MOVEMENT OF *CRYPTOLESTES FERRUGINEUS* (COLEOPTERA: CUCUJIDAE) IN GRAIN COLUMNS CONTAINING POCKETS OF HIGH MOISTURE CONTENT WHEAT AND CARBON DIOXIDE GRADIENTS

by

SHREEKANT PARDE

A THESIS
SUBMITTED TO THE FACULTY OF GRADUATE STUDIES
IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE
OF MASTER OF SCIENCE

DEPARTMENT OF BIOSYSTEMS ENGINEERING
UNIVERSITY OF MANITOBA
WINNIPEG, CANADA

0-612-62818-3

THE UNIVERSITY OF MANITOBA

FACULTY OF GRADUATE STUDIES

*****

COPYRIGHT PERMISSION

MOVEMENT OF *CRYPTOLESTES FERRUGINEUS* (COLEOPTERA: CUCUJIDAE) IN GRAIN COLUMNS CONTAINING POCKETS OF HIGH MOISTURE CONTENT WHEAT AND CARBON DIOXIDE GRADIENTS

BY

SHREEKANT PARDE

A Thesis/Practicum submitted to the Faculty of Graduate Studies of The University of Manitoba in partial fulfillment of the requirement of the degree of

MASTER OF SCIENCE

ABSTRACT

The movement of the adult rusty grain beetle, *Cyptolestes ferrugineus* (Stephens), in 100 cm long columns, filled with wheat of uniform moisture content (m.c.) of 12.1, 13.5 and 14.7%, was determined for horizontal and vertical placement of columns at 30°C. Also, the insect movement in columns, filled with high moisture content. wheat (above 16.5%) in the middle or top sections, and with uniform moisture content wheat (12 or 14.8%) in other sections, was determined. Insect movement was also determined under CO_2 gradients (with CO_2 introduced from either top or bottom) and no CO_2 gradients (control) in columns filled with either uniform moisture content wheat (12 or 14.6%) or with pockets of high moisture content wheat (15.5, 16, 16.6 or 17%) in the middle, bottom or top three sections.

Six columns made of polyvinyl chloride (PVC) were fabricated to hold wheat. Each column (10 cm diameter) was divided into 10 equal sections, each 10 cm long. One end of the column was closed and the other end remained opened to the atmosphere. Each section of the column had one port, except the two end sections, which had two ports each. The second port in the closed end section was for introducing CO_2 while the port at the open end was used for exit of gases. The first port in each section was used for introducing insects and for taking CO_2 samples. Carbon dioxide gradients were established in the columns by maintaining a flow of 6 mL/min from a compressed gas cylinder containing a mixture of 10% CO_2 and 90% air, by volume. The gas concentration in each section was measured using a gas chromatograph. *Cryptolestes ferrugineus* adults up to 2 mo old were taken from laboratory cultures and 200 adults, 100 each- marked and unmarked, were introduced, either

in the middle or at the top. The insect movement was determined in horizontal and vertical columns after 1, 3, 5, and 7 d, 1 and 5 d or 1 and 7 d.

In horizontal columns filled with wheat of 13.5% uniform m.c., 43% of adult *C. ferrugineus* remained in the adjacent sections from their point of introduction, but after 5 d, they exhibited the trend of distributing uniformly in the columns. In vertical columns filled with 12 or 14.8% uniform moisture content, 65% of the insects moved downward showing positive geotaxism. In horizontal or vertical columns containing pockets of high moisture content wheat, 55% of the beetles aggregated in the region of high moisture content, exhibiting hygrotactic response. Under CO_2 gradients in columns filled with uniform moisture content wheat, 60% of *C. ferrugineus* moved towards higher levels of CO_2. Also, higher levels of CO_2 were more influential in attracting *C. ferrugineus* (55%) in horizontal columns containing higher CO_2 levels at one end and high moisture content wheat at the other. In vertical columns filled with pockets of high moisture content wheat and subjected to CO_2 gradients, movement of *Cryptolestes ferrugineus* was more influenced by the combined effects of any two factors, out of the three, viz., gravity, higher levels of CO_2 and high moisture content.

ACKNOWLEDGMENTS

I express my deep sense of gratitude to my advisor, Dr. D. S. Jayas, for supporting and encouraging me throughout this research project. He has always been a source of inspiration for me and will remain so for ever. His command over the subject and his ability to explain things in the simplest possible way always helped me tackle any problem in this project with ease. I am particularly grateful to him for suggesting this topic to me for my research work which helped me expand my knowledge of stored grain as an insect habitat. His invaluable guidance and constructive comments helped me to complete this work.

I am indebted to Dr. N. D. G. White for his suggestions and guidance. He rendered great help to me in setting up the experimental apparatus in his laboratory and gave me useful advice during my experiments. I am thankful to Dr. W. E. Muir for his guidance and counsel on this study. I am also thankful to Dr. R. Roughley for serving on my thesis committee.

I owe a great deal to Colin Demianyk, who taught me the operation of all the equipment in Dr. White's laboratory. Without his technical expertise, this work would never have been completed.

Thanks are also due to Dale Bourne, who took special efforts to fabricate my apparatus in minimum possible time. I also thank the Natural Sciences and Engineering Research Council (NSERC) of Canada for providing financial support for this study.

Special thanks are to my wife, Alka, for her encouragement and support and also to my two daughters, Sneha and Puja, who showed immense patience and fortitude to stay in India to let me complete this work.

I thank Raj, Chitra, Fuji, and all others for their help.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF SYMBOLS

m.c.	Moisture content (% wet basis)
r.h.	Relative humidity (%)
T	Temperature (°C)
x	Distance (cm)
t	Time (d)
CO_2	Carbon dioxide
mo	Month

1. INTRODUCTION

1.1 Insect infestation in grain bulk

The total annual production of grains (cereals and oilseeds) in Canada is 64.2 Mt; of which wheat production is 22.4 Mt/yr (Canada Grains Council 1998). Canada ranks sixth in the world in wheat production. Wheat is mainly grown in the three Prairie Provinces- southern (Alberta), (southern) Saskatchewan, and (southwestern) Manitoba. Wheat harvest starts on the Canadian Prairies in late August or early September. The crop is swathed when the moisture content of grain is about 20-35% and is usually dried to below 14.5% in the field. After threshing the grain is stored in cylindrical steel bins on farms until it is moved to primary elevators, processing plants, or used by farmers for their own livestock.

Storage losses of grain are a significant factor in any country's food supply. Canada is no exception. Estimates of total losses as high as 50% have been reported for some countries (Parpia 1976). Most losses result from infestation by insects, microorganisms, rodents, and birds. A significant proportion of the total losses results from respiration and gradual deterioration of viability, nutritive quality, and end-use properties during storage. Nutrients are lost because of changes in carbohydrates, proteins, lipids, and vitamins (Pomeranz 1985). Functional properties, including germinability and flour quality, are lost, and aesthetic changes, including discoloration, caking , and abnormal odours occur. Also, mycotoxins can be produced that are toxic when the damaged grain is ingested.

Canada ranks second in the world in wheat export. It has legally set a zero tolerance limit for stored-product insects in exported grain. Elevators are not allowed to knowingly accept grain infested with one or more stored-grain insects. In the Prairie Provinces, newly harvested grain is seldom infested with stored grain insects and mites when first binned. During the storage period, infestation of grain depends on:

1. the proximity and abundance of stored-grain pests;

2. the ability of insects to gain a foot-hold and cause extensive damage; and

3. the initial storage condition of the grain.

The initial storage condition is influenced by temperature, moisture content (m.c.) and the amount of grain dust, chaff, and weed seed present (Liscombe and Watters 1962). On hot days insects can crawl or fly from one granary to another or from natural reservoirs and multiply rapidly if the temperature in the granary is between 30 to 35°C. High moisture content increases the internal breakdown of kernels due to an increase in enzymic activity and also causes microorganisms to multiply when the relative humidity exceeds 70%. Molds and fungi become visible on grain. Fungal deterioration of grain is a dynamic process that involves a succession of microorganisms, the breakdown of organic matter to yield carbon dioxide (CO_2) and water, and the generation of heat (Bothast 1978). At 14.0 and 18.0% m.c. (wet basis) in the starchy cereal seeds, a difference of less than 0.5% m.c. can make a great difference in the species of storage fungi that develop, in their rate of growth, and in the damage they do. Since fungi have lower temperature thresholds for development than insects do, they can cause grain to heat initially and then insects are attracted to this warm grain where they increase rapidly.

Grain in the Prairie Provinces is infested commonly by two beetles: the rusty grain beetle, *Cryptolestes ferrugineus* (Stephens); and red flour beetle, *Tribolium castaneum* (Herbst). Infestation by the rusty grain beetle is common in wheat. Adults and larvae feed mostly on the wheat germ and cause damage to grain due to the loss of germination and metabolic heat and moisture produced by their concentrated populations which allows molds to grow. The rate of reproduction of *C. ferrugineus* is 60 times per month at 32 to 35°C (Muir and White 1999). It aggregates in zones that become damp during storage (Surtees 1964). It also occurs in dry grain that contains a high a proportions of grain dust (Cotton 1954).

1.2 Importance of the study

Cryptolestes ferrugineus comprises the main insect threat to the safe storage of grain in the Prairie Provinces. Like any other adult insect, its activity and direction, and velocity of movement in a grain mass are influenced by:

1. physical factors, such as grain temperature and moisture content, gas concentrations, light, gravity, density of packing, and intergranular air movements, and
2. biological factors such as insect density, fungi, insect specie and time of movement.

Certain of these factors, individually or in combination, may affect its biology and behaviour in stored grain.

Stored grain is seldom homogeneous with respect to temperature, moisture content, and CO_2 that is produced during storage. Respiring insects add to the CO_2 production and may react differently to the gradients that develop in a grain mass. Natural CO_2 levels between 2 and 3% by volume in air (which is 100-fold higher than the ambient air levels) occur in infested non-airtight granaries (Sinha et al. 1986). Levels ranging from 5 to 18% occur in localized areas because of insect and microfloral respiration in wet and warm grain (White and Sinha 1980, White et al. 1982).

Infestations can be controlled by fumigating the grain mass. Physical factors like moisture content, temperature, and CO_2 gradient can be measured or predicted (Jayas 1995). To control infestations, knowledge about the locomotory behaviour and distribution of insects in a grain mass with respect to moisture content, temperature, and CO_2 gradient is necessary to identify the regions which are sensitive to infestations. Also, it would be necessary to know which factor, out of the three, is more influential in causing the movement and distribution of insects in a grain bin. A producer or elevator manager can then determine the likely sites of infestation and take suitable measures for detecting and then controlling this pest using chemical or physical methods such as controlled atmospheres. This study focused on determining the effects of moisture content, gravity, and CO_2 gradients only on insect movement because a separate study is being conducted to determine the effect of temperature gradients on adult insect movement.

1.3 Objectives

The objectives of this research were:

1. to determine the effects of grain moisture content on the movement of adult rusty grain beetles in grain columns at 30°C;
2. to determine the effects of CO_2 gradients, varying from 1 to 10% concentration, on beetle movement;

3. to determine the effects of the above two factors in combination, on the movement of beetles at 30°C; and
4. to describe the influence of these factors on the adult beetle movement.

2. REVIEW OF LITERATURE

2.1 Factors affecting distribution of insects in a grain bulk

2.1.1 Temperature and temperature gradients Spontaneous movement of insects are regulated by environmental stimuli. The primary influential factor in determining locomotion is temperature. Insects have limited ability to regulate their body temperature and temperature determines rate of movement, developmental time, fecundity, and population growth rate. Gunn and Hopf (1942) stated that temperature has a great effect on the speed of biological processes in *poikilotherms* (cold-blooded animals). They used the proportion of adults of *Ptinus tectus* (Bois.) (spider beetles) walking at specific temperatures as an indicator of activity. They found that the frequency of locomotion depended on temperature, temperature on the previous day, and the rate of temperature decrease or increase in the few minutes prior to testing.

Flinn and Hagstrum (1998) established temperature gradients in a 56 cm diameter cylindrical bin with 9 cm high sides filled with 19.9 kg of hard red winter wheat. They found that adult *C. ferrugineus* moved into and remained in warmer areas of the grain mass after 24 h. Beetles preferred to stay in the warmer area at 21-20°C, 24-20°C, and 42-20°C temperature gradients. They were able to locate the warmer area even at the 1°C temperature difference (21-20°C).

The thermal diffusivity in grain is low leading to a lag between ambient air temperature and grain temperature (Oxley 1949). The changes in the biological systems within a grain bulk are influenced by the interactions of its physical and biological components. Henson (1964) studying adult *Conophthorus coneperdus* (Schwarz). and

Perttunen and Pahoheimo (1964) studying adult *Tenebrio molitor* L. found that there was a middle temperature range within which the speed of movement did not change when the temperature was raised. For *C. coneperdus*, the middle range was between 20°C and 25°C, while for *T. molitor* it was between 20° and 30°C. They reported that as temperature increased to the lower limit of the middle temperature range locomotion increased. It also increased above the upper limit of the middle temperature range, but between these temperatures, locomotion did not increase.

Hagstrum et al. (1998) conducted experiments to test the dispersal of adult *T. castaneum* in a temperature gradient of 22-36°C in stored wheat. The dispersal was monitored for 20 h with eight microphones placed in the grain. They found that the adult male and female *T. castaneum* both preferred temperatures around 30°C with temperature preference much more evident with groups of six adults than with a single adult. A few adults were also detected near the cold end of the temperature gradient. Their tendency to spend more time at the preferred temperatures and at the locations where they were introduced may be because of the aggregation pheromone produced by both the sexes in their frass which is attractive to both sexes (Suzuki 1985). The tendency of adult *T. castaneum* to spend more time at the cold end of the temperature gradient when introduced there, was attributed to a thigmotactic response (touch stimulus) rather than a response to an aggregation pheromone (Yinon and Shulov 1970).

2.1.2 Moisture content Surtees (1964) studied the effects of pockets of damp wheat on the dispersion pattern of adults of *C. ferrugineus* using a Perspex box, with 300 mm sides, holding 25 kg wheat in four layers of 75 mm each. He used isolated pockets of non- mouldy wheat of 18% m.c. and of equally moist wheat supporting a mould flora to test the dispersion pattern after a week. He found that insects reared at 25°C and 70% r.h. accumulated in the pockets of damp wheat irrespective of whether it was mouldy or not. In another study, Watters (1969) found that more insects emigrated from wheat of 9.8% m.c. than from wheat of either 14.8% or 17.8% m.c. After 4 d at 28°C, however, emigration from wheat at 17.8% m.c. increased rapidly because of the growth of storage fungi *Aspergillus* spp. and *Penicillium* spp. which were less numerous at 22° and 15°C. Smith (1983) studied the relationship between wet grain, *C. ferrugineus*, and heating of wheat stored in granaries in the Prairie Provinces. He used two cylindrical metal granaries located at Glenlea, Manitoba for the study. After adding water to wheat in some pockets of the granaries, he recorded the temperature and also the population of adult *C. ferrugineus* at these locations for a period from April 1965 to June 1967. He found that the insect population reached maximum levels in December when outside temperature was decreasing. He, therefore, concluded that their development was accelerated by the high temperatures in wheat resulting from heating of the grain initiated by excess moisture in it. Loschiavo (1983) determined the movement of adult rusty grain beetle in columns of wheat at several moisture content with respect to time. He found that in wheat of uniform moisture content near 13%, adults of the rusty grain beetle moved down in the columns after 3 d. In columns filled with wheat of 16 and 17% m.c. at the top or in the middle zone, adults of the rusty grain beetle aggregated in these zones showing a positive hygrotactic response (moisture stimulus).

2.1.3 Fungi The occurrence of storage fungi in stored wheat influences the behavior and distribution of grain-infesting insects. Storage molds do not grow in grain at < 12% m.c.

(Agrawal et al. 1957). In his study on the association of grain storage fungi with *Sitophilus granarius*, he found that the invasion of stored wheat by this insect at 25°C and 75% r.h. invariably encouraged the growth of the *Aspergillus restrictus* species group. In bulk grain, damp pockets occur because of moisture seepage through the roof, walls, or floors, or by convection, translocation and adsorption of moisture. These damp pockets promote the growth of fungi and of stored grain insects which compound the damage. In one study, Sinha (1965) found that *C. ferrugineus* was able to sustain growth and ovipostion due to the nutrients supplied by certain fungi. He, therefore, concluded that zones that are heavily infected with fungi have sufficiently high moisture and temperature to support large insect infestations. In another study (Sinha 1966), the adults of *T. castaneum* and *T. confusum* were found feeding voraciously on fungi with *T. castaneum* laying eggs on 16 species of fungi and *T. confusum* on 10 species. Laying of eggs was maximum on fungi most suitable as adult food, while microorganisms like *Streptomyces* also resulted in mortality of larvae of *T. castaneum* and *T. confusum* (Sinha 1966). Dolinski and Loschiavo (1973) studied the effects of fungi and moisture on the locomotory behavior of *C. ferrugineus*. Adults of *C. ferrugineus* were placed on the surface of grain in plastic cylinders with perforated bottom. Insects were counted after 48 h when they had passed through the perforated brass screen at the bottom of cylinders and into the petri dishes containing wheat inoculated with different species of fungi. More adults were found in dishes of spoiled grain containing a mixture of fungi than in empty dishes or dishes of water. They concluded that response to fungi was probably induced by olfactory stimuli from volatile compounds in the fungi or by-products of fungi. With the exception of *Sitophilus granarius* (L.) and *Rhyzopertha dominica* (F.), most of the insects species can feed on fungi as an alternative food (Sinha 1971). These two species are found to be incapable of supporting populations on fungal diets. Pruthi and Singh (1945) reported that *R. dominica* does not flourish in grain infected with fungi, while *S. granarius* induces moldiness in grains harming its own population.

2.1.4 Aggregation pheromone Insect aggregation pheromones cause other members of the same species to aggregate in a particular area (Shorey 1973). Aggregating pheromones are released by either or both sexes when a suitable breeding habitat is located (Borden 1974). Borden et al. (1979) studied the effect of aggregation pheromone on *C. ferrugineus*. They developed an open arena airflow olfactometer to test the response of adult *C. ferrugineus* to various volatile stimuli. They found that beetles of mixed age and sex oriented positively upwind to the odor of beetles, frass, and pentane extracts of frass. Both sexes responded to the odor of beetle populations of mixed sex as well as to the odor of males, indicating that males produce a true population aggregation pheromone. The aggregation pheromone of the lesser grain borer, *R. dominica*, is a two component blend called Dominicalure 1 and 2 (Williams et al. 1981) that is effective in attracting adult males and females of *R. dominica* to traps placed inside and outside feed and seed warehouses (Leos-Martinez et al. 1986, 1987) and around metal farm bins containing stored rice (Cogburn et al.1984). Several stored-product insects also orient to stored grain odors (Barrer and Jay 1980, Freedman et al. 1982, Barrer 1983), and such odors may be important in helping females locate ovipositional sites (Crombie 1941).

2.1.5 Gravity *Sitophilus granarius* move upward in à heating bin of grain (Howe 1943). In another study, Howe (1951) found that *S. granarius* moved down in grain with tightness of packing being the major factor in affecting movement. He reported that tightness of packing restricted the movement but it did not prevent weevils from ovipostioning. He further found that smaller insects showed a greater tendency to move downward than the bigger ones. Adult *Tenebrio molitor* L. move downward (Cloudsley-Thompson 1953) and so do *T. castaneum* and *R. dominica* (Sharangpani and Pingale 1956). Watters (1969) studied the effect of geotaxism on the activity of adult *C. ferrugineus*. He found that more insects escaped from the bottoms of vertical columns of wheat than from the tops. Also, of those insects that remained in the grain, more were in the bottom half than in the top half. He concluded that the response to gravity was related to the quality of wheat in the columns. When the bottom half of a column contained sound wheat and the top half contained insect-conditioned wheat, more insects were present in the bottom half. Loschiavo (1974) in his study to determine the distribution of *C. ferrugineus* in wheat columns of uniform moisture content found that the highest number of beetles aggregated at the bottom after 24 h. He concluded that positive geotaxism was the predominant factor affecting the downward movement of *C. ferrugineus*.

2.1.6 Light Light does not significantly affect insect emigration during first hour of a test exposure but after 24 h, fewer insects emigrate from wheat held in darkness than from wheat illuminated for either 12 or 24 h (Watters 1969).Adult lesser grain borer, *R. dominica*, is a strong flyer (Leos-Martinez et al. 1986) with greatest flight activity occurring in late afternoon and evening, when temperature and light levels begin to decline (Sinclair and Haddrell 1985). High light intensity results in more *R. dominica* initiating flight than at medium and low intensities (Dowdy 1994). Barrer et al. (1993) reported higher flight activity of *R. dominica* from 135 to 85 min before dark than between 70 to 20 min before dark.

2.1.7 Starvation The survival of *C. ferrugineus* in dry grain depends on the availability of grain dust on which they can readily feed (Cotton 1954). Watters (1969) studied the effects of starvation on the locomotor activity of *C. ferrugineus*. He found that starvation depressed locomotor activity of insects in dry wheat. Fewer insects that had been starved for 3 d emigrated from dry wheat than those that had been kept on cracked wheat. Starved insects were less active in dry wheat than insects that had been exposed to food.

