearth period management of honeybee (*Apis mellifera*) colonies. Bee colonies were provided six different pollen substitutes viz., full fat soy flour (FFSF), roasted full fat soy flour (RFFSF), germinated soybean flour (GSF), defatted soy flour (DFSF), roasted defatted soy flour (RDFSF), soy protein concentrate (SPC) and compared with the control (no feeding) to determine their impact on desirable attributes of bee colonies. Results indicated that the per cent palatability of all the diets were more than 60%. A gradual increase in brood area, honey store and pollen store were observed after feeding in all the diet combination, viz SPC, followed by GSF, DFSF, FFSF and RDFSF. All the desirable parameters were found to be least in RFFSF. However, all the diets were found significantly superior over control. Significant increase in the foraging activity of the bees was found to be in colonies fed with pollen substitute. Cost and shelf-life of these patties were also calculated, in support of the adoption of these patties. Present work revealed that SPC was the best substitute for bees during the dearth period.

Key words: *Apis mellifera*, Dearth period, Defatted soy flour, Germinated soybean, Pollen substitute, Soy protein concentrate

Beekeeping is an agro-based rural industry, attaining a gradual popularity in most of the areas. Apicultural industries are being developed in hilly areas of Uttarakhand due to its rich biodiversity, ethnicity and floral sources. The success of beekeeping depends on the adequate availability of floral source. Bee receives carbohydrates from nectar and proteins from pollen (Javaheri *et al.* 2000). But in rainy season (dearth period) because of less floral rewards, supplement feeding is necessary for maintenance of bee population. Soy flour is acceptable as a protein feed for bees. It contains 47 to 50% crude protein and the amino acid profile is acceptable for bee metabolism. The amino acid isoleucine is well represented, and gives extra supplementation to the bees feeding on it (Stace and White 1994). Soy flour has been utilized extensively for a number of decades with various degrees of success in honeybee (*Apis mellifera* L.) supplementary feeds (Pokhrel *et al.* 2006, Prakash *et al.* 2007, Dodologlu and Emsen 2007, Siede *et al.* 2003, Manning 2006).

But some soy flours seem to contain antifeedant compounds that reduce the palatability to honeybees. When soy flour is heated during the manufacturing process, proteolytic enzyme inhibitors are destroyed and leads to improper digestion of the protein. Fresh soy flour is critical, as old flour has been shown to be toxic to bees (Somerville, 2005). Heat treatment to full fat soy flour (FFSF) and germination of soybean inactivate the antinutritional factors. Roasting is more effective than autoclaving for reduction of trypsin inhibitor, chymotrypsin inhibitor, and phytic acid content in soybean (El Aleem and Mohamed 2005). Therefor, the study was undertaken to develop an efficient pollen substitute from local resources for dearth period management of honebee to reduce the cost of feeding of the bees during off season.

MATERIALS AND METHODS

The present study was conducted in rainy season, i e June to October of 2007 and 2008 at Haldwani (Distt. Nainital, Uttarakhand). Bee colonies were provided by the six different pollen substitutes, viz full fat soy flour (FFSF), roasted full

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fat soy flour (RFFSF), germinated soybean flour (GSF), defatted soy flour (DFSF), roasted defatted soy flour (RDFSF) and soy protein concentrate (SPC). In control, no artificial feeding supplement was provided and bees were allowed to grow naturally. The experiment was conducted in randomozed block design and each treatment was replicated three times. The pollen substitutes were prepared as per the ratio described by Kencharaddi *et al.* (2003) with slight modification. The diet was prepared in the ratio of 10:1:1:6 (soy flour: skimmed milk powder: Brewer's yeast: honey). Multivitamin tablets (100 mg) were added in all combinations. Different substitutes (500 g) were provided to bees twice in a month at an interval of 15 days by placing them over the top bars of the frames inside the hive.

Brood area (cm^2), honey stores (cm^2) and pollen stores (cm^2) were studied by measuring the total area covered by brood (sealed and unsealed); honey (capped) and pollen (uncapped) respectively by using wire grid device (5 cm × 5 cm). Activity of forager honeybees was estimated before the treatment and 15 days interval of experimental period by counting the number of workers going out from the entrance of the hive for one minute after every two hours of interval from 10 AM to 4 PM (Srivastava *et al.* 2004). Shelf life of pollen patties was assessed at room and refrigeration temperature. Finally, the cost of different feeding materials was calculated and compared with each other to find out the cheapest pollen substitute.