2.1.8 Density There exists a direct relationship between density of *S. granarius* and emigration; the higher their density the greater is the emigration from the grain bin (Voute 1937, cited by Watters 1969). Crombie (1944) reported similar results for *R. dominica.* The emigration of *S. oryzae* from high density pockets of grain is due to the high temperature created by the insects themselves (Birch 1946). Watters (1969) investigated the effect of density on emigration of *C. ferrugineus* in wheat. He found that emigration was low at low densities during the first 2 d. However, he did not find any significant differences in the emigration thereafter from which he concluded that high insect density (0.5 g^{-1}) neither stimulated nor depressed emigration of *C. ferrugineus*.

2.1.9 Dockage The presence of dockage in grain is quite beneficial and critical at low moisture content for some of the externally infesting species. Flour beetles reproduce in dry grain in the presence of grain dockage or dust (Cotton et al. 1960). In wheat at 8% m.c., nearly100% of confused flour beetle adults survived over a period of about 4 mo. McGregor (1964) reported that the red flour beetles showed a preference for wheat containing increasing amounts of dockage. He established a gradient of percentage dockage containing equal amounts of wheat dust, wheat chaff, and broken kernels and studied its effect on the red flour beetle. He found that after 1 wk the percentage of insects recovered in the lots of wheat with 0.0, 0.3, 4.5, 9.0, and 13.5% dockage were 1.3, 2.5, 24.4, 34.1, and 37.7%, respectively. He also found that increased dockage favored increased progeny, with the respective percentages being 0.2, 0.4, 22.6, 33.8, and 43.0%.

2.1.10 Gas concentrations Adults of *Sitophilus granarius* are able to use small quantities of O_2 present in added N_2 and move close to the point at which N_2 is being introduced into the silo (Shejbal et al. 1973). Navarro et al. (1976, cited by Navarro et al. 1981) studied the effect of purging O_2 from a silo on the insect activity in it. They found that the adult insects aggregated around leaks in a silo from which O_2 had been removed. They attributed the relatively large number of insects found around the leaks to insect aggregation at the higher O_2 tension. In another study done to investigate the effect of O_2 and CO_2 gradients on vertical dispersion of grain insects in wheat, Navarro et al. (1981) found that in the columns containing air, adults of *Oryzaephilus surinamensis* (L.) dispersed from top to bottom within 24 h, while those of *S. oryzae* and *R. dominica* penetrated to a depth of only 50 cm in 72 h. However, under O_2 and CO_2 gradients varying from 0.9 to 18.5% and 70.5 to 3.3% from bottom to top in grain columns, respectively, the downward dispersion of *O. surinamensis* was restricted. The dispersion of adult *S. oryzae* and *R. dominica* remained unaffected as they did not penetrate deep enough to encounter unfavorable concentrations.

Carbon dioxide exerts its anesthetic effect directly on the nervous system of insects via the trachea (Nicolas and Sillans 1989). Pure CO_2 has an inhibitory effect on the bioelectrical responses of the nervous system of insects, while a smaller concentration (15%)

has a stimulatory effect (Boistel 1960, cited by Niclolas and Sillans 1989). An important consequence of increased CO_2 concentration is the permanent opening of the spiracles, which induces water loss and causes mortality (Bursell 1974). Barrer and Jay (1980) in their study on the movement of larvae of *Ephestia cautella* (Walker) to locate CO_2 reported that an addition of 30% CO_2 to a grain odor stream increased the attractiveness of the odor and the oviposition responses near the source.

Adler (1992) studied the vertical dispersion of adults of *S. granarius* in a column of wheat flushed with different low O_2 modified atmospheres (MA). He found that flushing the column from above caused stronger weevil migration than purging from below. Pressurized air (flow rate:5 and 10 ml/min) and 99% N_2 atmospheres (1% O_2), had an attractive effect, causing weevils to move closer to the gas inlet point, when purged from above. Gas mixtures containing19 or 95% CO_2 (1% O_2, balance N_2) however, produced a repelling effect, when purged from above. He also found that a small fraction of the weevils was above the starting point and towards the purging point when purged with gas mixtures containing 19% CO_2 (1% O_2, balance N_2) from above. This led him to conclude that small amounts of CO_2 are attractive while higher amounts have a repelling effect. In another study, adult *C. ferrugineus* placed at the top of vertical columns moved down three times more rapidly through a CO_2 concentration difference of 1-43% CO_2 (top to bottom) than in controls in ambient air (White et al. 1993). When adult *C. ferrugineus* were added to the center of horizontal columns with CO_2 concentration varying from 3 to 37%, they were attracted to the higher levels of CO_2, while in controls of ambient air they moved equally in both directions. Work by White et al. (1995) on the effect of concentrations of CO_2 that can be produced by biological respiration (7.5-19.2%) on the activity of adult *T. castaneum*, *Cryptolestes pusillus* (Schonberr) and *C. ferrugineus* showed that the numbers of offspring in *T. castaneum*, *C. pusillus*, and *C. ferrugineus* exposed to 7.5% CO_2 were reduced by 43, 94, and 50%, respectively, and the total population at 6 wk was reduced by 53, 84, and 19%, respectively, relative to the controls.

2.2 Models for predicting insect movement

A mathematical model is an equation or a set of equations that describes observed data or a theory. Mathematical models are useful because they can be used to simulate and predict the progress of events under conditions different from those for which they are developed (Throne 1995).

Barrer et al. (1993) studied the dispersal and food-finding behavior of adult *R. dominica* considering the influences of population density, starvation, age, and time of day on initiation of flight of adult insects. They developed a predictive model which described the increase in insect progeny with respect to the density. The model was expressed as:

$$y = c - 0.236 \ln d$$

where: c = constant for the regression for an individual replicate;

y = logarithm of the total number of progeny per founding individual; and

d = density in number of individuals per 125 ml flour.

A second predictive model describing the effect of starvation, time of day, and age on flight response (p) was expressed as:

$$\log (p) = c + 1.47\, t_s - 0.283\, (t_s)^2 - 0.0919\, t_a - 0.15\, t$$

where: t_s = number of days starved;

t_a = age in days;

t =0 or 1(earlier or later time of day); and

c is the constant for the regression.

Dowdy (1994) developed a quadratic predictive model describing the relationship between temperature and the percent of adult *R. Dominica* initiating flight. He expressed it as:

$$y = -240.03 \pm 25.40 + 17.83\,(\pm 1.76)T - 0.29\,(\pm 0.03)T^2$$

where: y = percent of beetles initiating flight and T = temperature (°C).

Based on the model the minimum and maximium temperature at which adults of *R. dominica* initiated the flight were determined to be 19.9 and 41.6°C, respectively (Dowdy 1994).

3. MATERIALS AND METHODS

3.1 Experimental apparatus and set up Six columns were constructed using polyvinyl chloride, 10 cm inside diameter tube, to hold wheat (Fig.3.3). The columns were used in two sets (three columns each) for either vertical or horizontal configurations. One set of columns represented three replicates in a test. Each column (100 cm long, 10 cm diameter) was divided into 10 equal sections, each 10 cm long. A 3 mm slit was cut across one half of the column circumference between the sections. A port of 8-mm diameter was made at the center of each section of the column. This port was used for adding insects to the sections as well as for taking intergranular air samples. All ports were sealed with rubber septa. For studying insect movement under CO_2 gradients, the two end sections were provided with one additional port each. The additional port in the opened end section was covered with fine mesh sieve to let CO_2 out and air in, but prevent insects from moving out, while the port at the closed end section was used to introduce CO_2.

Each column had one end sealed and the other end fitted with a removable cover. A fine screen (30 mm diameter) was glued to the center of this cover from inside for exposing the grain in the column to ambient air conditions through a 8 mm opening. The sections of the column were designated 1 to 10, starting with #1 for the closed end section and ending with #10 for the open end section (Fig. 3.3). Before the test, the slits between the sections of the column were covered with duct tape. Each column held 6.1 kg of hard red spring wheat.

3.2 Wheat Hard red spring wheat,'AC Barrie', was used in the experiments. It was dried to the desired low moisture content by spreading it on plastic bags in thin layers on the floor at a room temperature of 22 ± 1°C and turning it frequently to achieve uniform drying. The moisture content during drying was checked at 1 h interval using a moisture meter (Model # 919, Serial # 12002, LABTRONICS, Winnipeg, Canada). When it reached the desired level, the final moisture content was reconfirmed using a standard oven-drying method (ASAE 1997a). The moisture content (% wet basis; all moisture content are on % wet basis) determined by the oven method was considered final and reported in the thesis. The dried wheat was then packed in plastic bags and kept overnight before it was placed in the grain columns.

Wheat was conditioned to the desired high moisture content by adding calculated amounts of distilled water to it in a wooden mixer, which was then rotated for 2 h with an electric motor to ensure thorough mixing. The moisturized wheat was then transferred to plastic bags and kept overnight for moisture equilibration before being used in grain columns.

3.3 Insects Adults of the rusty grain beetle, *Cryptolestes ferrugineus*, were taken from the laboratory cultures contained in glass jars. The culture medium was a mixture of 95% wheat and 5% germ, by weight. The cultures were stored in rearing cabinets maintained at 30 ± 1°C and 65 ± 5% r.h. The insects were of mixed sexes and were 1 to 2 mo old.

3.4 Insect marking A piece of 240 x 360 mm grey paper was placed in a ceramic tray of the same size. *Cryptolestes ferrugineus* adults, 150 in number, were taken from the cultures and put onto the paper. A water soluble, flourescent spray solution (saturn yellow splash colour, DAY-GLO flourescent pigments, A.R. Morteith Inc., Mississauga, ON) was gently sprayed on the insects using a Paasche air brush (Paasche Air Brush Co., Chicago, IL 60614) with a No. 5 nozzle at 69 kPa pressure. The pigment spots on the surface of the insects were allowed to dry for 10 min, and subsequently 100 insects, which had visible spots on their backs were selected using an ultraviolet lamp at 365 nm (Spectronics Corp., Westbury, NY) for introducing into the grain columns (Fig. 3.1).

3.5 Measurement of CO_2 Carbon dioxide generated in the columns during the experiments was measured by obtaining gas samples through rubber septums attached to protrusions from each section of the columns using a 10 mL gas-tight syringe with a 5 cm needle. The samples were analyzed using a Perkin-Elmer Sigma 3B gas chromatograph that had a thermal conductivity detector (Fig. 3.2). The carrier gas was helium, the oven was held at 70°C and the detector at 150°C. Carbon dioxide was separated from other gases by a 1.8-m column packed with Porapak N. Data were recorded as percentage concentration by volume with a Hewlett-Packard 3380S integrator. The gas chromatograph was regularly calibrated with fresh air samples before taking the readings.

3.6 Experimental design The experiments were carried out in three parts. In the first part, top or middle sections of the grain columns were filled with wheat of high moisture content and the insect movement was studied in the columns (Table 3.1). In the second part, CO_2 gradients were established in the columns containing wheat of uniform moisture content and insect movement was studied therein (Table 3.2). In the third part, insect movement was studied under CO_2 gradients in columns containing pockets of high moisture content wheat (Table 3.3).

3.7 Test procedure

3.7.1 Insect movement in grain columns with pockets of high moisture content wheat

Three sections of the columns, at the top or in the middle, were filled with wheat of high moisture content (16.5 to 18.2%) and the remaining sections held wheat at low moisture content (12.1 to 14.7%) (Table 3.1). The columns filled with wheat were flushed initially with air for 2 min at a flow rate of 1000 mL/min to bring wheat held inside the column to ambient air conditions. The columns were then transferred to a growth cabinet maintained at a temperature of 30 ± 1°C.

The tests were carried out in one set of columns for 1 and 3 d, and in the other set of columns for 5 and 7 d, for either horizontal or vertical configurations of columns. In each series of tests, 100 insects (unmarked) were first added to the columns at 30 ± 1°C. A further 100 marked insects were added to the columns 2 d after the introduction of the unmarked insects. The movement of the marked insects was counted for 1 d in 1 and 3 d test and for 5 d in 5 and 7 d test. The movement of unmarked insects was counted for 3 and 7 d in 1 and 3 d and 5 and 7 d test, respectively. Where only 1 and 5 d test was done, the movement of marked insects was for 1 d and that of unmarked insects was for 5 d. At the end of the test, the duct tape, covering the slits between the sections, was removed and circular galvanized steel plates were inserted into the slots to block off each section (Fig. 3.4). Grain and insects were

Fig. 3.1 Set up for marking insects.

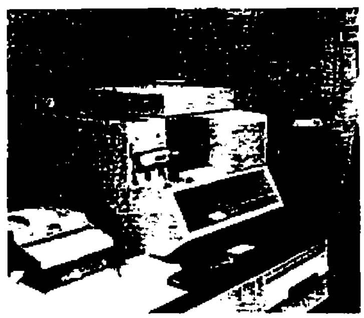


Fig. 3.2 Gas chromatograph and Integrator.

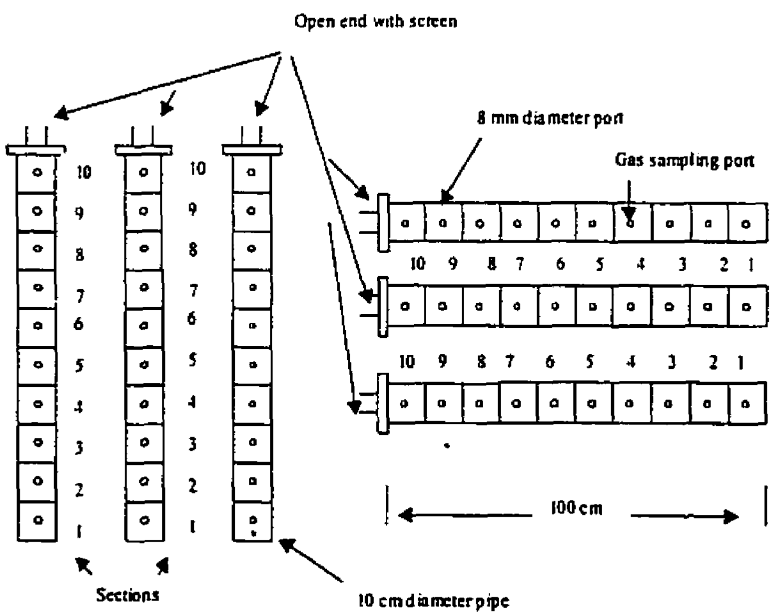


Fig.3.3. Grain columns.

Table 3.1 Experiments on insect movement in pockets of high moisture content wheat at 30°C.

Vertical columns		Horizontal columns	
Test (treatment)*	Test (control)	Test (treatment)	Test (control)
M16.5O12V15-t	U12.1V15-t	M17.5O13.6H1357-6	U13.5H1357-6
M16.9O14.8V1357-6	U14.7V1357-6		
T16.6O14.8V1357-6	"		
T18.2O14.8V15-t	U14.7V15-t		

*First character refers to: M - middle three sections, T - top three sections, U - uniform, and O - other sections. The number following M, T, U or O refers to moisture content (m.c.). Characters V and H refer to Vertical and Horizontal directions and numbers followed after V or H refer to days of sampling (1, 3, 5 and 7 days). Number or t followed after hyphen refers to insect introduction point.

For example, M16.5O12V15-t means 16.5% m.c. in the three middle sections and 12% m.c. in other sections. Insect count was taken after 1 and 5 d when columns were in vertical orientation and insects were introduced in the top section (section 10).

Table 3. 2 Experiments on insect movement under CO_2 gradients.

Vertical columns			Horizontal columns		
Introduction of CO_2	Test (CO_2 grad.)	Test (control)	Introduction of CO_2	Test (CO_2 grad.)	Test (control)
From top (Section 1)	U12InV15-6 *	U12InV15-6	In sec. 1	U12H15-6	U12H15-6
From bottom (Section 1)	U12V15-t	U12V15-t			
From bottom (Section 1)	U14.6V1735-6	U14.6V1735-6			

*'In' refers to the columns in inverted direction, i.e., closed end at the top and opened end at the bottom. The other Test codes indicate the same meaning as given under Table 3.1.

Table 3. 3 Experiments on insect movement under CO_2 gradients with pockets of high moisture content wheat in grain columns.

Position of Columns	Introduction of CO_2	Test (moisture content and CO_2 grad.)☼	Test (Control)
Vertical	From bottom (Sec. 1)	M15.5O12.1V15-t	M15.5O12.1V15-t
Vertical	From bottom (Sec. 1)	T16O14.1V17-6	T15.9O14.1V17-6
Vertical	From top (Sec. 1)	B16O13.9InV15-6	B15.9O13.9InV15-6
Vertical	From top (Sec. 1)	T17O12.8InV15-6	T17.2O12.8InV15-6
Horizontal	In sec. 1	OE16.6O14.4H15-6	OE16.7O14.3H15-6

☼'OE' refers to the three sections of columns from open end (Section 10). B refers to the bottom three sections. The other Test codes indicate the same as given under Table 3.1 and Table 3.2.

removed separately from each section and placed in vented plastic bags for 24 h. The grain was then sifted using a # 10 (2.00 mm apertures) sieve to remove all the insects, marked and unmarked, which were then counted from each section under an ultraviolet lamp at 365 nm and discarded.

The CO_2 generated in the columns due to the respiration of grain and insects was measured using the gas chromotagraph.

3.7.2 Insect movement under CO_2 gradients in grain columns The grain columns were filled with wheat of uniform moisture content and were transferred to a growth cabinet maintained at a temperature of 30 ± 1°C. They were flushed initially at 500 mL/min for 20 min using gas from a compressed gas cylinder containing a mixture of 10% CO_2 and 90% air, by volume. A stable CO_2 gradient was established, thereafter, in each of the three columns by maintaining a flow of 6 mL/ min. The flow was regulated by an autoflow controller (Model SA 202- 3(5)2, Serial # 11037, max. pressure 200 psi, VICI CONDYNE, INC., Duarte, CA) connected through a nalgene tubing (8 mm diameter) to the CO_2 cylinder (Fig.3. 5). The gas was passed through a gas washing bottle containing sulphuric acid solution to regulate relative humidity (r.h.)(Solomon 1951). The density of the solution was adjusted to give a relative humidity that was in equilibrium with the moisture content of wheat. The following equilibrium relationships between relative humidity and moisture content at 30°C were used, based on modified Chung-Pfost equation (ASAE 1997b):

wheat m.c. (%)	r.h.(%)
12.0	55.0
14.6	70.5

Fig. 3.4 Grain column with slats inserted in the slots.

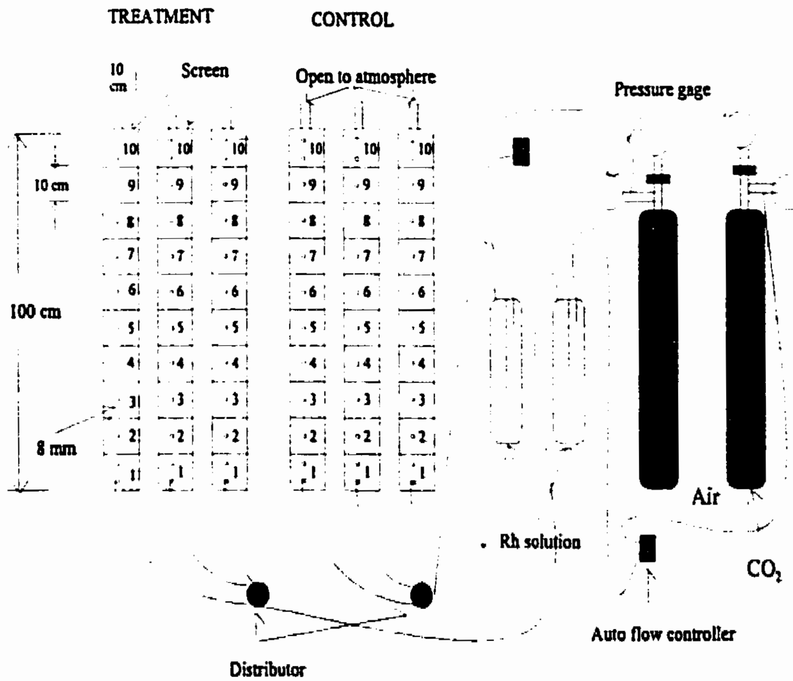


Fig. 3.5 Experimental set up for experiments in CO_2 gradients (Part II and III).

The tests were carried out for 1 and 5 d except for one test, which was carried out for 1, 3, 5 and 7 d (Table 3.2). Three columns of grain were treatments and three were controls (no CO_2) (Fig 3.5). In the tests conducted for 1 and 5 d, 100 unmarked insects were first added to the columns, followed by the addition of 100 marked insects after 4 d. In the test for 1 and 7 d, and 3 and 5 d, 100 marked insects were added after 6 and 2 d, respectively. At the end of each test, gas samples were collected and analyzed and insects were removed and counted.

3.7.3 Insect movement under CO_2 gradients with pockets of high moisture content wheat in grain columns After filling the top or middle three sections of the columns with wheat of high moisture content (15.5 to 17%) and the remaining sections with wheat of low moisture content (12.1 to 14.4%) (Table 3.3), the columns were transferred to the growth cabinet maintained at a temperature of 30 ± 1°C. The density of the sulphuric acid solution in the gas bottle, through which the gas passed, was adjusted to give an relative humidity of 70.5%. Three columns of grain were treatments and three were controls (no CO_2). The tests were carried out for 1 and 5 d, except one test, which was carried out for 1 and 7 d (Table 3.3). In all the tests, 100 unmarked insects were first added to the columns and, thereafter, 1 d before the columns were dismantled, 100 marked insects were added. At the end of each of the tests, gas samples were collected and analyzed and insects were removed and counted.

3.8 Data identification

The point where insects were introduced in the grain column was taken as the origin (0 cm) for measuring the distance moved by the insects. In vertical columns, the movement of the insects in a downward direction was considered as, "+ ", and in an upward direction as, "-

". In horizontal columns, insect movement towards the right was termed as, "- ", and towards the left as, " + ".

3.9 Data analysis

3.9.1 Analysis of insect movement in grain columns with pockets of high moisture content wheat The procedure GLM of SAS (1988) was used to perform student's *t* test on the data to determine the significant differences in mean insect counts between treatment and controls at the 5% significance level. The data for all the experiments were then modeled using linear and non-linear regressions (SigmaPlot 3.02, Jandel Scientific, San Rafel, CA). The procedure REG (regression) with selection FORWARD (for fitting the best model) and MAXR (for improvement of R^2) of SAS (1988) were used to analyze the best fit equation. The best fit equation was evaluated on the basis of F- value, coefficient of determination (R^2), standard error of estimates(SE), and randomness of residuals. The standard error of estimates (SE) was defined as:

$$SE = \sqrt{\frac{\sum (Y - Y')^2}{df}}$$

where:

Y = observed insect count (%)

Y' = predicted insect count by the model (%)

df = degree of freedom of the regression model (N minus the number of constants in the model; N = number of data points).

The effects of distance, time, and moisture content on the insects moved, were then analyzed for the best fit model.

3.9.2 Analysis of insect movement under CO_2 gradients in grain columns The same procedure was followed to analyze the data of insect movement under CO_2 gradients. Here an additional factor, mean relative percent error(e), was calculated to compare the models and select the best among them. The mean relative percent error (e) was defined as:

$$e = \frac{100}{N} \sum \frac{|Y - Y'|}{Y}$$

The dependent variable in these experiments was insect count and the independent variables were: distance, time, and CO_2 gradient.

3.9.3 Analysis of insect movement under CO_2 gradients with pockets of high moisture content wheat in grain columns The data were analyzed using the same statistical procedure as given in sec.3.9.1. The dependent variable in these experiments was insect count, while distance, time, moisture content, and CO_2 were the independent variables.

4. RESULTS

4.1 Insect movement in grain columns with pockets of high moisture content wheat

4.1.1 Insects introduced near the middle in grain columns In horizontal grain columns with high moisture pockets in the middle (test: M17.5O13.6H1357-6; Table 3.1), an average of 60% of the adult insects remained in the region of high moisture content after 1, 3, 5, and 7 d, exhibiting hygrotactic response (Fig. 4.1). In the control columns, they showed a nearly uniform distribution pattern after 5 d (Fig. 4.2). In vertical columns with high moisture pockets in the middle (test: M16.9O14.8V1357-6; Table 3.1), 55% of the insects remained in the high moisture zone after 3 d, but after 7 d, they distributed further with only 41% remaining in that region (Fig. 4.3). In the control columns, 65% of the insects moved down towards the bottom of the columns after 7 d (Fig. 4.4). There were more than double the insects in high moisture zone in the treatment columns compared with the controls in both the tests (Tables 4.1 and 4.2). With increase in time, however, more insects left the wet pockets and were distributed throughout the grain columns.

The vertical grain columns filled with high moisture content wheat at the top (test: T16.6O14.8V1357-6; Table 3.1), showed an average of 45% of the insects moving up to the high moisture zone (Fig. 4.5). The treatment columns had significantly different numbers of insects in sections 8, 9, 10, and 1 (bottom) compared with the controls (Table 4.3).

4.1.2 Insects introduced at the top In tests in vertical grain columns with insects introduced at the top (test: M16.5O12V15-t; Table 3.1), 85% of the insects moved into the pockets of high moisture content after 1 and 5 d (Fig. 4 6). In the control columns, they predominantly

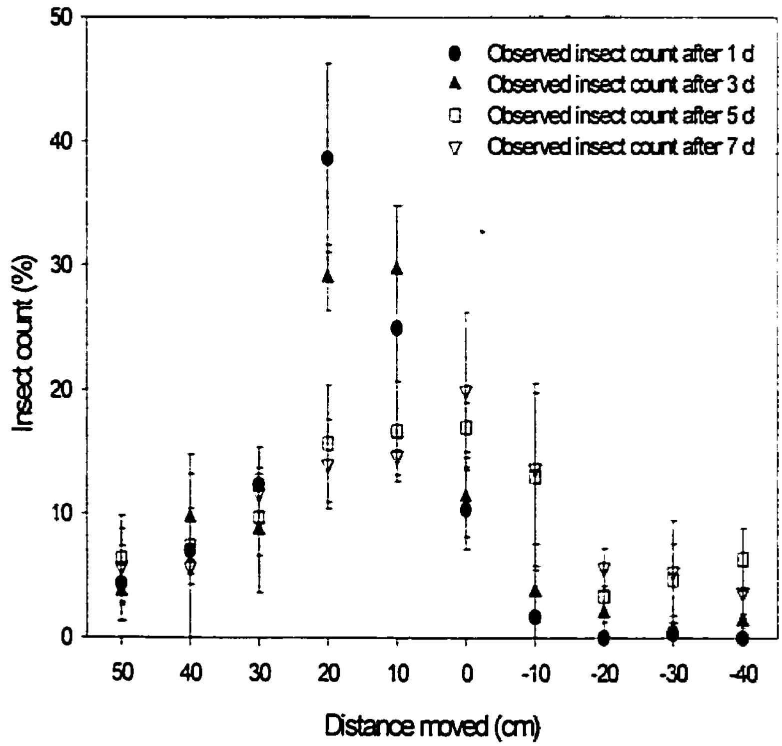


Fig. 4.1 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1, 3, 5, and 7 d in horizontal columns of wheat at 30°C. The middle sections of the columns (20 to 0 cm) had wheat at 17.5% m.c., while other sections had wheat at 13.6% m.c. Insects were introduced near the middle at 0 cm distance.

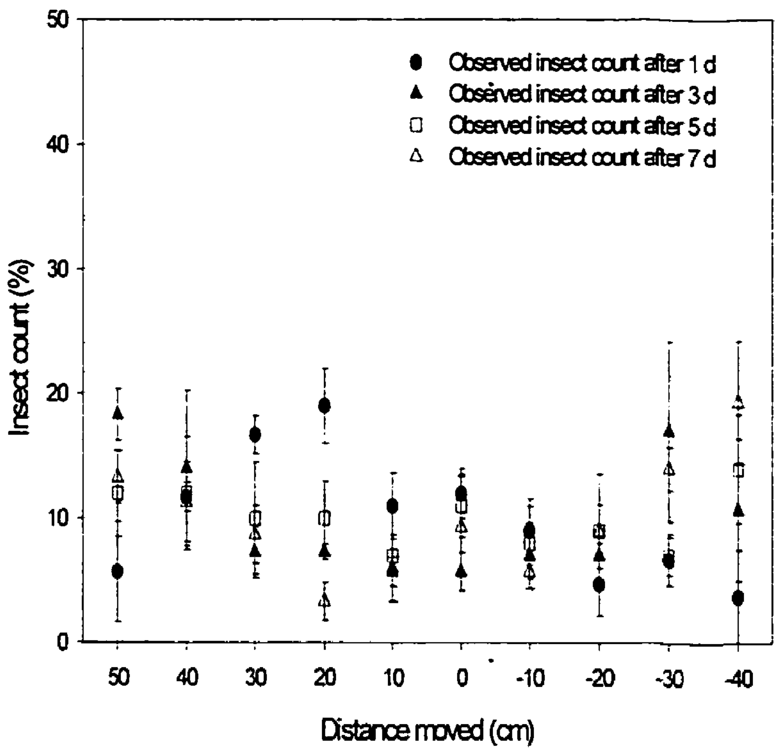


Fig. 4.2 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1, 3, 5, and 7 d in horizontal columns of wheat held at 13.5% m.c. and at 30°C. Insects were introduced near the middle at 0 cm distance.

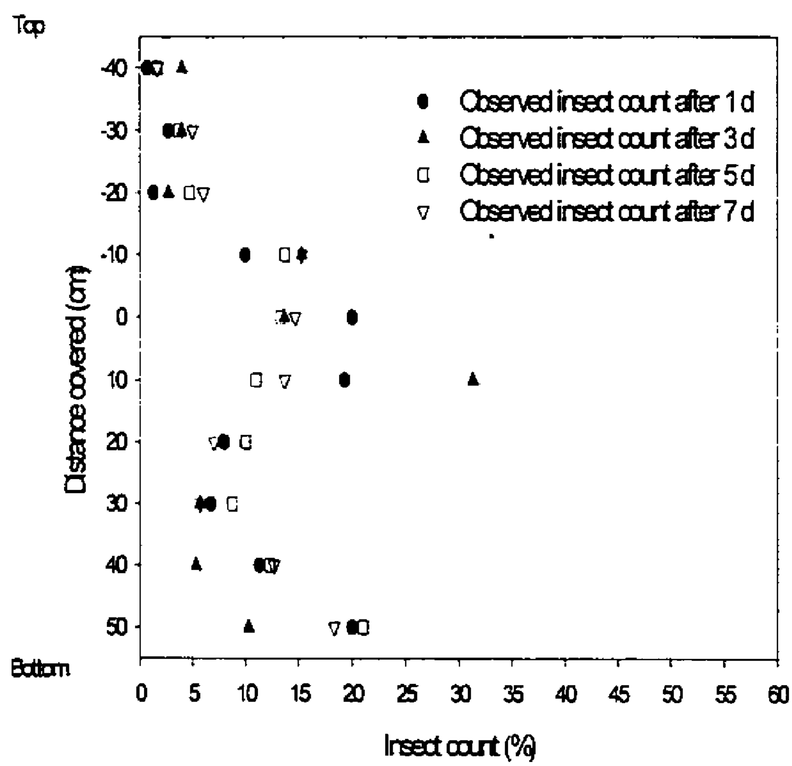


Fig. 4.3 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1, 3, 5, and 7 d in vertical columns of wheat at 30°C. The middle sections of the columns (10 to -10 cm) had wheat at 16.9% m.c., while other sections had wheat at 14.8% m.c. Insects were introduced near the middle at 0 cm distance.

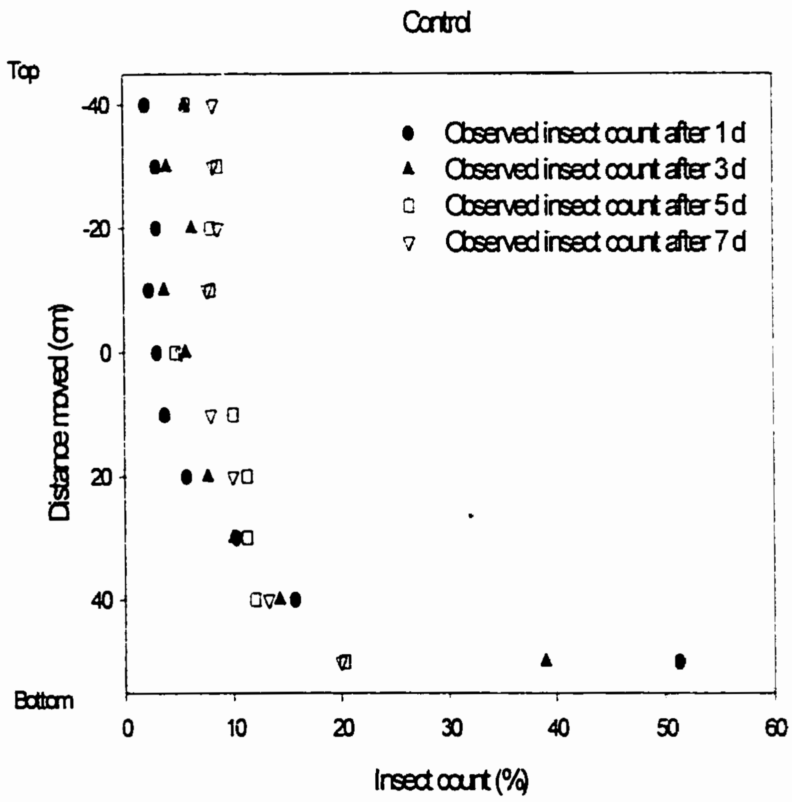


Fig. 4.4 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1, 3, 5, and 7 d in vertical columns of wheat held at 14.7% m.c. and at 30°C. Insects were introduced near the middle at 0 cm distance.

Table 4. 1 Comparison of the mean insect movement between treatment (test:M17.5O13.6H1357-6) and control (13.5 ± 0.1% m.c.) in horizontal columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control°	
		Moisture content (%)	Mean*	S.D**	Mean	S.D.
4	20	17.3 ± 0.1	24.3[a]	11.3	9.9[b]	6.3
5	10	17.8 ± 0.1	21.5[a]	8.1	7.3[b]	3.2
6	0	17.4 ± 0.1	14.6[a]	5.3	9.5[b]	3.4

S.D**:Standard deviation based on n = 12.

°The control columns contained wheat at 13.5 ● 0.1% m.c. based on n = 60

* Means with the same letter (along rows) do not differ significantly at the 5% level.

Table 4. 2 Comparison of the mean insect movement between treatment (test:M16.9O14.8V1357-6) and control (14.7 ± 0.1% m.c.). in vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control°	
		Moisture content (%)	Mean*	S.D**	Mean	S.D.
1(bottom)	50	14.8 ± 0.1	17.4^a	6.4	32.6^b	17.1
2	40	14.7 ± 0.1	10.4^a	3.9	13.8^b	3.3
3	30	14.7 ± 0.0	6.6^a	2.1	10.5^b	3.0
5	10	16.9 ± 0.1	18.8^a	8.7	6.3^b	3.3
6	0	17.1 ± 0.1	15.4^a	5.4	4.6^b	3.1
7	-10	16.7 ± 0.2	13.5^a	2.8	5.4^b	3.8

S.D**:Standard deviation based on n = 12.

°The control columns contained wheat at 14.7 ± 0.1% m.c. based on n =60

* Means with the same letter (along rows) do not differ significantly at the 5% level.

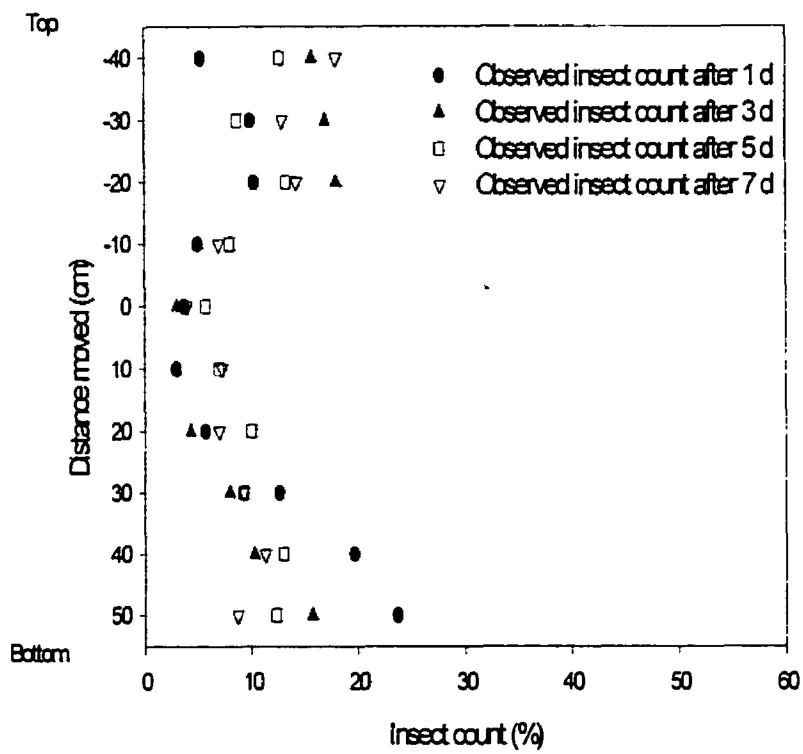


Fig. 4.5 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1, 3,5, and 7 d in vertical columns of wheat at 30°C. The top sections of the columns (-40 to -20 cm) had wheat at 16.6% m.c., while other sections had wheat at 14.8% m.c. Insects were introduced near the middle at 0 cm distance.

Table 4. 3 Comparison of the mean insect movement between treatment (test:T16.6O14.8V1357-6) and control (14.7 ± 0.1% m.c.) in vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control°	
		Moisture content (%)	Mean*	S.D**	Mean	S.D.
1 (bottom)	50	14.8 ± 0.0	15.0[a]	6.7	32.6[b]	17.1
2	40	14.8 ± 0.0	13.5[a]	5.8	13.8[a]	3.3
3	30	14.8 ± 0.1	9.0[a]	3.5	10.5[a]	3.0
8	-20	16.5 ± 0.1	14.0[a]	3.8	6.5[b]	3.5
9	-30	16.6 ± 0.0	12.1[a]	4.7	6.2[b]	3.8
10 (top)	-40	16.6 ± 0.0	12.9[a]	5.4	5.4[b]	3.4

**Standard deviation based on n = 12.

°The control columns contained wheat at 14.7 ± 0.1% m.c. based on n = 60

* Means with the same letter (along rows) do not differ significantly at the 5% level.

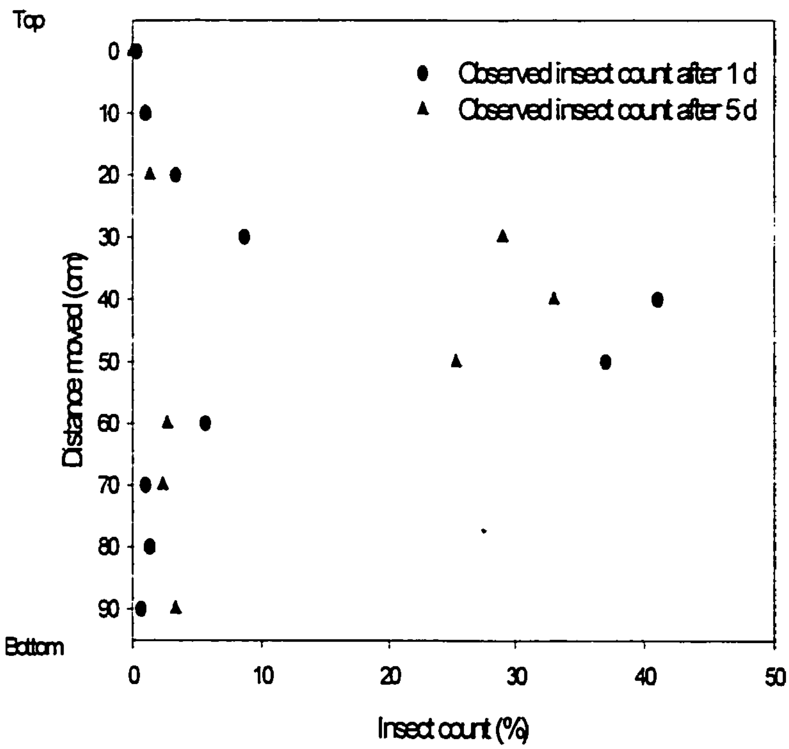


Fig. 4.6 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in vertical columns of wheat at 30°C. The middle sections of the columns (30 to 50 cm) had wheat at 16.5% m.c., while other sections had wheat at 12% m.c. Insects were introduced at the top at 0 cm distance.

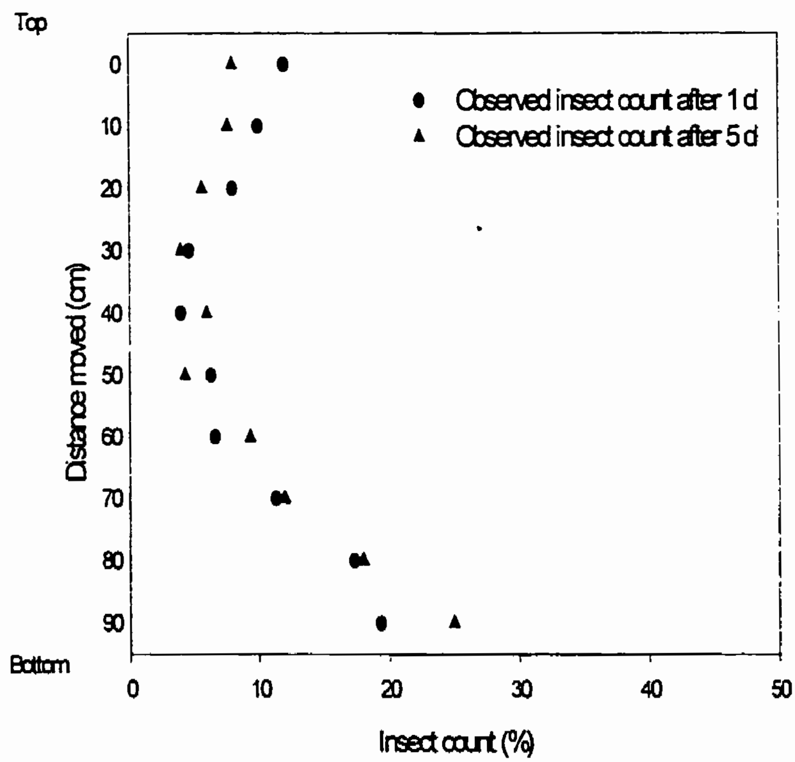


Fig. 4.7 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in vertical columns of wheat held at 12.1% m.c.and at 30°C. Insects were introduced at the top at 0 cm distance.

exhibited geotactic effect (Fig. 4.7). Because the moisture content of wheat in the other sections in the treatment columns was 12%, very few insects moved down, preferring to stay in the region of high moisture content compared with the controls (Table 4.4). In vertical columns (test: T18.2O14.8V15-t; Table 3.1), an average of 62% of the insects remained in the high moisture zone, but the rest moved down towards the bottom (Fig. 4.8). In the control columns,80% of the insects moved down in the grain columns (Fig. 4.9). There were three times more insects in high moisture zone in the treatment columns compared with controls (Table 4.5).