Quantity of given pollen substitute was recorded initially and after the 15 days interval of total experimental period. The weight of leftout and supplied substitutes in each colonies was worked out and per cent palatability was calculated by using the formula used by Kencharaddi *et al.* (2003).

$$\text{Per cent palatability} = \frac{\text{Initial weight of substitute} - \text{weight of leftover substitute} \times 100}{\text{Initial weight of substitute}}$$

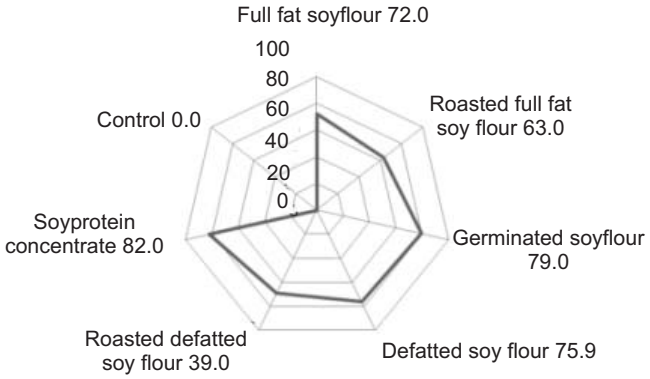
Randomized block design (RBD) was used to compute the variance. After the determination of significance of difference between the treatments means at 0.05% probability, critical difference was calculated to compare the treatment means (Snedecor and Cochran 1968).

RESULTS AND DISCUSSION

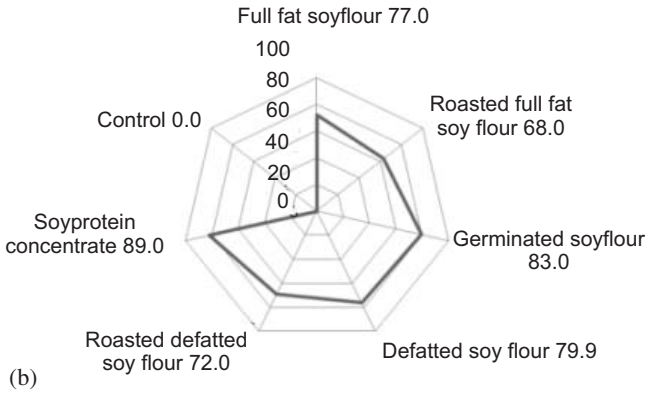
All the treatments have shown profound effect on brood development, store of honey and pollen and foraging activity of bees than control (no feeding) during both 2007 and 2008 hold the opinion that pollen is an indispensable food to be given to honeybee colonies in the spring. Honeybee colonies fed with substitutes in absence of pollens stimulated egg-laying and maintain brood rearing under less-than-optimum conditions. It was also reported that beekeepers feed artificial sources of protein such as soybean flour to enable the queen bee to keep laying eggs which when hatched and fully develop into bees, increasing colony population (Manning 2007).

Per cent palatability

Consumption of patties was significantly different from each other (Fig 1a). Honeybees preferentially consumed SPC (82%), followed by GSF (79%), DFSF (75.9%), FFSF (72%), RDFSF (69%) and RFFSF (63%) as indicated by left over patties, viz 2160 g, 2520 g, 2892 g, 3360 g, 3720 g and 4440 g respectively in 2007. Similar trend was also observed in 2008 (Fig 1b). Bees did respond positively to the low fat, high protein soybean protein concentrate rather than the plain soybean flours whether used as defatted or full fat product (Manning 2007) which was similar to the present findings. Bees consumed SPC patty more, might be due to its high protein content and the softness.



(a)



(b)

Fig 1 Per cent palatability of different soy flour pollen substitute patties (dearth period, 2007 and 2008)

Effect of pollen substitute feeding treatments on brood area

In 2007, maximum brood area was observed in SPC (785.67 cm^2), followed by GSF (766.00 cm^2), DFSF (695.33 cm^2), FFSF (655.67 cm^2) and RDFSF (616.33 cm^2) while it was the lowest in control (300.00 cm^2). Similar trend was found during 2008 also.