4.1.3 Regression model One linear and two polynomial models were examined for fitting the experimental data of insect movement in pockets of high moisture content wheat (Appendix A (Tables A10 and A11)). The dependent variable 'insect count' was transformed to 'Log (insect count + 1)' since the experimental data had '0' insect count in the data points. The independent variables were: distance, time, and moisture content. Parameters used to compare the models were: coefficient of determination (R^2), standard error of estimates (SE) and distribution of residuals. The analysis showed that the quadratic model 'RegGMCq' with the terms d^2 and mc^2 had the highest regression coefficients and better distribution of residuals than the other models (Fig. 4.10). The SE was marginally higher than the linear model, but the F statistics for these quadratic terms were significant at the 5% level. Also, the relationship between the insect count and independent variables indicated a curvilinear fit and it was, therefore, concluded that this model gave the best description of the relationship between the insect count and other variables in the grain columns. The best estimations of the insect count were obtained with this model.

Table 4.4 Comparison of the mean insect movement between treatment (test:M16.5O12V15-t) and control (12.1 ● 0.1% m.c.) in vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control°	
		Moisture content (%)	Mean*	S.D**	Mean	S.D.
1(bottom)	90	11.9 ● 0.0	2.0^{a}	1.5	22.1^{b}	3.2
2	80	12.0 ± 0.0	1.3^{a}	0.8	17.6^{b}	0.8
3	70	12.0 ± 0.1	1.6^{a}	1.6	11.6^{b}	2.5
5	50	16.3 ± 0.0	31.1^{a}	6.6	5.3^{b}	1.5
6	40	16.6 ± 0.0	37.0^{a}	4.8	5.0^{b}	2.0
7	30	16.4 ± 0.0	18.8^{a}	11.3	4.3^{b}	1.0

** Standard deviation based on n = 6.

°The control columns contained wheat at 12.1 ± 0.1% m.c. based on n =30

* Means with the same letter (along rows) do not differ significantly at the 5% level.

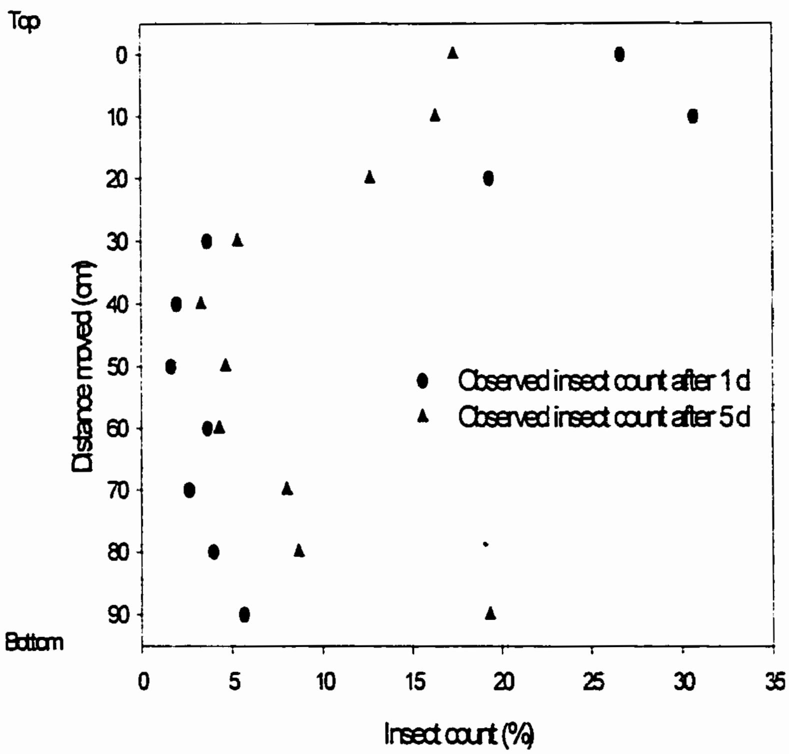


Fig. 4.8 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1and 5 d in vertical columns of wheat at 30°C. The top sections of the columns (0 to 20 cm) had wheat at 18.2% m.c., while other sections had wheat at 14.8% m.c. Insects were introduced at the top at 0 cm distance.

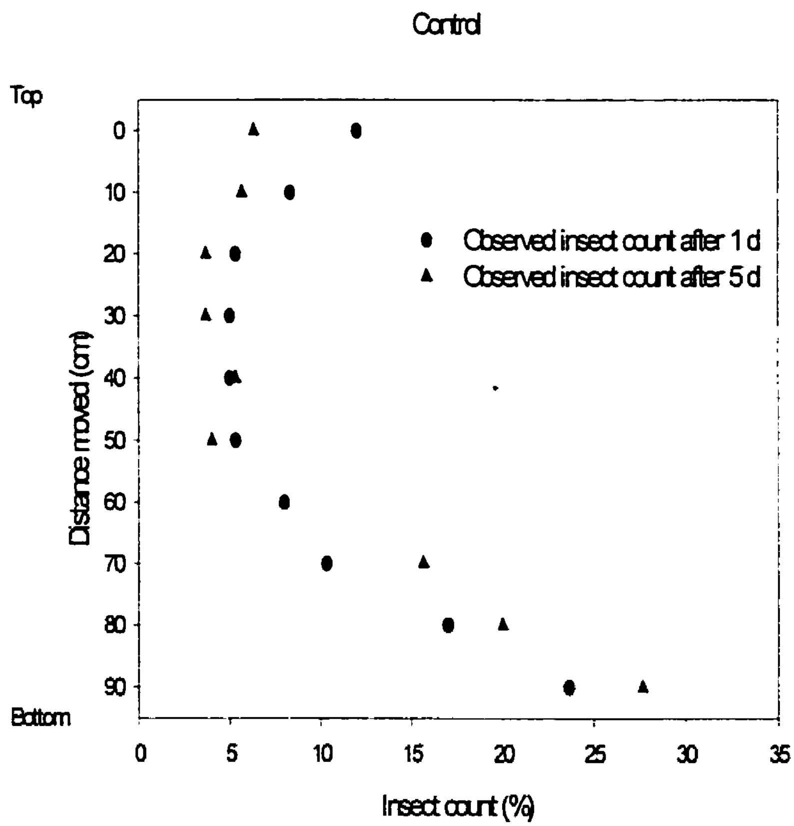


Fig. 4.9 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in vertical columns of wheat at 30°C and at 14.7% m.c. Insects were introduced at the top at 0 cm distance.

Table 4. 5 Comparison of the mean insect movement between treatment (test:T18.2O14.8V15-t) and control (14.7 ± 0.1% m.c.) in vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control°	
		Moisture content (%)	Mean*	S.D**	Mean	S.D.
1(bottom)	90	14.7 ± 0.0	12.5[a]	9.0	25.6[b]	3.5
2	80	14.8 ± 0.1	6.3[a]	2.6	18.5[b]	3.8
3	70	14.9 ± 0.2	5.3[a]	4.1	13.0[b]	3.2
8	20	18.0 ± 0.1	16.0[a]	6.1	4.5[b]	1.3
9	10	18.3 ± 0.0	23.5[a]	9.2	7.0[b]	2.3
10 (top)	0	18.2 ± 0.1	22.0[a]	5.7	9.1[b]	3.6

** Standard deviation based on n = 6.

°The control columns contained wheat at 14.7 ± 0.1% m.c. based on n =30

* Means with the same letter (along rows) do not differ significantly at the 5% level.

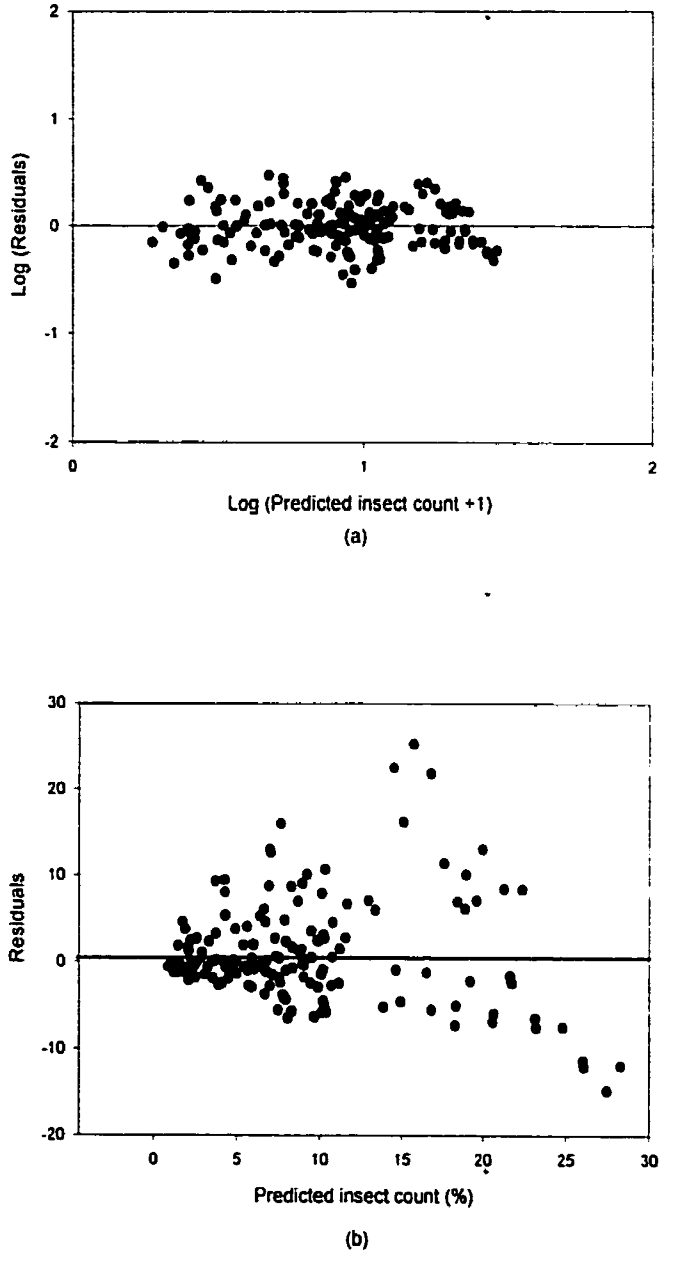


Fig. 4.10 Residual plots obtained with the best fit equation (1).

The best fit equation is:

$$\text{Log (insect count + 1)} = -3.121 + 0.005\,x + 0.025\,t + 0.354\,(\text{m.c.}) - 0.00003\,x^2 - 0.006\,(\text{m.c.})^2 \quad (1)$$

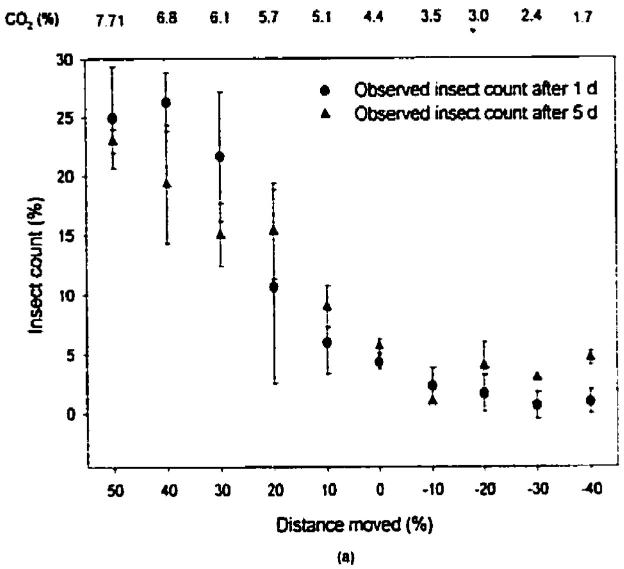
when $16.5\% < \text{m.c.} < 18.2\%$.

The R^2 value of the model is 0.64. The F-value of the overall model (55.83, $P<0.01$) indicated that it was significant. The effects of moisture content . distance, and time on the insect count were all significant ($P<0.05$). Based on equation (1), the predicted insect count was determined for all the experimental data and compared with the measured insect count (Appendix A, Figs. A1 - A5).

4.2 Insect movement under CO_2 gradients in grain columns with wheat at uniform moisture content.

4.2.1 Insects introduced near the middle Insects introduced at 0 cm distance in the horizontal treatment columns (test: U12H15-6; Table 3.2) that had CO_2 varying in concentrations from 7.7% to 1.7%, moved noticeably (≈65%) towards higher CO_2 levels after 1 and 5 d. In the control columns, in absence of CO_2 gradients, they preferably moved into the adjacent sections after 1 d and then showed a nearly uniform distribution pattern after 5 d (Fig. 4.11). Sections with higher concentrations of CO_2 in treatment columns had significantly more insects than those in controls (Table 4.6).

In vertical columns under treatment (test:U14.6V1735-6; Table 3.2), with CO_2 concentrations higher in the bottom sections, insects moved down one and half times faster than controls after 1, 3, 5, and 7 d (Figs. 4.12 and 4.13). The differences in their movement



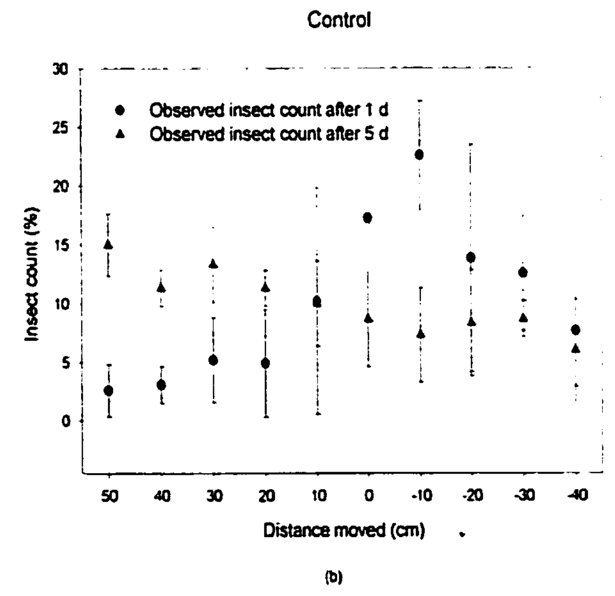
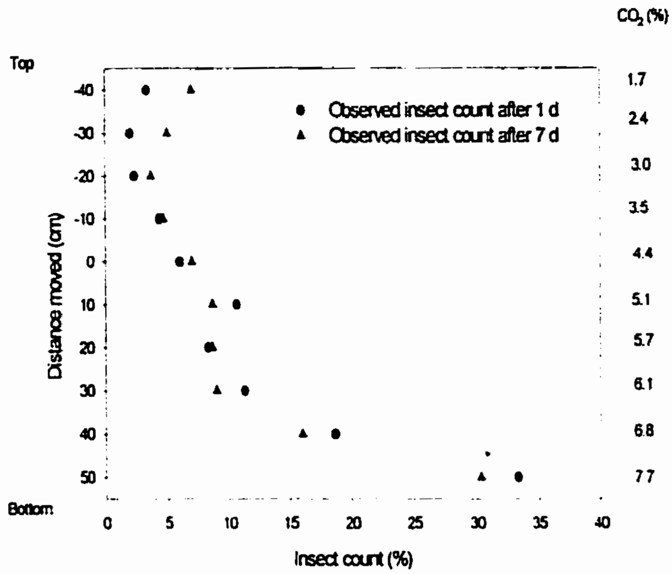
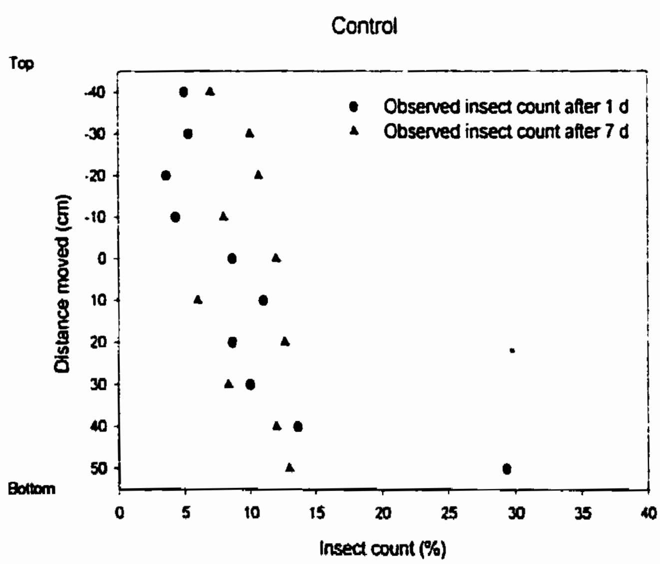


Fig. 4.11 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in horizontal columns of wheat at 30°C under CO_2 gradients and in controls. The columns held wheat at 12% m.c. Insects were introduced near the middle at 0 cm distance.

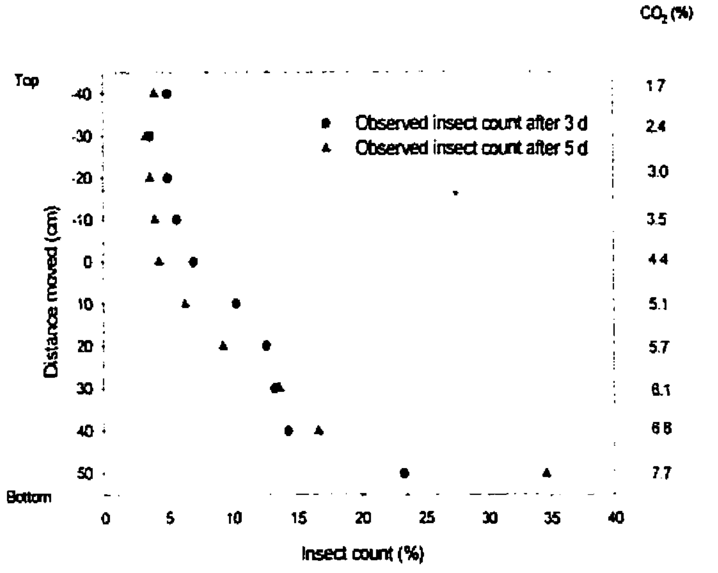


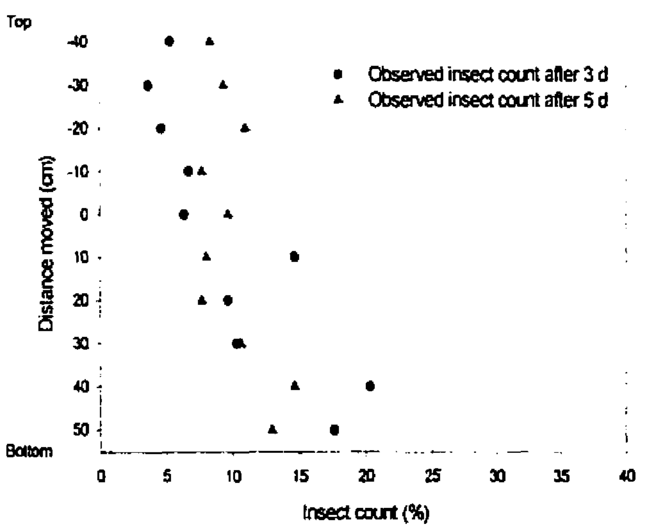
(a)



(b)

Fig. 4.12 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 7 d in vertical columns of wheat at 30°C under CO_2 gradients and in controls. The columns held wheat at 14.6% m.c. Insects were introduced near the middle at 0 cm distance.





(b)

Fig. 4.13 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 3 and 5 d in vertical columns of wheat at 30°C under CO_2 gradients and in controls. The columns held wheat at 14.6% m.c. Insects were introduced near the middle at 0 cm distance.

Table 4.6 Comparison of the mean insect movement under CO_2 gradients (test:U12H15-6) and controls in horizontal columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control	
		CO_2 (%)	Mean*	S.D*	Mean	S.D.
1(closed)	50	7.7	24.0^a	3.0	8.8^b	7.1
2	40	6.8	22.8^a	5.2	7.1^b	4.7
3	30	6.1	18.3^a	5.3	9.3^b	5.3
8	-20	3.0	2.8^a	2.0	11.1^b	7.5
9	-30	2.4	1.8^a	1.4	10.6^b	3.9
10 (open)	-40	1.7	2.8^a	2.1	6.8^a	4.1

S.D*:Standard deviation based on n=6.

* Means with the same letter (along rows) do not differ significantly at the 5% level.

Table 4.7 Comparison of the mean insect movement under CO_2 gradients (test:U14.6V1735-6) and controls in vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control	
		CO_2 (%)	Mean*	S.D*	Mean	S.D.
1 (bottom)	50	7.7	30.4[a]	6.9	18.2[b]	7.9
2	40	6.8	16.4[a]	3.9	15.1[a]	6.4
3	30	6.1	11.8[a]	3.1	9.8[a]	3.3
8	-20	3.0	3.6[a]	1.8	7.5[b]	4.0
9	-30	2.4	3.5[a]	2.1	7.0[b]	3.5
10 (top)	-40	1.7	4.8[a]	2.1	6.4[a]	3.2

S.D*:Standard deviation based on n=12.