During both the years, brood area was almost similar at

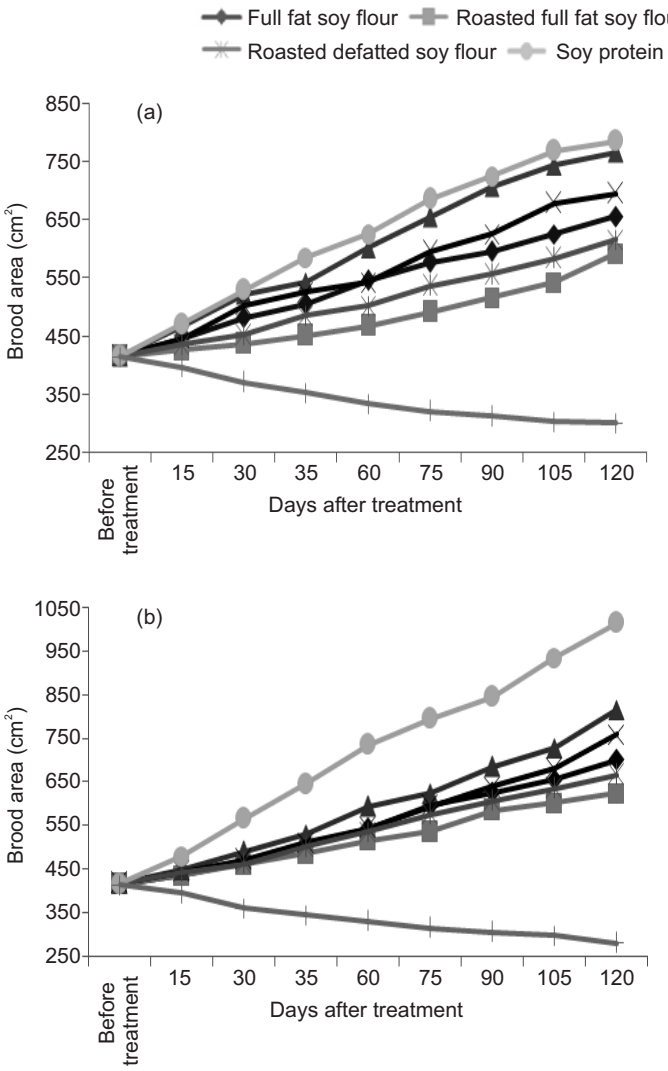


Fig 2 Brood area in different soy flour pollen substitute-fed colonies (dearth period, 2007 and 2008)

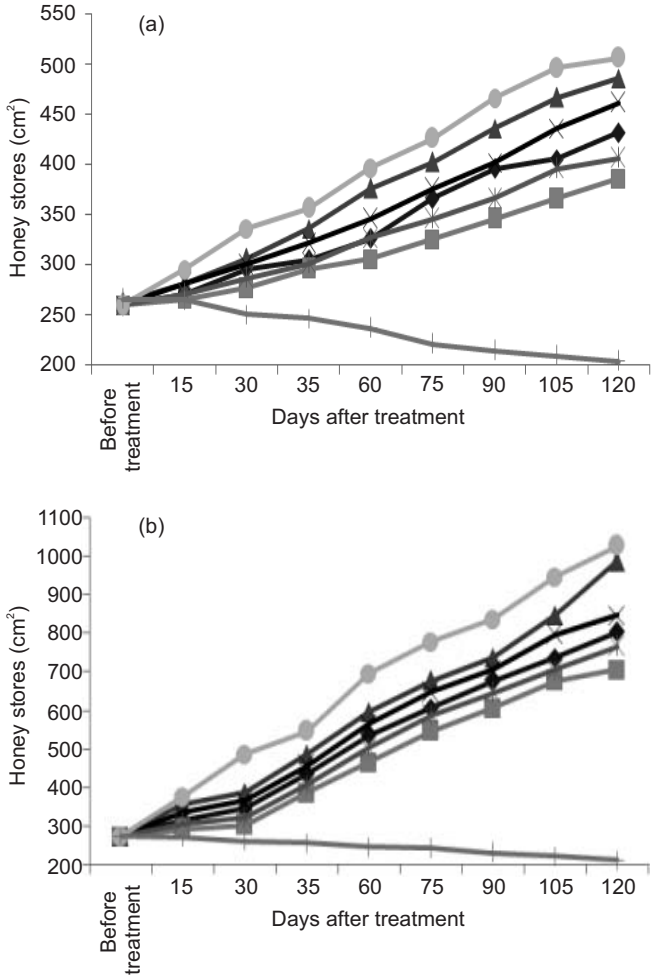


Fig 3 Honey stores in different soy flour pollen substitute-fed colonies (dearth period, 2007 and 2008)

the beginning, but a significant increase was observed after feeding on all the combinations of pollen substitutes compared to the control throughout the trial period (Fig 2a, 2b). This is supported by the findings of Kencharaddi *et al.* (2003) and Srivastava *et al.* (2004) for *A. cerana indica*. Szymaoe and Przybyl (1995) observed the suitability of soybean (90% higher than that in the non-supplemented food) with other ingredients for higher brood area. Pokhrel *et al.* (2006) also observed that three weeks feeding of soybean flour, sugar powder, skim milk powder, yeast powder, egg yolk and honey resulted higher brood rearing (158.8%) and stronger colonies (15.0%). Thapa and Pokhrel (2005) reported maximum flight activity, honey production and brood area due to soybean substitute feeding. Dastouri and Sis (2007) observed that pollen group showed positive effect on bee brood area increase.

Effect of pollen substitute feeding treatments on honey store

During 2007, significant increase in honey store was observed in SPC (506.00 cm^2), followed by GSF (485.33 cm^2), DFSF (461.33 cm^2), FFSF (431.33 cm^2), RDFSF (405.67 cm^2) and RFFSF (385.67 cm^2) while, it was less in control (203.67 cm^2) (Fig. 3a.). Similar trend was also found during 2008 (Fig 3b.)

These findings are supported by Doull (1980) who hypothesized that supplement feeding could increase the average lifespan of foragers which boosts in honey production. Utility of soy flour as a ingredient for higher honey production also reported and three weeks feeding of mixture of soybean flour, sugar powder, skim milk powder, yeast powder, egg yolk and honey has resulted in higher hive storage (Pokhrel *et al.* 2006). Similarly, Prakash *et al.* (2007) observed that a positive and significant effect of