* Means with the same letter (along rows) do not differ significantly at the 5% level.

were significant in the bottom two sections and in the top sections, 8 and 9, which showed the attractive effect of CO_2 in causing this rapid movement (Table 4.7).

In inverted vertical columns (test: U12InV15-6; Table 3.2), with CO_2 introduced from the top, ≈64% of the insects moved up towards higher CO_2 levels after 1 and 5 d, while in controls only ≈27% moved up (Fig. 4.14). There were three times more insects in the top portion of the treatment columns than the controls (Table 4.8).

4.2.2 Insects introduced at the top Insects introduced in the top section of the vertical treatment columns (test: U12V15-t; Table 3.2), purged with CO_2 from the bottom, moved downwards to the bottom after 1 and 5 d. The control columns also showed the same type of insect movement, possibly due to geotaxism (Fig. 4.15). There were no significant differences in the insect movement in any of the sections and particularly in the bottom sections between the treatment columns and the controls (Table 4.9).

4.2.3 Regression model Linear and nonlinear regressions (SigmaPlot 3.02, Jandel Scientific, San Rafael, CA) were performed on the experimental data. One linear and three polynomial models were examined for fitting the experimental data (Appendix B (Table B9 and B10)). The dependent variable 'insect count' was transformed to 'Log (insect count)' in three models, while the last model 'Reg CO_2' was fitted without transformation. The independent variables were: distance, time, and CO_2. Parameters used to compare the models were: coefficient of determination (R^2), standard error of estimates (SE), mean relative percent error (e), and distribution of residuals. The analysis showed that the cubic model '$RegCO_2$' had the highest regression coefficients and better distribution of residuals than the other models (Fig. 4.16). The SE also was comparatively lower than the other models.

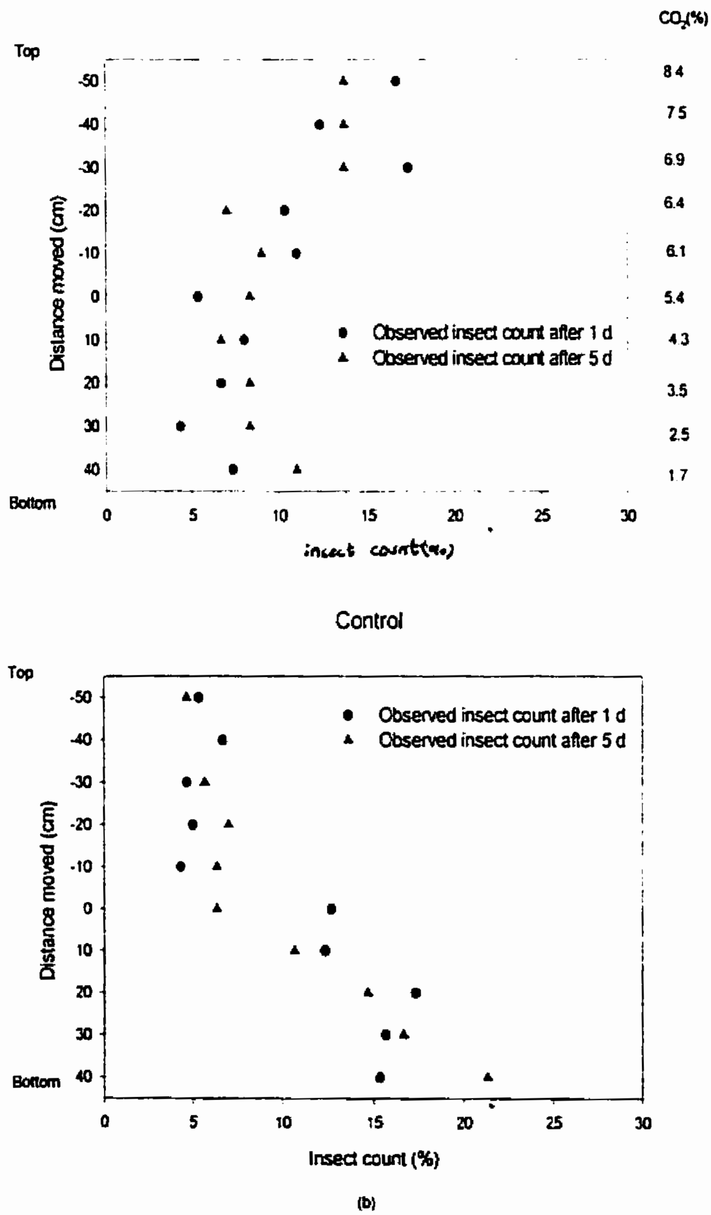
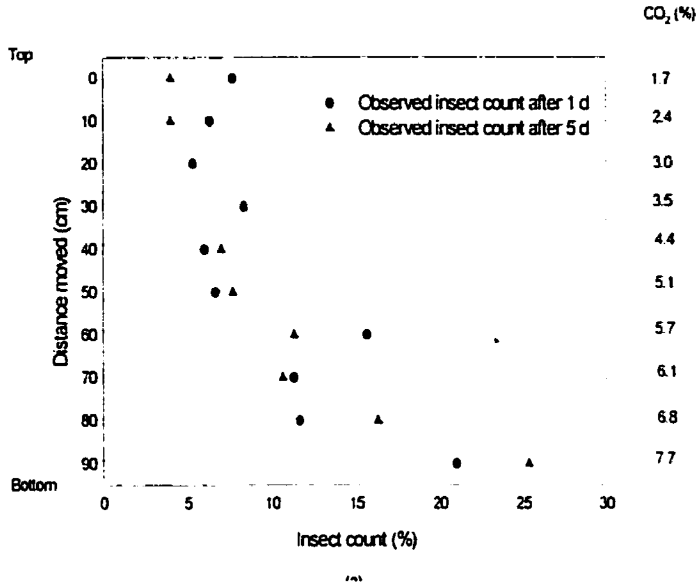


Fig. 4.14 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in inverted vertical columns of wheat at 30°C under CO_2 gradients and in controls. The columns held wheat at 12% m.c. Insects were introduced near the middle at 0 cm distance



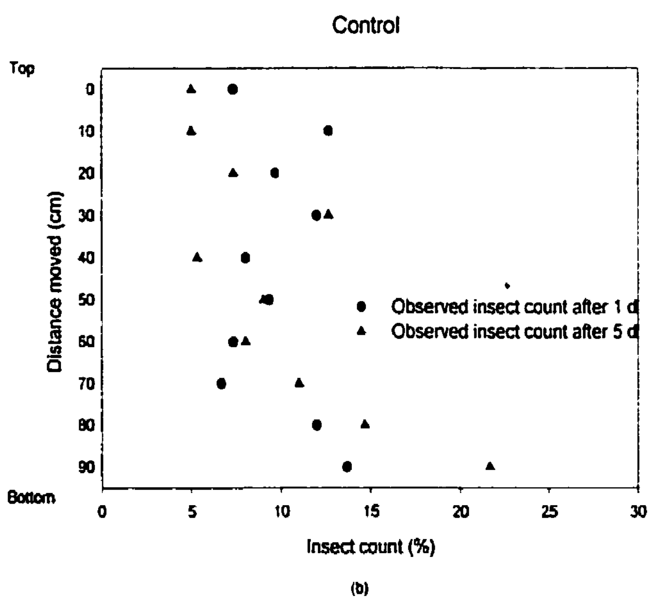


Fig. 4.15 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in vertical columns of wheat at 30°C under CO_2 gradients and in controls. The columns held wheat at 12% m.c. Insects were introduced at the top at 0 cm distance.

Table 4.8 Comparison of the mean insect movement under CO_2 gradients (test:U12InV15-6) and controls in inverted vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control	
		CO_2 (%)	Mean*	S.D*	Mean	S.D.
1(top)	-50	8.4	15.1[a]	2.7	5.0[b]	1.8
2	-40	7.5	13.0[a]	3.0	6.6[b]	2.2
3	-30	6.9	15.5[a]	3.1	5.1[b]	1.8
8	20	3.5	7.5[a]	4.5	16.0[b]	3.5
9	30	2.5	6.3[a]	4.0	16.1[b]	3.7
10 (bottom)	40	1.7	9.1[a]	4.1	18.3[b]	6.0

S.D*:Standard deviation based on n=6.

* Means with the same letter (along rows) do not differ significantly at the 5% level.

Table 4.9 Comparison of the mean insect movement under CO_2 gradients (test: U12V15-t) and controls in vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)				
		Treatment			Control	
		CO_2 (%)	Mean*	S.D*	Mean	S.D.
1(bottom)	90	7.7	23.1[a]	4.6	17.6[a]	4.8
2	80	6.8	14.0[a]	3.1	13.3[a]	2.3
3	70	6.1	11.0[a]	7.3	8.8[a]	4.2

S.D*:Standard deviation based on n=6.

* Means with the same letter (along rows) do not differ significantly at the 5% level.

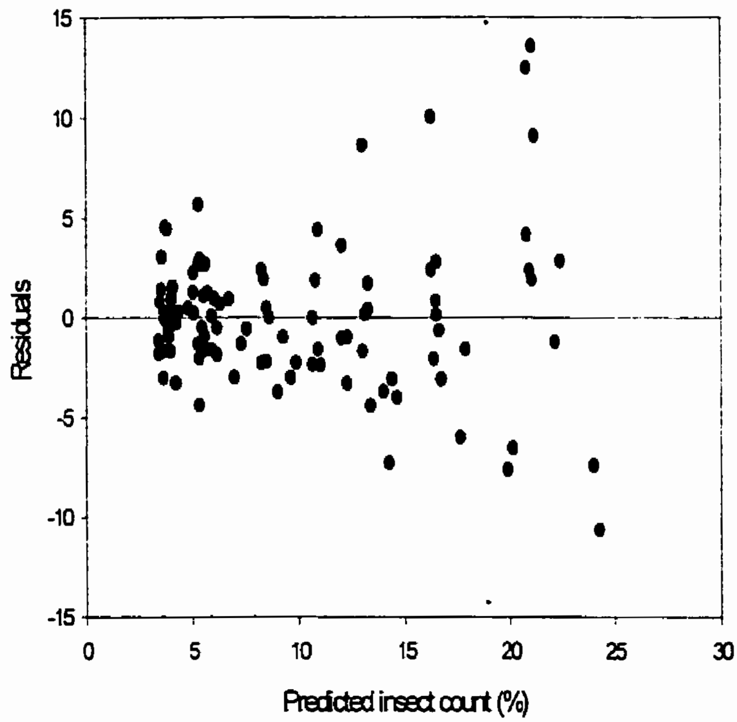


Fig. 4.16 Residual plot obtained with the best fit equation (2).

It was, therefore, concluded that this model was the best among the models tested to describe the relationship between the insect count and other variables in the grain columns. The best estimations of the insect count were obtained with this model.

The best fit equation is :

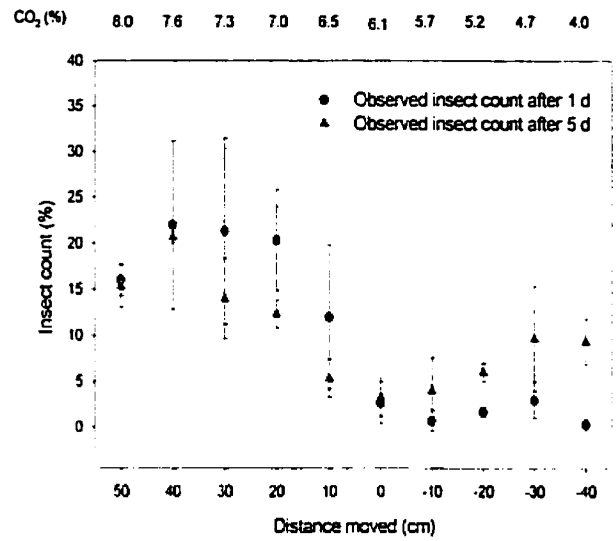
$$\text{Insect count} = 17.80 + 0.034\,x + 0.063\,t - 9.60\,CO_2 + 1.937\,CO_2^2 - 0.087\,CO_2^3 \quad (2)$$

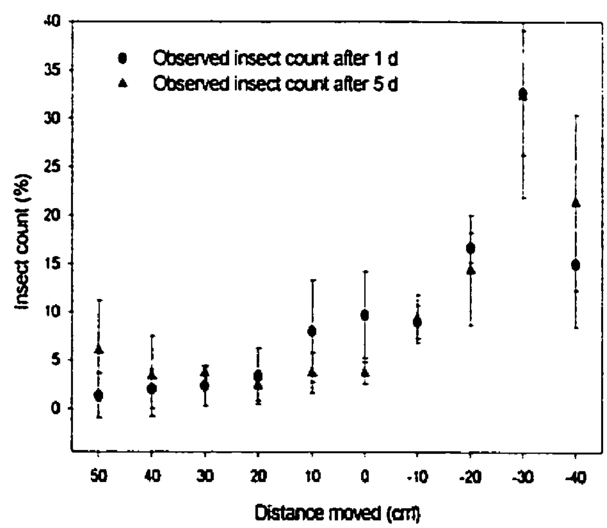
when $2\% < CO_2 < 9\%$.

The R^2 value of the model is 0.72. The effects of CO_2, distance and time on the insect count were analyzed using procedure REG of SAS (1988). The F-value of the overall model (48.03, $P<0.01$) indicated that the model was significant. The effects of distance, CO_2, and CO_2^2 were highly significant. ($P<0.05$). Time had no significant effect on the insect movement ($P>0.05$). The best fit equation (2) was used to predict the insect count for all the experimental data and these values were compared with the measured insect count (Appendix B, Figs. B1 - B4).

4.3 Insect movement under CO_2 gradients with pockets of high moisture content wheat in grain columns

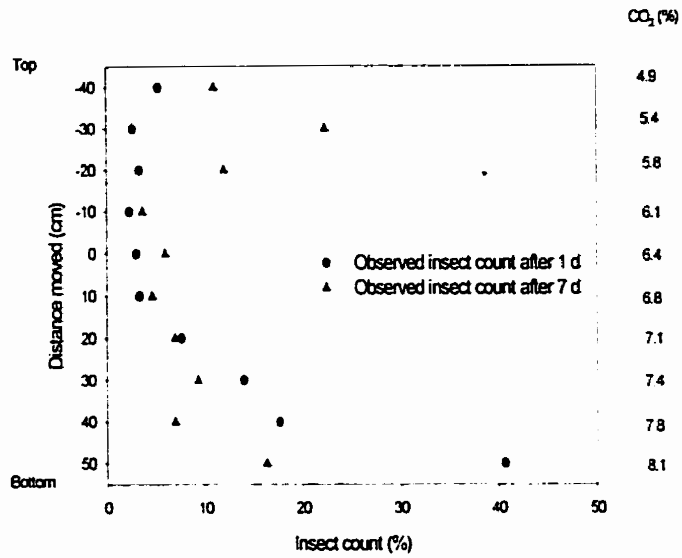
4.3.1 Insects introduced near the middle In horizontal columns with CO_2 and moisture content gradients in opposing directions (test: OE16.6O14.4H15-6; Table 3.3), 90% of the insects moved to higher CO_2 after 1 d. After 5 d, they distributed more evenly and 24% of them were found in the high moisture zone. In the control columns with no CO_2 gradient, 64% of the insects moved into the high moisture zone after 1 and 5 d (Fig. 4.17). The



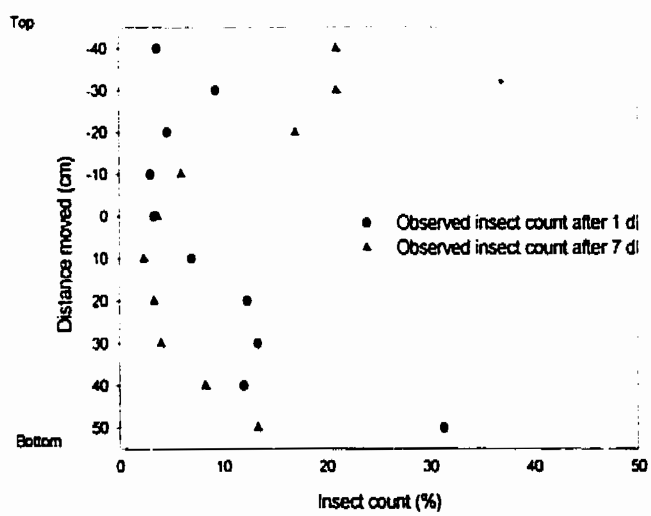


(b)

Fig. 4.17 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in treatment (CO_2 gradients and high moisture content) and in controls (no CO_2 gradients but high moisture content present) in horizontal columns of wheat at 30°C. The side sections of the columns (-20 to -40 cm) had wheat at16.6% m.c., while other sections had wheat at 14.4% m.c. Insects were introduced near the middle at 0 cm distance.



Control



(b)

Fig. 4.18 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 7 d in treatment (CO_2 gradients and high moisture content) and in controls (no CO_2 gradients but high moisture content present) in vertical columns of wheat at 30°C. The top sections of the columns (-20 to -40 cm) had wheat at 16.0% m.c., while other sections had wheat at 14.1% m.c. Insects were introduced near the middle at 0 cm distance.

Table 4.10 Comparison of the mean insect movement between treatment (test:OE16.6O14.4H15-6; in CO_2 gradients and high moisture content) and controls (no CO_2 gradients but high moisture content present) in horizontal columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)					
		Treatment				Control**	
		Moisture content (%)	CO_2 (%)	Mean	S.D.	Mean*	S.D.*
1 (closed)	50	14.3 ± 0.0	8.0 ± 0.6	15.6^a	1.8	3.6^b	4.4
2	40	14.3 ± 0.0	7.6 ± 0.6	21.3^a	5.8	2.6^b	3.0
3	30	14.3 ± 0.1	7.3 ± 0.7	17.6^a	8.0	3.0^b	1.5
4	20	14.3 ± 0.6	7.0 ± 0.8	16.3^a	5.6	2.8^b	2.1
7	-10	14.6 ± 0.0	5.7 ± 0.9	2.3^a	3.0	9.1^b	1.9
8	-20	16.5 ± 0.2	5.2 ± 0.9	3.8^a	2.4	15.5^b	3.9
9	-30	16.6 ± 0.1	4.7 ± 0.8	6.3^a	5.2	32.5^b	7.7
10 (open)	-40	16.7 ± 0.1	4.0 ± 0.8	4.8^a	5.1	18.1^b	7.8

S.D*:Standard deviation based on n=6.

** The control columns contained wheat at 16.7 ± 0.1% m.c. in sections 8, 9, and 10, and 14.3 ± 0.1% m.c. in the other sections.

* Means with the same letter (along rows) do not differ significantly at the 5% level.

attractive effect of CO_2 clearly outweighed the effect of moisture content, as 55% of the insects were attracted to higher CO_2 levels compared with only 15% of them moving into the high moisture zone. Also the differences in the insect movement were significant in sections 1, 2, and 3 in the treatment columns as compared to the controls (Table 4.10)

In vertical columns with CO_2 and moisture content gradients in opposing directions (test: T16O14.1V17-6; Table 3.3), the response of ≈81% of the insects was to move down after 1 d, but ultimately ≈41% of them moved up into the region of high moisture content after 7 d (Fig. 4.18). Their movement to the bottom of the grain columns was either due to higher CO_2 concentration at the bottom or due to geotaxism. The treatment columns did not show any significant differences in the insect movement in any of sections, when compared with the controls (Table 4.11).

In inverted vertical columns with CO_2 introduced from the top (test: B16O13.9In V15-6;Table 3.3), 28% of the insects moved up being attracted to CO_2 after 1 and 5 d. In control columns with no CO_2 gradient, only 6% of the insects moved up (Fig. 4.19). The differences in the movement were noticeably significant in sections 1, 2, 3, and 10 between treatment columns and controls (Table 4.12).

In another inverted vertical columns experiment (test: T17O12.8InV15-6; Table 3.3), both high CO_2 and high moisture content wheat were at the top. Approximately 84% of the insects moved up after 1 and 5 d. In the control columns, 65% of them moved up into the high moisture zone after the same period (Fig. 4.20). There were more insects in sections 1, 2, and 3 in the treatment columns than controls (Table 4.13).

Table 4.11 Comparison of the mean insect movement between treatment (test: T16O14.1V17-6;in CO_2 gradients and high moisture content) and controls (no CO_2 gradients but high moisture content present) in vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)					
		Treatment				Control**	
		Moisture content (%)	CO_2 (%)	Mean	S.D.	Mean*	S.D.*
1(bottom)	50	14.0 ± 0.1	8.1 ± 0.0	28.5[a]	13.5	22.3[a]	10.7
2	40	14.0 ± 0.2	7.8 ± 0.1	12.3[a]	6.3	10.1[a]	4.9
3	30	13.9 ± 0.1	7.4 ± 0.1	11.6[a]	3.3	8.6[a]	5.7
8	-20	15.9 ± 0.2	5.8 ± 0.1	7.6[a]	4.8	10.8[a]	7.9
9	-30	16.0 ± 0.0	5.4 ± 0.2	12.5[a]	11.3	15.1[a]	7.9
10 (top)	-40	16.0 ± 0.1	4.9 ± 0.1	8.1[a]	4.7	12.3[a]	10.0

S.D*:Standard deviation based on n=6.