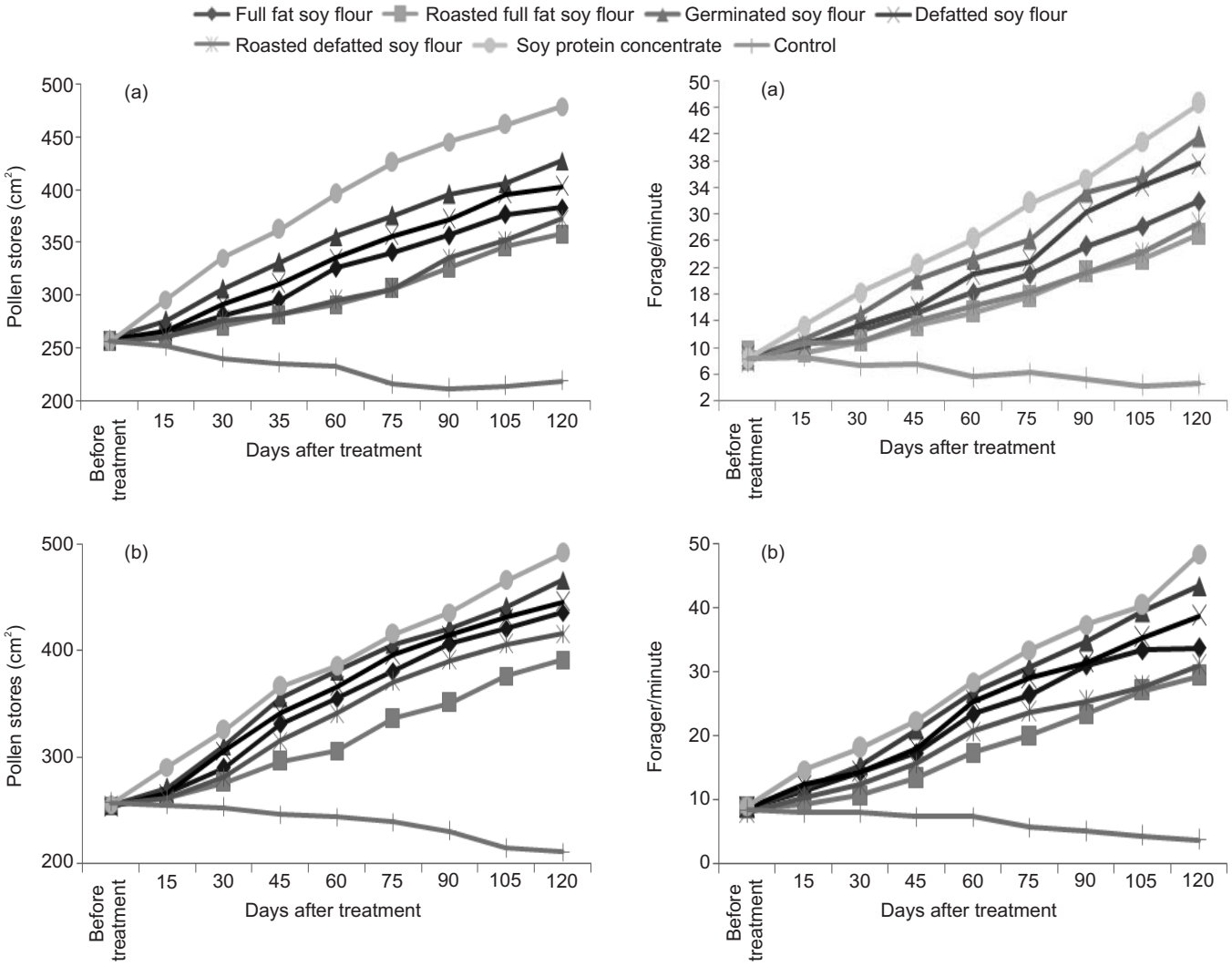


Fig 4 Pollen stores in different soy flour pollen substitute-fed colonies (dearth period, 2007 and 2008)

Fig 5 Forager/minute in different soy flour pollen substitute-fed colonies (dearth period, 2007 and 2008)

supplementary and substitution feeding of soy flour on bee colonies. However, these findings are differed from Dastouri and Sis (2007), who found that soybean meal does not have any statistically different effect in terms of increasing honey.

Effect of pollen substitute feeding treatments on pollen store

After 15 days of treatment, maximum pollen store was observed in SPC (296.00 cm^2), followed by GSF and the lowest was in RDFSF (261.00 cm^2) during 2007. Finally, pollen store in the substitute-fed colonies increased significantly and was maximum in SPC (478.67 cm^2), followed by GSF (427.33 cm^2), DFSF (403.00 cm2), FFSF (383.33 cm^2), RDFSF (372.33 cm^2) and RFFSF (358.33 cm^2) (Fig 4a). Similar trend was also found during 2008 (Fig 4b). Pollen store in all the treatments remained significantly higher throughout the experimental period than in control (218.67 cm^2).

Present results are in accordance with Prakash *et al.* (2007) who reported that pollen store increased with the feeding of soy flour steadily from the beginning to the end of the experiment and remained significantly higher than the control. Srivastava *et al.* (2004) reported that after feeding of pollen supplement, pollen hoarding capacity was better than the control. This is also supported by, Erickson and Herbert (1980) and Sabir *et al.* (2000), who reported that soybean products were good substitute as pollen supplement and had positive effect.

Effect of pollen substitute feeding treatments on foraging activity

During 2007, foraging activity was significantly higher in all the treatments than the control (4.67 forager/min.) and it was the highest in SPC (46.67 forager/min.), followed by GSF (41.67 forager/min), DFSF (37.67 forager/min), FFSF

Table 1 Cost of pollen substitute patties (made by different treated soybean flour)

Treatment	Required amount of raw material (g) for 1 kg of pollen substitute		Cost of raw material (₹/kg)	Cost of raw material used in 1 kg of pollen substitute (₹)	Total cost (₹)
	Material required	Quantity (g)			
FFSF	Flour	555.56	19.75	10.97	137.20
	SMP	55.56	152	8.45	
	Yeast	55.56	1 520	84.45	
	Honey	333.33	100	33.33	
RFFSF	Flour	555.56	19.75	10.97	137.20
	SMP	55.56	152	8.45	
	Yeast	55.56	1 520	84.45	
	Honey	333.33	100	33.33	
GSF	Flour	555.56	19.75	10.97	137.20
	SMP	55.56	152	8.45	
	Yeast	55.56	1 520	84.45	
	Honey	333.33	100	33.33	
DFSF	Flour	555.56	20	11.11	137.34
	SMP	55.56	152	8.45	
	Yeast	55.56	1 520	84.45	
	Honey	333.33	100	33.33	
RDFSF	Flour	555.56	20	11.11	137.34
	SMP	55.56	152	8.45	
	Yeast	55.56	1 520	84.45	
	Honey	333.33	100	33.33	
SPC	Flour	555.56	100	55.56	181.79
	SMP	55.56	152	8.45	
	Yeast	55.56	1 520	84.45	
	Honey	333.33	100	33.33	

(32.00 forager/min), RDFSF (28.67 forager/min) and RFFSF (27.00 forager/min) (Fig 5a). Similar pattern was also observed during 2008 (Fig 5b).

Kencharaddi *et al.* (2003) reported that supplement feeding encouraged foraging. Thapa and Pokhrel (2005) also reported high rate of flight activities due to soybean as a pollen substitute. Dreller *et al.* (1999) observed that the number of pollen foragers was significantly higher in colonies with artificially enlarged brood areas. Painkiw and Page (1999) found that the foraging activities remarkably increased with the increment of broods in the hive. Whereas, present findings were in contradiction with Schulz *et al.* (2002) who observed that starved colonies had significantly greater numbers of foragers than well-fed colonies.

Cost of pollen substitute feeding prepared from different soy flour and soy protein concentrate

Feeding cost of 1 kg pollen substitute for bee colonies (Table 1) varied greatly between FFSF, RFFSF, GSF, DFSF, RDSF and SPC. The highest cost of feeding was recorded for SPC (₹ 181.79), followed by DFSF, RDFSF, FFSF, RFFSF and GSF (₹ 137.2) pollen substitute.

Shelf-life of pollen substitute patties

In room and refrigeration temperature: During both the years, it was observed that patties kept at room temperature were not preferred by the honeybees after 15 days of preparation. It might be due to the increase in hardness of patties due to loss of moisture or grittiness or microbial growth. By putting the pollen substitute patties (formulated in trial I and trial II) in refrigerator (8ºC to 10ºC) the shelf life and bee acceptance was extend up to 25 days after preparation.

The overall study has great significance for bee keepers. The use of pollen substitute is important for growth and development of the bee colonies not only in dearth periods but also at other times (during foraging, pollination process and to overcome pesticide exposures.). From the present findings it can be concluded that although bees have accepted and grew on all the diets. However, soy protein concentrate (SPC) found to be superior for bees, followed by germinated soybean flour (GSF). Since the former contains more protein (70%) and later having less antinutritional factors. The roasted soy flour and roasted defatted soy flour showed no appreciable increase in all the parameters. All the treatment were superior to control. Soy protein concentrate showed better result than all other treatment, followed by germinated soybean flour. None of the pollen substitute was found to be toxic to bees.

REFERENCES

Dastouri M R and Sis N M. 2007. The effect of replacement feeding of some protein sources with pollen on honey bee population

and colony performance. *Journal of Animal and Veterinary Advances* **6**(11): 1258–61.

Dodologlu A and Emsen B. 2007. Effect of supplementary feeding on honey bee colony. *Journal of Applied Animal Research* **32**(2): 199–200.

Doull K M. 1980. Relationships between consumption of a pollen supplement honey production and brood rearing in colonies of honeybees *Apis mellifera. Apidologic* **11**: 361–5.

Dreller C, Page R E and Fondrk M K. 1999. Regulation of pollen foraging in honeybee colonies, effects of young brood, stored pollen and empty space. *Behavioural Ecology and Sociobiology* **45**: 227–33.