** The control columns contained wheat at 15.9 ± 0.15% m.c. in the top three sections and 14.1 ± 0.2% m.c. in other sections

• Means with the same letter (along rows) do not differ significantly at the 5% level.

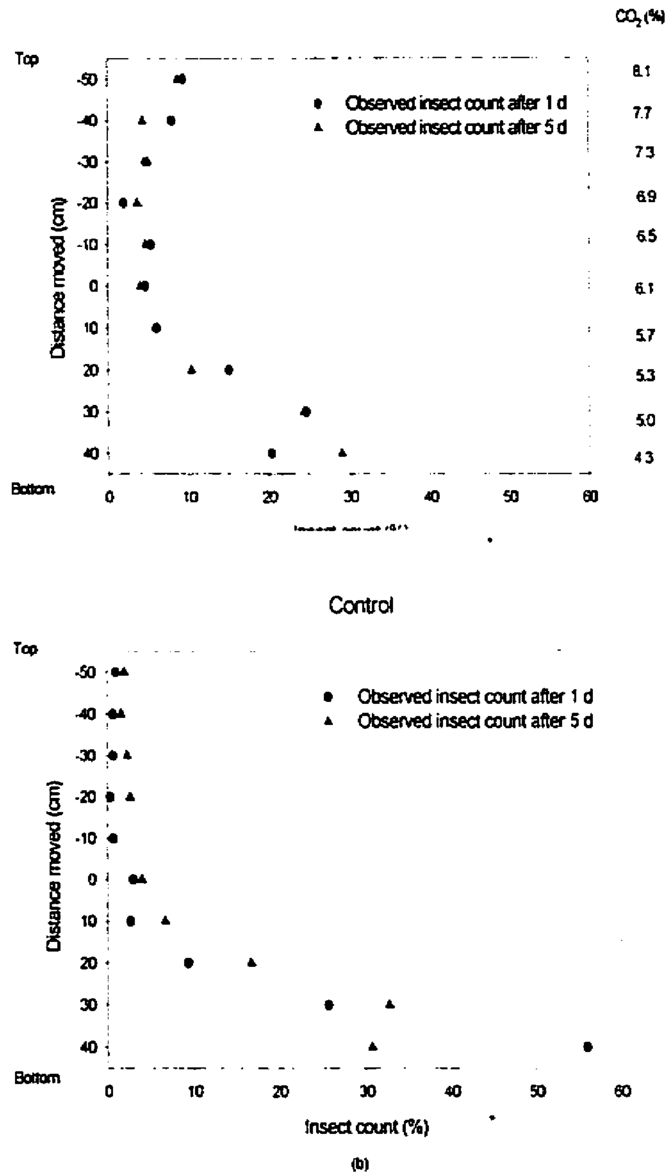
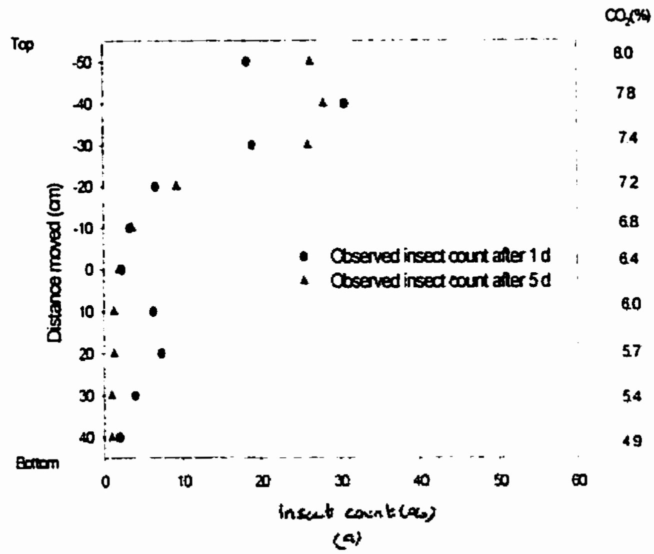


Fig. 4.19 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in treatment (CO_2 gradients and high moisture content) and in controls (no CO_2 gradients but high moisture content present) in inverted vertical columns of wheat at 30°C. The bottom sections of the columns (20 to 40 cm) had wheat at 16.0% m.c., while other sections had wheat at 13.9% m.c. Insects were introduced near the middle at 0 cm distance.



Control

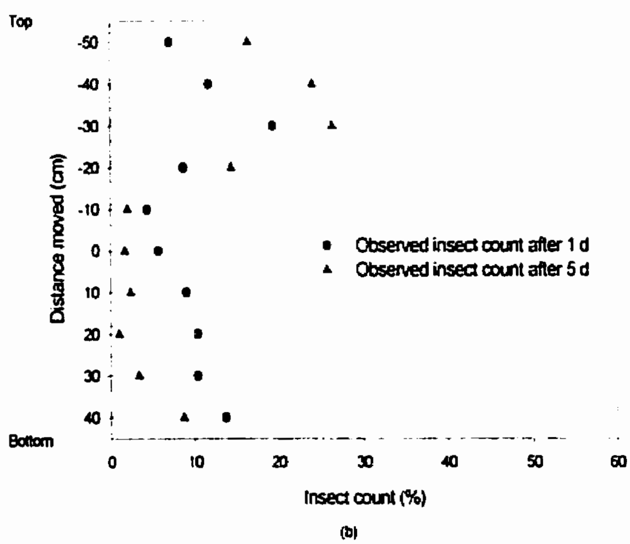


Fig. 4.20 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in treatment (CO_2 gradients and high moisture content) and in controls (no CO_2 gradients but high moisture content present) in inverted vertical columns of wheat at 30°C. The top sections of the columns (-50 to -30 cm) had wheat at 17.0% m.c., while other sections had wheat at 12.8% m.c. Insects were introduced near the middle at 0 cm distance.

Table 4.12 Comparison of the mean insect movement between treatment (Test:B16O13.9InV15-6; in CO_2 gradients and high moisture content) and controls (no CO_2 gradients but high moisture content present) in inverted vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)					
		Treatment				Control **	
		Moisture content (%)	CO_2 (%)	Mean	S.D.	Mean*	S.D.*
1 (top)	-50	13.8 ± 0.1	8.1 ± 0.2	9.0[a]	1.0	1.5[b]	0.8
2	-40	13.8 ± 0.1	7.7 ± 0.0	6.1[a]	2.3	1.1[b]	1.1
3	-30	13.9 ± 0.1	7.3 ± 0.0	4.8[a]	1.1	1.5[b]	1.6
8	20	15.9 ± 0.0	5.3 ± 0.0	12.6[a]	5.2	13.0[a]	4.3
9	30	16.0 ± 0.1	5.0 ± 0.1	24.5[a]	2.8	29.1[a]	5.1
10 (bottom)	40	16.1 ± 0.1	4.3 ± 0.2	24.6[a]	6.8	43.3[b]	14.1

S.D*:Standard deviation based on n=6.

** The control columns contained wheat at 15.9 ± 0.14% m.c. in the bottom three sections and 13.9 ± 0.17% m.c. in the other sections.

* Means with the same letter (along rows) do not differ significantly at the 5% level.

Table 4.13 Comparison of the mean insect movement between treatment (Test:T17O12.8InV15-6); in CO_2 gradients and high moisture content) and controls (no CO_2 gradients but high moisture content present) in inverted vertical columns of wheat at 30 ● 1°C.

Column section	Distance moved (cm)	Insect count (%)					
		Treatment				Control**	
		Moisture content (%)	CO_2 (%)	Mean	S.D.	Mean*	S.D.*
1(top)	-50	17.0 ± 0.2	8.0 ± 0.1	22.3[a]	6.0	11.6[b]	5.6
2	-40	17.2 ± 0.0	7.8 ± 0.1	29.3[a]	7.4	17.8[b]	8.54
3	-30	17.1 ± 0.1	7.4 ± 0.1	22.5[a]	4.3	22.8[a]	8.7
8	20	12.6 ± 0.0	5.7 ± 0.1	4.3[a]	3.6	5.6[a]	5.8
9	30	12.6 ± 0.1	5.4 ± 0.2	2.5[a]	1.7	6.8[a]	5.6
10 (bottom)	40	12.6 ± 0.1	4.9 ± 0.1	1.5[a]	1.3	11.1[b]	7.5

S.D*:Standard deviation based on n=6.

** The control columns contained wheat at 17.2 ± 0.1% m.c. in the top three sections and 12.8 ± 0.3% m.c. in the other sections.

* Means with the same letter (along rows) do not differ significantly at the 5% level.

4.3.2 Insects introduced at the top In vertical columns with CO_2 introduced from the bottom (test: M15.5O12.1V15-t), only 5% of the insects remained at the top at the point of introduction, while 81% of them moved into the high moisture zone after 1 and 5 d. The rest (14%) crossed the high moisture zone and proceeded down either because of the geotactic effect or being attracted to higher CO_2. In the control columns with no CO_2 gradient, the percentage of insects remaining at the top was 23 (Fig. 4.21). There was a significant difference in insect movement between the treatment and controls in the bottom portion of the columns (Table 4.14).

4.3.3 Regression model Four polynomial models were examined for fitting the experimental data (Appendix C (Table C11 and C12)). The dependent variable 'insect count' was transformed to 'Log (insect count)' to get a better fit. The independent variables were: distance, time, m.c and CO_2. The models were compared using coefficient of determination (R^2), mean relative percent error (e), standard error of estimates (SE), and distribution of residuals. The analysis showed that the cubic model 'RegMCCO$_2$ -5a' gave the highest R^2 value and better distribution of residuals than the other models (Fig. 4.22). The SE value was marginally higher than the other two models, but 'e' value was the lowest. Also, the plots of insect count versus individual independent variable showed a cubic relationship and it was, therefore, concluded that this model gave the best description of the relationship between the insect count and the other variables in the grain columns. The best estimations of the insect count were obtained with this model.

The best fit equation is:

$$\text{Log (insect count)} = 59.781 + 0.001\,x + 0.005\,t + 0.178\,CO_2 - 13.530\,(\text{m.c.}) + 0.988\,(\text{m.c.})^2 - 0.023\,(\text{m.c.})^3 \qquad (3)$$

when $15.5\% < \text{m.c.} < 17\%$, and $2\% < CO_2 < 9\%$.

The R^2 value of the model is 0.56. The F-value of the overall model (19.35, $P<0.01$) indicated that the model was significant. The effects of moisture content, CO_2, and distance on the insect count were all significant ($P<0.05$). The effect of moisture content was more significant than CO_2. The effect of time was, however, non-significant ($P>0.05$). Based on the equation (3), the predicted insect count was determined for all the experimental data and compared with the measure insect count (Appendix C, Figs. C1 - C5).

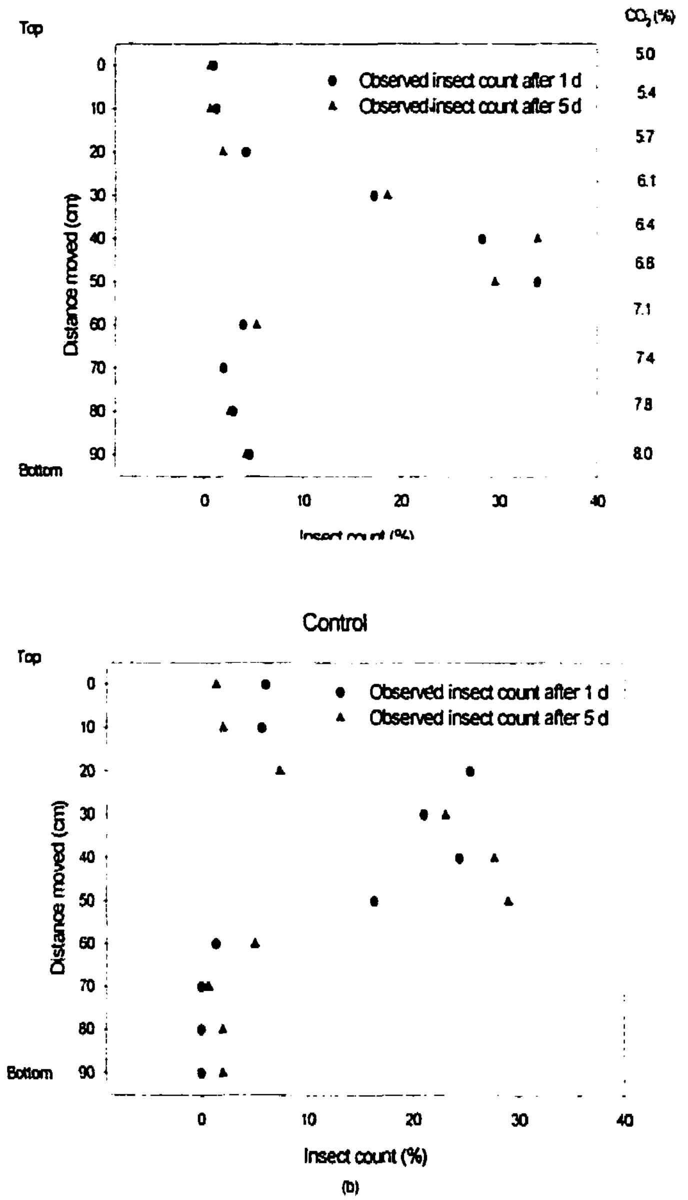


Fig. 4.21 Mean movement (three replicates) of adult *Cryptolestes ferrugineus* after 1 and 5 d in treatment (CO_2 gradients and high moisture content) and in controls (no CO_2 gradients but high moisture content present) in vertical columns of wheat at 30°C. The middle sections of the columns (30 to 50 cm) had wheat at 15.5% m.c., while other sections had wheat at 12.1% m.c. Insects were introduced near the middle at 0 cm distance.

Table 4.14 Comparison of the mean insect movement between treatment (Test:M15.5O12.1V15-t; in CO_2 gradients and high moisture content) and controls (no CO_2 gradients but high moisture content present) in vertical columns of wheat at 30 ± 1°C.

Column section	Distance moved (cm)	Insect count (%)					
		Treatment				Control **	
		Moisture content (%)	CO_2 (%)	Mean	S.D.	Mean*	S.D.*
1 (bottom)	90	12.1 ± 0.0	8.0 ± 0.1	4.5^a	2.2	1.0^b	1.2
2	80	12.1 ± 0.1	7.8 ± 0.1	2.8^a	1.7	1.0^a	1.0
3	70	12.0 ± 0.0	7.4 ± 0.1	2.0^a	1.2	0.3^b	0.8
5	50	15.5 ± 0.1	6.8 ± 0.1	31.8^a	4.1	22.6^b	7.6
6	40	15.5 ± 0.1	6.4 ± 0.0	31.1^a	4.9	26.0^a	6.0
8	20	12.3 ± 0.0	5.7 ± 0.1	3.1^a	2.3	16.3^b	10.3

S.D*:Standard deviation based on n=6.

** The control columns contained wheat at 15.5 ± 0.1% m.c. in sections 5, 6, and 7, and 12.2 ± 0.1% m.c. in the other sections.

* Means with the same letter (along rows) do not differ significantly at the 5% level.

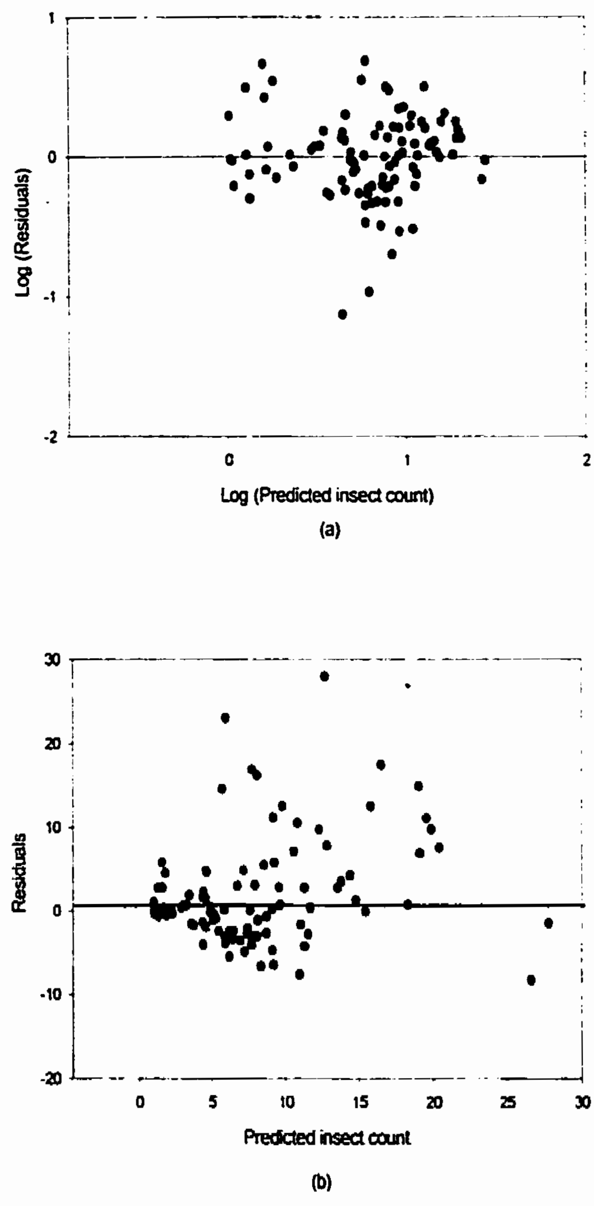


Fig. 4.22 Residual plots obtained with the best fit equation (3)

5. DISCUSSION

5.1 Insect movement in grain columns with pockets of high moisture content wheat

Rusty grain beetles introduced in vertical grain columns filled with pockets of high moisture content wheat at the top or near the middle, aggregated in the region of high moisture content, exhibiting a hygrotactic response. In these columns, they also showed a secondary preference to move down. In horizontal control columns, they showed a trend toward distributing themselves uniformly after 5 d, but in vertical control columns, they consistently showed a geotactic effect. This remained independent of their point of introduction, i.e., irrespective of whether they were introduced at the top or near the middle in these columns.

When high moisture content wheat was above the point of introduction and insects were introduced near the middle in vertical columns, their movement into the high moisture zone was gradual, taking more than 3 d (Fig. 4.5). This may be either because they detected the high moisture content after 3 d, or, because they had to move up against their natural tendency of moving down in the grain columns. Freedman et al. (1982) had reported that several stored-product insects are oriented to stored grain odor emitted by the wet grain.

They moved very little, 10 to 20 cm , on either side, on day 1, when they were introduced directly into the high moisture zone. Where the difference between the two moisture content used was high, the percentage of insects preferring to stay in the high moisture zone for more than 1 d was high, but with a marginal difference in the two moistures used, insect movement was observed right after the day 1 in grain columns (tests: T18.2O14.8V15-t and M16.9O14.8V1357-6).

Similar results were observed by Loschiavo (1983) in his study on movement of adult rusty grain beetles in vertical grain columns at 25°C. He used wheat of 17% m.c. in the middle sections and 13.4 % m.c. in other sections in one of his experiments, while in another experiment, he used 16.5% m.c. wheat at the top and 12.5% in other sections (Fig. 5.1 and 5.2). Insects were introduced at the top. The slight differences in insect movement between my test and that of Loschiavo (1983) might be due to the difference in the temperatures at which the tests were performed.

A second noteworthy feature of the experiments performed with pockets of high moisture content wheat was that it indicated a decline in locomotor activity of adults of *C. ferrugineus* with time. Although this activity was high when beetles moved from dry to high moisture content wheat, continuous exposure to high moisture content resulted in rapid decline in activity. The decrease in level of activity with time may be due to an ortho-kinetic (movement in response to stimulus) adaptation, such as shown by adults of *Tenebrio molitor* in their reaction to high humidity (Ewer and Bursell 1950).

In grain columns of uniform moisture content wheat, adult *C. ferrugineus* showed a geotactic response. This indicates that insects can penetrate to the bottom of dry wheat in a grain bin. It also explains the fact that infestations can develop deep in a pile of apparently dry wheat. Adult *C. ferrugineus* also showed a geotactic response in grain columns filled with high and low moisture content wheat. This suggests that surface infestations may result in the establishment of insects at various depths in a bulk of wheat.