El Aleem I M A and Mohamed M H. 2005. Preparing some meat products using soybean seeds after removing the antinutritional matters. *Annals of Agricultural Science Moshtoher* **43**(2): 627–47.

Erickson E H and Herbert E W J. 1980. Soybean Products replace expeller processed soy flour for pollen supplements and substitutes. *American Bee Journal* **120**: 122–6.

Javaheri S D, Esmaili M, Nkkhaohi A, Mirhadi S A and Tahnasebi H. 2000. Honeybees with protein supplement and pollen substitute and its effects on development and resistance of honeybee's colonies and honey production. *7th IBRA Conference/5th AAA Conference*, 76 pp. Changmai, Thailand.

Kencharaddi R N, Reddy M S and Bhat N S. 2003. Evaluation of new pollen supplement for dearth period management of Indian bee *Apis Cerana* Fab. Colonies. *Indian Bee Journal* **65**(3&4): 128–30.

Manning R. 2006. Fatty acid composition of pollen and the effect of two dominant fatty acids linoleic and oleic in pollen and flour diets on longevity and nutritional composition of honeybees (*Apis mellifera* L.) Chapter **7:** Longevity of honeybees fed linoleic and oleic acid enhanced redgum (*Corymbia calophylla*) pollen diets; high and low-fat soya bean flour diets and lupin and flour diets mixed with redgum pollen. Ph D thesis, Mudorch University.

Manning R. 2007. The effect of high and low fat pollens on honeybee longevity. (*in*) *Rural Industries Research and Development Corporation Report*. Publication No. 08/031 and Project No. DAW-105A, Canberra, Australia.

Painkiw T and Page R E J. 1999. Brood pheromone and pollen forages. *Abstracts of the Proceeding of 23rd International Beekeeping Congress*. 12–17 September 1999. Vancovar, Canada.

Pokhrel S, Thapa R B, Neupane F P and Shrestha S M. 2006. Absconding behaviour and management of *Apis cerana* f. honeybee in chitwan, Nepal. *Journal of the Institute of Agricultural and Animal Science* **27**: 77–86.

Prakash S, Bhat N S, Naik M I and Hanumanthaswamy B C. 2007. Evaluation of pollen supplement and substitute on honey and pollen stores of honeybee, *Apis cerana* Fabricius. *Karnatak Journal of Agricultural Sciences* **20**(1): 155–6.

Sabir A M, Suhail A, Akram W, Sarwar G and Saleem M. 2000. Effect of some pollen substitute diets on the development of *Apis mellifera* L. colonies. *Pakistan Journal of Biological Sciences* **3**(5): 890–1.

Schulz D J, Vermiglio M J, Hvang Z Y and Robinson G E 2002. Effects of honey food shortage on social interaction in honey bee colonies. *Insectes Sociaux* **49**: 50–55.

Siede R, Buchler R and Schulz A. 2003. Detection of transgenic soyabean material in pollen substitute and honey samples. *Bee World* **84**(3): 107–11.

Snedecor G N and Cochran W G. 1968. *Statistical Methods*, 6th edn, Oxford & IBH Publishing Co. New Delhi.

Somerville D. 2005. Fat bees skinny bees- a manual on honeybee nutrition for bee-keeper. Chapter **4**: Pollen Supplements / Substitutes. *Rural Industries Research and Development Corporation Publication*.

Srivastava B G, Tiwari A and Meenakshi. 2004. Development of pollen supplements for *Apis cerana indica* Fabricius. *Indian Journal of Entomology* **66**(2): 121–3.

Stace P and White E. 1994. The use of isoleucine as a supplement feed for honeybees (*Apis mellifera*) in Australia. *The Australian Beekeeper* **96**: 159–61.

Szymaoe B and Przbyl A. 1995. Zastosowanie bialka ziemniaka wzywieniu Pszczoly miodnej (*Apis mellifera* L.). *Pszczelnicze Zeszyty Naukowe* **39**: 49–53.

Thapa R B and Pokhrel S. 2005. Impact of supplement diets on flights of crossbreed honeybee (*Apis mellifera* L.) *Journal of the Institute of Agricultural and Animal Science* **26**: 71–6.