In many grainaries, higher moisture often accumulates at the floor of the bin which may be due to snow or rain entering the bin. Geotactic response exhibited by beetles will

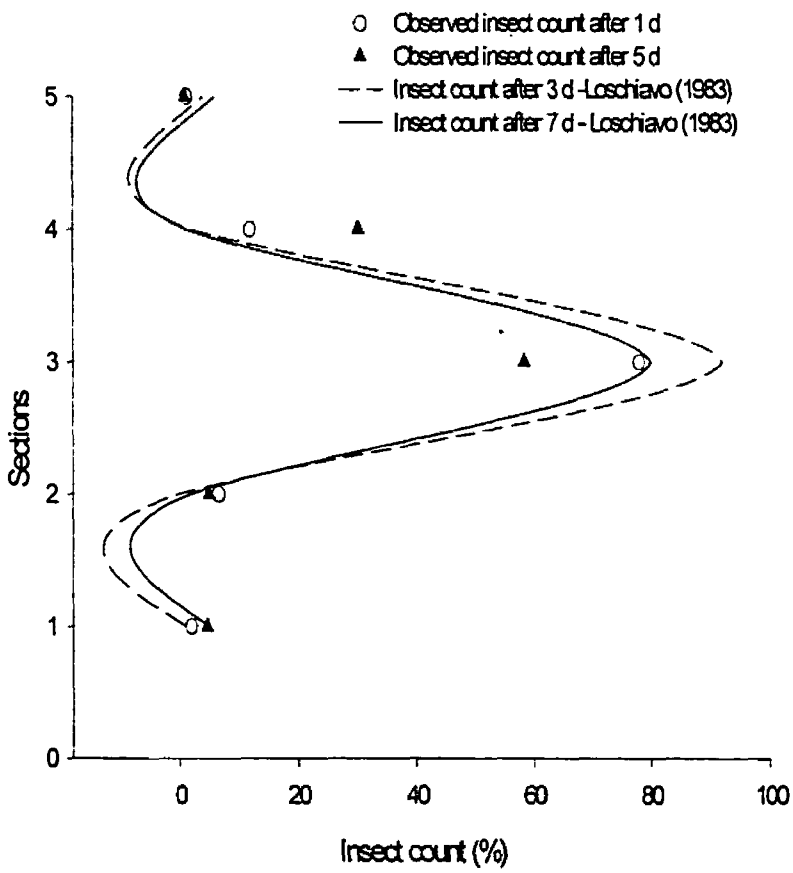


Fig.5.1 Comparison of the insect movement in vertical columns between my test M16.5O12V15-t (at 30°C) and that performed by Loschiavo (1983). Vertical columns used by Loschiavo (1983) had 17% m.c. wheat in the middle section and 13.4% m.c. wheat in other sections at 25°C. Two sections of the vertical columns in my test and five sections of the columns used by Loschiavo (1983), were combined into one to compare the results.

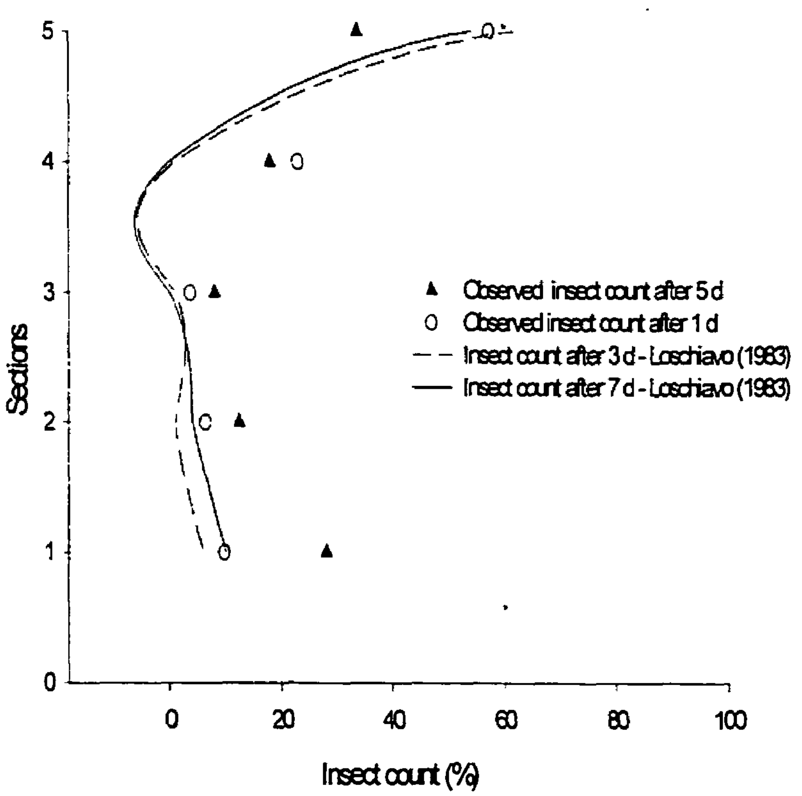
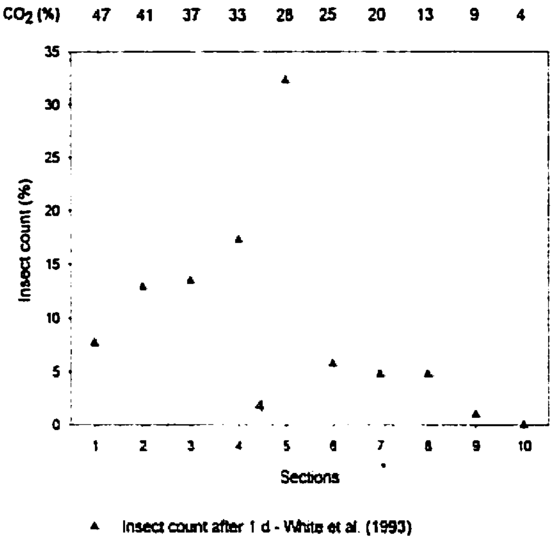


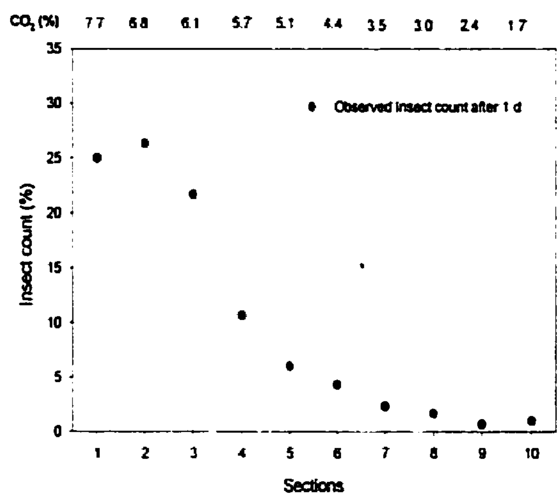
Fig. 5.2 Comparison of the insect movement in vertical columns between my test T18.2O14.8V15-t (at 30°C) and that of Loschiavo (1983). Vertical columns used by Loschiavo (1983) had 16.5% m.c. wheat at the top and 12.5% m.c. wheat in other sections at 25°C. Two sections of the vertical columns in my test and five sections of the columns used by Loschiavo (1983), were combined into one to compare the results.

bring them down to the bottom of the bin where high moisture may enhance their development. Sampling at the bottom of the bin may, therefore, increase the chances of detecting infestation which could then be controlled by taking suitable measures. The regression model of the insect count gave predicted values which agreed fairly well with (R^2= 0.64) the observed values. These predicted values of the insect count could tell the level of infestation at any particular point in a grain bin when moisture content varied from 16.5% to 18.2%.

5.2 Insect movement under CO_2 gradients in grain columns Adult rusty grain beetles introduced into the grain columns purged with CO_2 from any end of the column, were attracted to higher levels of CO_2. In the horizontal columns, their attraction could be clearly seen, as more than 65% of the insects moved towards higher CO_2. The same phenomenon was noticed in inverted vertical columns. In vertical columns, when insects were introduced in the middle, they moved at a faster rate down the columns where higher CO_2 levels existed compared to the controls. These results could be compared with similar results observed by White et al. (1993) in their experiments on movement of adult rusty grain beetles under CO_2 gradients. Their results in horizontal columns are compared with my results (Fig. 5.3 and 5.4). Although CO_2 gradients in White et al. (1993) varied from 47% to 3% in 1 and 3 d experiments, the trend indicated that rusty grain beetles were attracted to higher levels of CO_2. Boistel (1960, cited by Nicolas and Sillans 1989) had stated that pure CO_2 inhibits the movement of insects, but a smaller concentration (15%) stimulates it. In all my experiments, I used a 10% CO_2:90% air mixture which produced gradients ranging from 2 to 9% of CO_2.

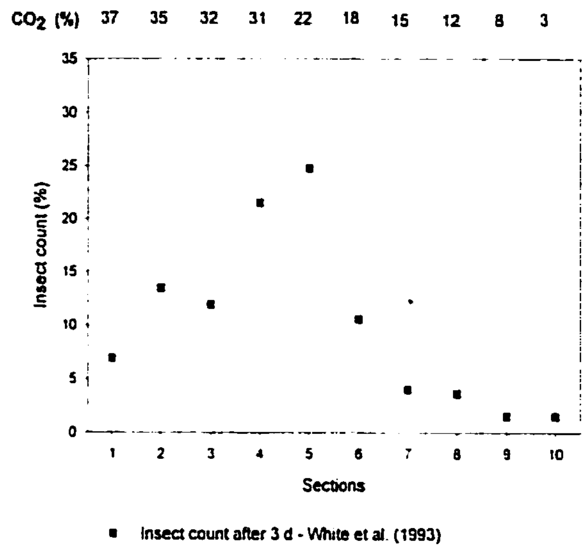


a) After 1 d - White et al. (1993)

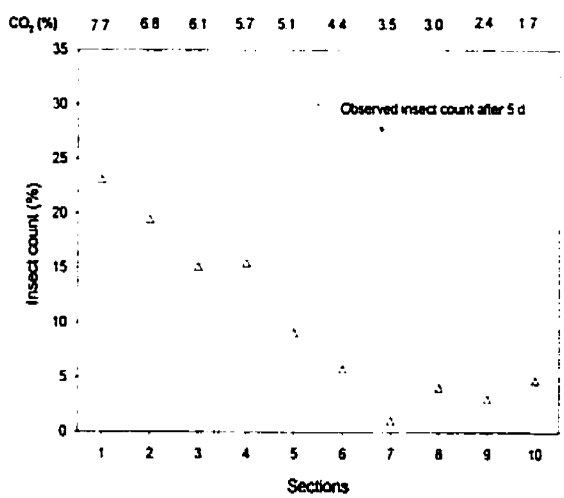


b) After 1 d- present study

Fig. 5.3 Comparison of insect movement in horizontal columns under CO_2 gradients between the current study and that of White et al. (1993). Hundred insects were introduced in section 5 in (a) and in section 6 in (b).



c) After 3 d -White et.al (1993)



d) After 5 d-present study

Fig. 5.4 Comparison of insect movement in horizontal columns under CO_2 gradients between the current study and that of White et al. (1993). Hundred insects were introduced in section 5 in (c) and in section 6 in (d).

The levels of concentration of biological CO_2 produced by the insects and grain in grain bins varies from 5 to 18% (White and Sinha 1980, White et al. 1982).These levels are similar to the levels used in my experiments. Insects in my experiments did not show any repulsive effect.

In a controlled atmosphere storage, keeping the bin airtight for a longer period is a problem. There are always some leakages from which CO_2 gas can escape. Insects may get attracted towards low CO_2 levels, if the purging process lasts for a longer time. They may survive the treatment, if they remain in the region of low CO_2 concentrations. A possibility that insects may re-infest the grain, after the normal atmospheric composition is restored, cannot be ruled out. This will, of course, depend on the prevailing CO_2 concentration and exposure time in the controlled atmosphere storage. Nevertheless, this suggests that CO_2 concentration should be held long enough in the maintenance phase to achieve maximum efficiency in the production of a lethal atmosphere after purging has been accomplished.

The tests conducted under CO_2 gradients also showed that adult rusty grain beetles were attracted to CO_2 levels produced biologically in the grain columns. Carbon dioxide being heavier than air settles at the bottom of the bin (White et al. 1990). Rusty grain beetles showing geotactic response and also attraction to CO_2 move down to the bottom of the bin where CO_2 concentration is the highest.

The regression model could predict ($R^2 = 0.72$) the insect count for concentrations of CO_2 less than 9%.

5.3 Insect movement under CO_2 gradients with pockets of high moisture content wheat in grain columns Insects introduced in horizontal grain columns with high moisture content at one end and high CO_2 at the other were found to prefer higher CO_2 levels. Higher moisture content affected insect movement, but only after 3 d. Similar results were observed for insect movement in columns with pockets of high moisture content wheat (sec. 4.1). There was no introduced CO_2 there, but it still took time for the insects to detect the high moisture content, particularly when they had to move up in the grain columns. Here, the effect of CO_2 was more pronounced than that of high moisture content.

In the vertical columns test with high moisture content at the top and high CO_2 levels at the bottom (T16O14.1V17-6), insects behaved in the same way as in the controls (Table 4.11). However, after 1 d, there were more insects at the bottom of the treatment columns than the controls (Fig. 4.18). This indicate that insects moved down speedily after 1 d in the treatment columns due to higher CO_2 levels at the bottom, and then moved up and became less evenly distributed in the columns after 7 d (Fig. 4.18). In the results observed by White et al. (1993) on the insect movement under CO_2 gradients in vertical columns (insects introduced in the middle) with no high moisture content at the top, they found that insects moved down faster to the bottom. In my experiments on 'insect movement under CO_2 gradients in uniform moisture content wheat (test: U14.6V1735-6)', insects moved one and half times faster toward the bottom of the columns and toward the higher CO_2 levels compared with the controls. This clearly shows that when high m.c is above them, insects tend to move up after a certain period. This may explain why an infestation starting at the bottom of the bin subsequently spreads to all the parts of the bin where increase in moisture may result from moisture migration to the top or central portion of a bulk.

In the inverted vertical columns with higher CO_2 levels at the top and high moisture content wheat at the bottom (test: B16O13.9InV15-6), insects moved down to the bottom of the columns, but their number was lower than those observed at the bottom of the control columns (Table 4.12). This was mainly because higher CO_2 levels held the insects up, slowing down their speed to move down. In my earlier test on 'insect movement in inverted vertical columns in CO_2 gradients (U12InV15-6),' insects were attracted to CO_2 at the top in the absence of high moisture content at the bottom (Table 4.8). In this test (B16O13.9InV15-6), however, there were two factors acting on the insect. The more important was geotaxism and the less important was high moisture content

The test (T17O12.8InV15-6) clearly demonstrated the combined effects of high moisture content. and CO_2 (Fig. 4.20). Insects moving down were very few. These two factors outweighed the effect of geotaxism.

The test (M15.5O12.1V15-t) (Fig. 4.21) showed that once the insects are in the zone of high m.c. then their locomotor activity declines. Despite this, 14% of the insects crossed the high moisture zone and moved down. This may be because of the combined two factors acting on them; their natural tendency to move down, and higher CO_2 levels.

The regression model showed a stronger effect of high moisture content ($P < 0.0001$) than CO_2 ($P < 0.01$) in causing the movement. This may be mainly because in most of my tests, the effect of high moisture content was strengthened by the effect of geotaxism. This was the factor which could not be taken into account in my regression model. In horizontal columns, where CO_2 levels outweighed the effect of high moisture content. this effect was minimum. In all other tests in vertical columns, insects moved down in response to geotaxism and high moisture content or CO_2 gradients. Perhaps, this could explain the reason for obtaining an R^2 value of 0.56 in my regression model.

6. CONCLUSIONS

The conclusions that can be drawn from this study are:

1. An average of 55% of adult *Cryptolestes ferrugineus* aggregated in the high moisture zone in both horizontal and vertical columns, exhibiting a positive hygrotactic response.
2. Where the difference between the two moisture contents used in the vertical columns was small (e.g.,16.6% and 14.8%), adult *C. ferrugineus* took time (3 d) to detect the high moisture zone, but eventually, 45% moved to the zone of high moisture content.
3. In vertical columns filled with uniform moisture content wheat, 65% of the beetles exhibited a positive geotactic response.
4. In horizontal columns containing uniform moisture content wheat, 43% of adult *C. ferrugineus* moved initially into the adjacent sections from the point of introduction, but subsequently, after 5 d, they exhibited a nearly uniform distribution pattern.
5. In horizontal and vertical columns filled with uniform moisture content wheat, 65 and 60% of beetles, respectively, moved towards high levels of CO_2 under CO_2 gradients (2 to 9%).
6. In horizontal columns containing pockets of high moisture content wheat at one end and high CO_2 levels at the other end, 55% of adult *C. ferrugineus* moved towards higher levels of CO_2, when introduced in the middle.
7. In vertical columns filled with pockets of high moisture content wheat under CO_2 gradients, combined effects of any two factors, out of the three, viz., gravity, higher levels of CO_2 and high moisture content, outweighed the effect of any single factor in influencing the insect movement.

7. RECOMMENDATIONS FOR FUTURE STUDY

Following studies should be conducted in continuation of this study:

1. The movement of the adult rusty grain beetles should be studied in a two dimensional and three dimensional cases, preferably in a bin with wheat containing pockets of high moisture content and subjected to CO_2 gradients.
2. Based on this study, a three dimensional model on the movement of the adult rusty grain beetles should be developed and validated for use in a stored-grain ecosystem model.

8. REFERENCES

Adler, C. 1992. Vertical dispersion of adult *Sitophilus granarius* (L.) (Coleoptera: Curculiondae) in a wheat column flushed with modified atmospheres. *Journal of Stored Products Research* 28:201-209.

Agrawal, N.S., C. M. Christensen and A.C. Hudson. 1957. Grain storage fungi associated with the granary weevil. *Journal of Economic Entomology* 50:659-663.

ASAE. 1997a. Moisture measurement - Unground grain and seeds. ASAE S352.2. In *ASAE Standards* 1997, 555. St. Joseph, MI: ASAE.

ASAE. 1997b. Moisture relationships of plant-based agricultural products. ASAE D245.5. In *ASAE Standards* 1997, 500. St. Joseph, MI: ASAE.

Barrer P. M. 1983. A field demonstration of odor-based, host food finding behavior in several species of stored grain insects. *Journal of Stored Products Research* 19:105-110.

Barrer, P. M. and E. G. Jay. 1980. Laboratory observations on the ability of *Ephestia cautella* (Walker) (Lepidoptera: Phycitidae) to locate, and to oviposit in response to a source of grain odor. *Journal of Stored Products Research* 16:1-7.

Barrer, P. M., N. T. Starick, R. Morton and E. J. Wright. 1993. Factors influencing initiation of flight by *Rhizopertha dominica* (F.) (Coleoptera: Bostrichidae). *Journal of Stored Products Research* 29:1-5.

Birch, L. C. 1946. The movements of *Calandra oryzae* L. (Small strain) in experimental bulks of wheat. *Journal of Australian Institute of Agriculture Science* 11:21-26.

Boistel, J. 1960. *Characteristiques fonctionelles des fibres nerveuses et des recepteurs tactiles et olfictifs des insectes.* Paris: Arnette.147 pp. (cited by Nicolas and Sillans 1989).

Borden, J. H. 1974. Aggregation pheromones in the Scolytidae. In *Pheromones*, ed. M.C. Birch, 135-160. Amsterdam, NL: North-Holland Publishing Company.

Borden, J. H., M. G. Dolinski, L. Chong, V. Verigin, H. D. Pierce (Jr.) and A. C. Oehlshlager. 1979. Aggregation pheromone in the rusty grain beetle, *Cryptolestes ferrugineus* (Coleoptera:Cucujidae). *Canadian Entomologist* 111:681-688.

Bothast, R. J. 1978. Fungal deterioration and related phenomena in cereals. In *Postharvest Biology and Biotechnology*, eds. H. O. Hultin and M. Milner, 210-243. Westport, CT: Food and Nutrition Press.

Bursell, E. 1974. Environmental aspects - humidity. In *The Physiology of Insecta*, ed. M. Rockstein, 43-84. New York/ London: Academic Press.

Canada Grains Council. 1998. *Statistical handbook*, Winnipeg, MB: Canada Grains Council.

Cloudsley-Thomson, J. L. 1953. Studies in diurnal rhythms. IV. Photoperiodism and geotaxis in *Tenebrio molitor* L. (Coleoptera: Tenebrionidae). *Proceedings of Royal Entomological Society of London* 28:117-132.

Cogburn, R. R., W. E. Burkholder and H. J. Williams. 1984. Field tests with the aggregation pheromone of the lesser grain borer (Coleoptera: Bostrichidae). *Environmental Entomology* 13:162-166.

Cotton, R. T. 1954. Insects. In *Storage of Cereal Grains and Their Products*, ed. J.A. Anderson and A.W. Alcock, 221-274. St. Paul, MN: American Association of Cereal Chemists, Inc.

Cotton, R. T., H. H. Walkden, G. D. White and D. A. Wilbur. 1960. Causes of outbreak of stored grain insects. *Kansas Agriculture Experiment Station Bulletin* 416, 35p.

Crombie, A. C. 1941. On ovipostion, olfactory conditioning and host selection in *Rhizopertha dominica* Fab. (Coleoptera: Bostrichidae). *Journal of Experimental Biology* 18:62-79.

Crombie, A. C. 1944. On intraspecific and interspecific competition in larvae of graminivorous insects. *Journal of Experimental Biology* 20:135-151.

Dolinski, M. G. and S. R. Loschiavo. 1973. The effect of fungi and moisture on the locomotory behavior of the rusty grain beetle, *Cryptolestes ferrugineus* (Coleoptera: Cucujidae). *Canadian Entomologist* 105:485-490

Dowdy, A. K. 1994. Flight initiation of lesser grain borer (Coleoptera: Bostrichidae) as influenced by temperature, humidity, and light. *Journal of Economic Entomology* 87:1714-1717.

Ewer, D. W. and E. Bursell. 1950. A note on the classification of elementary behaviour patterns. *Behaviour* 3:40-47.

Flinn, P. W. and D. W. Hagstrum. 1998. Distribution of *Cryptolestes ferrugineus*

(Coleoptera: Cucujidae) in response to temperature gradients in stored wheat. *Journal of Stored Products Research* 34:107-112.

Freedman, B., K. L. Mikolajczak, C. R. Smith, Jr., W. F. Kwolek and W. E. Burkholder. 1982. Olfactory and aggregation responses of *Oryzaephilus surinamensis* (L.) to extracts from oats. *Journal of Stored Products Research* 18:75-82.

Gunn, D. L. and H. S. Hopf. 1942. The biology and behavior of *Ptinus tectus Boie.* (Coleoptera: Ptinidae), a pest of stored products. II. The amount of locomotory activity in relation to experimental and previous temperatures. *Journal of Experimental Biology* 18:278-289.

Hagstrum, D. W., P. W. Flinn and J. J. Gaffney. 1998. Temperature gradient on *Tribolium castaneum* (Coleoptera: Tenebrionidae) adult dispersal in stored wheat. *Environmental Entomology* 27:123-129.

Henson, W. R. 1964. Laboratory studies on the adult behavior of *Conophthorus coniperdus* (Coleoptera: Scolytidae). IV. Responses to temperature and humidity. *Annals of Entomological Society of America* 57:77-85.

Howe, R. W. 1951. The movement of grain weevils through grain. *Bulletin of Entomological Research* 42:125-134.

Howe, R. W. 1943. An investigation of the changes in a bin of stored wheat infested by insects. *Bulletin of Entomological Research* 34:145-158

Jayas, D. S. 1995. Mathematical modeling of heat, moisture, and gas transfer in stored-grain ecosystems. In *Stored -Grain Ecosystems*, eds. D. S. Jayas, N. D. G. White and W. E. Muir, 527-568. New York, NY: Marcel Dekker, Inc.

Leos-Martinez, J., T. A. Granovsky, H. J. Williams, S. B. Vinson and W. E. Burkholder. 1986. Estimation of aerial density of the lesser grain borer (Coleoptera:Bostrichidae) in a ware house using dominicalure traps. *Journal of Economic Entomology* 79:1134-1138.

Leos-Martinez, J., T. A. Granovsky, H. J. Williams, S. B. Vinson and W. E. Burkholder. 1987 Pheromonal trapping methods for lesser grain borer, *Rhyzopertha dominica* (Coleoptera: Bostrichidae). *Environmental Entomology* 16:747-751.

Liscombe, E. A. R. and F. L. Watters. 1962. Insect and mite infestations in empty granaries in the prairie provinces. *Canadian Entomologist* 94:433:441.

Loschiavo, S. R. 1974. Laboratory studies of a device to detect insects in grain, and of the distribution of adults of the rusty grain beetle, *Cryptolestes ferrugineus* (Coleoptera: Cucujidae), in wheat- filled containers. *Canadian Entomologist* 106:1309-1318

Loschiavo, S. R. 1983. Distribution of the rusty grain beetle (Coleoptera: Cucujidae) in columns of wheat stored dry or with localized high moisture content. *Journal of Economic Entomology* 76:881-884.

McGregor, H. E. 1964. Preference of *Tribolium castaneum* for wheat containing various percentages of dockage. *Journal of Economic Entomology* 57:511-513.

Muir, W. E. and N. D. G. White. 1999. Insects and mites in stored grain. In *Grain Preservation Biosystems*, ed. W. E. Muir, 5-1-9.Winnipeg, MB: Department of Biosystems Engineering. University of Manitoba.

Navarro, S., M. Gonen, Y. Kasanchi and H. Frandji.. 1976. The use of controlled atmosphere for the control of stored product insects: I Experiments in a flat metal bin. In *Progress Report on Israel Agriculture Research Organization Stored Products Division*, 85-103 (cited by Navarro et al. 1981).

Navarro, S., T. G. Amos and P. Williams. 1981. The effect of oxygen and carbon dioxide gradients on the vertical dispersion of grain insects in wheat. *Journal of Stored Products Research* 17:101-107.

Nicolas, G., and D. Sillans. 1989. Immediate and latent effects of carbon dioxide on insects. *Annual Review of Entomology* 34:97-116.

Oxley, T. A. 1949. The movement of heat and water in stored grain. *Transactions of American Association of Cereal Chemists* 6:84-100.

Parpia. H. A. B. 1976. Postharvest losses- Impact of their prevention on food supplies, nutrition,and development. In *Nutrition and Agriculture Development*, eds. N. S. Scrimshaw and M. Behar, New York, NY: Plenum Press.

Perttunen, V. and L. Pahoheimo. 1964. Locomotory activity of *Tenebrio molitor* L. (Coleoptera: Tenebrionidae) at different temperatures and light intensities. *Annales Entomologici Fennici* 30:156-172.

Pomeranz, Y. 1985. *Functional Properties of Food Components*. Orlando, FL: Academic Press.

Pruthi, H. S. and M. Singh. 1945. Stored grain pests and their control. *Miscellaneous Bulletins Council of Agriculture Research (India)* 57:39-42.

SAS. 1988. *SAS User's Guide: Statistics*. Version 6.0 Statistical Analysis System Inc., Raleigh, NC

Sharangpani, M. V. and S. V. Pingale. 1956. A study of the movement of some insect pests through grain stored in bags. *Indian Journal of Entomology* 18:243-250.

Shejbal, J., A. Tonolo and G. Careri. 1973. Conservation of wheat in silos under nitrogen. *Annales de Technologie. Agricole* 22:773-785.

Shorey, H. H. 1973. Behavioral responses to insect pheromones. *Annual Review of Entomology* 18:349-380.

Sinclair, E. R. and R. L. Haddrell. 1985. Flight of stored products beetles over a grain farming area in southern Queensland. *Journal of Australian Entomological Society* 24:9-15.

Sinha, R. N. 1965. Development of *Cryptolestes ferrugineus* (Stephens) and *Oryzaephilus mercator* (Fauvel) on seed borne fungi. *Entomologia Experimenta et Applicata* 8:309-313

Sinha, R. N. 1966. Development and mortality of *Tribolium castaneum* and *T. confusum* (Coleoptera: Tenebrionidae) on seed-borne fungi. *Annals of Entomological Society of America* 59:192-201.

Sinha, R. N. 1971. Fungus as food for stored- product insects. *Journal of Economic Entomology* 64: 1-6.

Sinha, R. N., D. Waterer and W. E. Muir. 1986. Carbon dioxide concentrations associated with insect infestations of stored grains. 1. Natural infestation of corn, barley, and wheat in farm granaries. *Sciences des Aliments* 6: 91-98.

Smith, L. B. 1983. The relationship between wet grain, *Cryptolestes ferrugineus* (Coleoptera: Cucujidae) populations, and heating in wheat stored in granaries. *Canadian Entomologist* 115:1383-1394.

Solomon, M. E.1951. Control of humidity with pottasium hydroxide, sulphuric acid, or other solutions. *Bulletin of Entomological Research* 42: 543-554.

Surtees, G. 1964. Laboratory studies on dispersion behavior of adult beetles in grain. XII. The effect of isolated pockets of damp and moldy wheat on *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Cucujidae). *Bulletin of Entomological Research* 55:673-680.

Suzuki, T. 1985. Presence of another aggregation pheromone substance(s) in the frass of red flour beetles, *Tribolium castaneum* (Coleoptera, Tenebrionidae). *Applied Entomology and Zoology*. 20:90-91.

Throne, J. E. 1995. Computer modeling of the population dynamics of stored-product pests. In *Stored-Grain Ecosystems*, eds. D. S. Jayas, N. D. G. White and W. E. Muir, 169-196. New York, NY: Marcel Dekker, Inc.

Voute, A. D. 1937. Bevolkingsproblemen II Emigratie van *calandra oryzae* L. Natuurk. Tijdschr. Ned.-Ind. 97:210-213 (cited by Watters 1969)

Watters, F. L. 1969. The locomotor activity of *C. ferrugineus* (Stephens) (Coleptera:Cucujidae) in wheat. *Canadian Journal of Zoology* 47:1177-1182.

White, N. D. G. and R. N. Sinha. 1980. Changes in stored-wheat ecosystems infested with two combinations of insect species. *Canadian Journal of Zoology* 58:1524-1534.

White, N. D. G., D. S. Jayas and R. N. Sinha. 1990. Carbon dioxide as a control agent for the rusty grain beetle (Coleoptera: Cucujidae) in stored grain. *Journal of Economic Entomology* 83:277-288.

White, N. D. G., D. S. Jayas and W. E. Muir. 1995. Toxicity of carbon dioxide at biologically producible levels to stored-product beetles. *Environmental Entomology* 24:640-647.

White, N. D. G., R. N. Sinha and W. E. Muir. 1982. Intergranular carbon dioxide as an indicator of biological activity associated with the spoilage of stored wheat. *Canadian Agricultural Engineering* 24:35-42.

White, N. D. G., R. N. Sinha, D. S. Jayas and W. E. Muir. 1993. Movement of *Cryptolestes ferrugineus* (Coleoptera:Cucujidae) through carbon dioxide gradients in stored wheat. *Journal of Economic Entomology* 86:1846-1851.

Williams, H. J., R. M. Silverstein, W. E. Burkholder and A. Khorramshahi. 1981. Dominicalure 1 and 2: components of aggregation pheromone from male lesser grain borer *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae). *Journal of Chemical Ecology* 7:759-780.

Yinon, U. and A. Shulov. 1970. The dispersion of *Trogoderma granarium* in a temperature gradient and comparison with other stored product beetles. *Entomologia Experimentalia* et *Applicata* 13:107-121.

Appendix A

Table A1 Adult *Cryptolestes ferrugineus* count in test: M17.5O13.6H1357-6*.

After 1 and 3 d

Section	Insect count (%)						Moisture content		
	After 1 d			After 3 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	1	5	7	5	5	1	13.5	13.5	13.7
2	16	3	2	13	6	10	13.4	13.4	13.5
3	13	9	15	14	8	4	13.8	13.7	13.9
4	47	37	32	30	26	31	17.1	17.3	17.2
5	14	33	28	24	31	34	17.8	17.7	17.9
6	8	9	14	10	15	9	17.3	17.6	17.3
7	0	4	1	2	6	3	13.7	14.3	14.1
8	0	0	0	1	1	4	13.5	13.9	13.6
9	1	0	0	0	1	1	13.5	13.6	13.4
10	0	0	0	1	1	2	13.5	13.7	13.6

After 5 and 7 d

Section	Insect count (%)						Moisture content		
	After 5 d			After 7 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	6	3	10	5	3	9	13.5	13.8	13.8
2	8	10	4	6	6	5	13.5	13.7	13.6
3	13	9	7	10	12	13	13.8	13.8	13.8
4	14	12	21	13	18	11	17.3	17.6	17.5
5	13	16	21	13	15	16	17.8	17.7	17.6
6	15	17	19	22	13	25	17.5	17.4	17.4
7	21	12	6	19	15	7	13.9	13.8	13.8
8	1	4	5	4	6	7	13.6	13.7	13.7
9	3	8	3	2	10	4	13.5	13.6	13.6
10	6	9	4	6	2	3	13.6	13.6	13.5

* For description of the code, refer Table 3.1.

Table A2 Adult *Cryptolestes ferrugineus* count in test: U13.5H1357-6 (Control)*.

After 1 and 3 d

Section	Insect count (%)						Moisture content		
	After 1 d			After 3 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	2	5	10	20	16	19	13.4	13.4	13.4
2	13	11	11	7	19	16	13.6	13.6	13.6
3	17	15	18	8	5	9	13.6	13.5	13.5
4	22	19	16	8	7	7	13.6	13.6	13.6
5	12	13	8	7	5	5	13.5	13.4	13.4
6	12	14	10	7	6	4	13.6	13.5	13.4
7	9	11	7	9	4	8	13.5	13.6	13.6
8	5	2	7	6	7	8	13.6	13.6	13.5
9	6	9	5	11	25	15	13.4	13.5	13.5
10	2	1	8	17	6	9	13.5	13.5	13.5

After 5 and 7 d

Section	Insect count (%)						Moisture content		
	After 5 d			After 7 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	10	10	16	15	11	14	13.7	13.6	13.3
2	12	8	17	10	9	15	13.6	13.6	13.5
3	10	14	5	10	10	6	13.5	13.6	13.5
4	13	10	7	3	5	2	13.6	13.5	13.5
5	4	11	5	4	5	9	13.7	13.4	13.6
6	9	14	11	7	7	14	13.5	13.5	13.6
7	11	4	9	5	6	6	13.5	13.5	13.5
8	7	10	11	8	14	5	13.6	13.6	13.6
9	5	8	7	13	16	13	13.6	13.5	13.4
10	19	11	12	25	17	16	13.5	13.5	13.5

* For description of the code, refer Table 3.1.

Table A3 Adult *Cryptolestes ferrugineus* count in test: M16.9O14.8V1357-6*.

After 1 and 3 d

Section	Insect count (%)						Moisture content		
	After 1 d			After 3 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	12	30	18	9	15	7	14.7	14.6	14.6
2	6	12	16	4	4	8	14.7	14.4	14.6
3	4	8	8	3	8	6	14.6	14.7	14.7
4	8	8	8	9	7	7	15.0	15.0	15.2
5	22	12	24	35	31	28	16.7	16.9	16.9
6	32	16	12	14	14	13	17.0	17.1	16.8
7	10	10	10	19	15	12	16.5	16.5	16.4
8	0	2	2	0	4	4	14.9	14.7	14.8
9	6	2	0	2	0	10	14.8	14.7	14.8
10	0	0	2	5	2	5	14.7	14.8	14.7

After 5 and 7 d

Section	Insect count (%)						Moisture content		
	After 5 d			After 7 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	25	18	20	19	21	15	14.8	14.9	14.9
2	12	13	12	13	12	13	14.9	14.9	14.8
3	7	8	11	5	6	6	14.8	14.8	14.8
4	14	8	8	9	7	5	14.9	15.0	14.9
5	12	12	9	14	13	14	16.9	16.8	17.1
6	10	15	15	14	14	16	17.2	17.2	17.4
7	12	15	14	15	14	17	17.0	16.9	17.1
8	3	5	6	6	5	7	14.9	15.1	15.0
9	4	4	3	5	5	5	14.8	14.9	14.8
10	1	2	2	0	3	2	14.9	14.8	14.8

* For description of the code, refer Table 3.1.

Table A4 Adult *Cryptolestes ferrugineus* count in test: U14.7V1357-6 (Control)*.

After 1 and 3 d

Section	Insect count (%)						Moisture content		
	After 1 d			After 3 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	47	71	36	28	57	32	14.8	14.7	14.7
2	20	13	14	19	11	13	14.5	14.8	14.7
3	10	5	16	14	7	9	14.8	14.8	14.7
4	4	2	11	10	5	8	14.7	14.8	14.8
5	3	2	6	2	2	7	14.8	14.9	14.7
6	5	1	3	8	6	3	14.5	14.9	14.9
7	2	1	4	1	3	7	14.7	14.9	14.6
8	3	2	4	4	4	11	14.6	14.8	14.8
9	3	1	5	3	4	5	14.5	14.8	14.8
10	3	2	1	11	1	5	14.7	14.7	14.9

After 5 and 7 d

Section	Insect count (%)						Moisture content		
	After 5 d			After 7 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	21	20	20	18	21	21	14.7	14.8	14.8
2	10	13	13	11	18	11	14.8	14.8	14.9
3	12	13	9	10	12	9	14.8	14.8	14.9
4	14	10	10	12	7	11	14.8	14.7	14.9
5	9	10	11	10	8	6	14.9	14.8	14.9
6	9	3	2	11	2	3	14.9	14.8	14.8
7	3	12	9	4	9	10	14.8	14.8	14.8
8	12	4	8	9	6	11	14.7	14.9	14.8
9	5	7	14	6	7	12	14.9	14.7	14.9
10	5	8	4	9	10	6	14.8	14.8	14.8

*For description of the code, refer Table 3.1.

Table A5 Adult *Cryptolestes ferrugineus* count in test: T16.6O14.8V1357-6*.

After 1 and 3 d

Section	Insect count (%)						Moisture content		
	After 1 d			After 3 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	22	19	30	10	20	17	14.7	14.8	14.8
2	13	29	17	10	11	10	14.6	14.8	14.8
3	12	16	10	7	10	7	14.5	14.9	14.8
4	3	4	10	2	6	5	14.6	14.7	14.7
5	6	2	1	4	2	3	14.7	14.8	14.9
6	5	5	1	5	2	2	14.6	14.8	14.8
7	9	1	5	7	3	5	14.8	15.0	14.8
8	11	8	12	22	17	15	16.6	16.7	16.7
9	13	9	8	23	10	18	16.6	16.6	16.6
10	5	6	5	10	19	18	16.5	16.6	16.6

After 5 and 7 d

Section	Insect count (%)						Moisture content		
	After 5 d			After 7 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	11	13	13	11	7	8	14.7	14.8	14.8
2	12	16	11	5	15	14	14.8	14.8	14.9
3	10	11	7	10	10	8	14.8	14.8	14.9
4	8	14	8	7	7	7	14.8	14.7	14.9
5	8	7	6	12	5	5	14.9	14.8	14.9
6	9	3	5	4	4	4	14.9	14.8	14.8
7	8	5	11	5	8	8	14.8	14.8	14.8
8	11	11	18	13	15	15	16.4	16.2	16.5
9	11	5	10	15	11	13	16.5	16.6	16.5
10	12	15	11	18	18	18	16.7	16.6	16.6

* For description of the code, refer Table 3.1.

Table A6 Adult *Cryptolestes ferrugineus* count in test: M16.5O12V15-t*.

Section	Insect count (%)						Moisture content		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	0	1	1	3	3	4	11.9	12.0	12.0
2	2	1	1	2	0	2	12.0	12.1	12.0
3	1	1	1	5	1	1	11.9	11.9	12.2
4	2	6	9	4	1	3	12.3	12.2	12.2
5	36	40	35	24	26	26	16.4	16.3	16.4
6	43	38	42	31	35	33	16.6	16.7	16.7
7	10	9	7	27	32	28	16.5	16.4	16.5
8	6	2	2	2	1	1	12.3	12.3	12.2
9	0	1	2	1	1	1	12.1	12.0	11.9
10	0	1	0	0	0	0	12.0	12.1	12.0

Table A7 Adult *Cryptolestes ferrugineus* count in test: U12V15-t (Control)*.

Section	Insect count (%)						Moisture content		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	20	18	20	25	24	26	12.1	12.0	12.2
2	17	17	18	17	18	19	11.9	11.9	12.1
3	13	11	10	13	15	8	12.2	12.0	12.2
4	6	8	6	13	7	8	12.3	12.2	12.3
5	6	8	5	4	4	5	12.4	12.1	12.2
6	4	2	6	5	5	8	12.3	11.9	12.1
7	3	6	5	4	4	4	12.1	12.3	12.2
8	10	7	7	4	6	7	12.2	12.2	12.2
9	10	11	9	7	9	7	12.0	12.1	11.9
10	11	12	13	8	8	8	12.1	12.0	12.0

* For description of the code, refer Table 3.1.

Table A8 Adult *Cryptolestes ferrugineus* count in test: T18.2O14.8V15-t*.

Section	Insect count (%)						Moisture content		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	10	5	2	27	18	13	14.7	14.7	14.7
2	5	4	3	8	9	9	15.0	14.8	14.7
3	2	3	3	7	4	13	15.2	15.0	14.6
4	3	3	5	5	2	6	15.1	14.7	14.6
5	1	2	2	5	5	4	15.1	14.8	14.9
6	1	3	2	2	6	2	15.3	14.7	14.6
7	4	5	2	9	5	2	15.1	15.4	14.8
8	12	19	27	10	14	14	18.2	17.8	17.9
9	36	25	31	10	20	19	18.3	18.3	18.3
10	26	31	23	17	17	18	18.3	18.3	18.1

Table A9 Adult *Cryptolestes ferrugineus* count in test: U14.7V15-t (Control)*.

Section	Insect count (%)						Moisture content		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	25	19	27	28	26	29	14.8	14.7	14.7
2	15	18	18	14	24	22	14.8	14.8	14.7
3	8	12	11	16	16	15	14.6	14.7	14.6
4	5	10	9	7	9	8	14.6	14.5	14.6
5	7	5	4	4	4	4	14.7	14.6	14.8
6	7	3	5	4	6	6	14.7	14.7	14.8
7	5	6	4	5	4	2	14.8	14.8	14.8
8	6	6	4	5	3	3	14.9	14.9	14.7
9	8	10	7	8	3	6	14.8	14.8	14.6
10	14	11	11	9	5	5	14.8	14.8	14.8

* For description of the code, refer Table 3.1.

Table A10 Coefficients, F-statistics and R^2 in regression models fitted to the data on movement of adult *Cryptolestes ferrugineus* in pockets of high moisture content wheat in grain columns.

Parameters	Coefficients		
	RegGMC	RegGMCq1	RegGMCq
Distance	0.003	0.004	0.005
Time	0.026	0.025	0.025
m.c.	0.176	0.167	0.354
Intercept	-1.887	-1.722	-3.121
$Distance^2$	****	-0.00003	-0.00003
$m.c.^2$	****	****	-0.006
F-statistics ($P<0.01$)	87.85	69.37	55.83
R^2	0.62	0.64	0.64

Table A11 Standard error of *Cryptolestes ferrugineus* adult count for the tests on insect movement in pockets of high moisture content wheat in grain columns.

Test	Standard error		
	RegGMC*	RegGMCq1	RegGMCq
M175O13.6H1357-6	6.71	6.74	6.67
M16.9O14.8V1357-6	5.34	5.36	5.3
T16.6O14.8V1357-6	4.95	5.29	5.3
M16.5O12V15-t	9.94	10.24	10.49
T18.2O14.8V15-t	8.03	8.46	8.2

* RegGMC denotes a regression model fitted to the data on movement of adult *Cryptolestes ferrugineus* in grain columns containing pockets of high moisture content wheat.

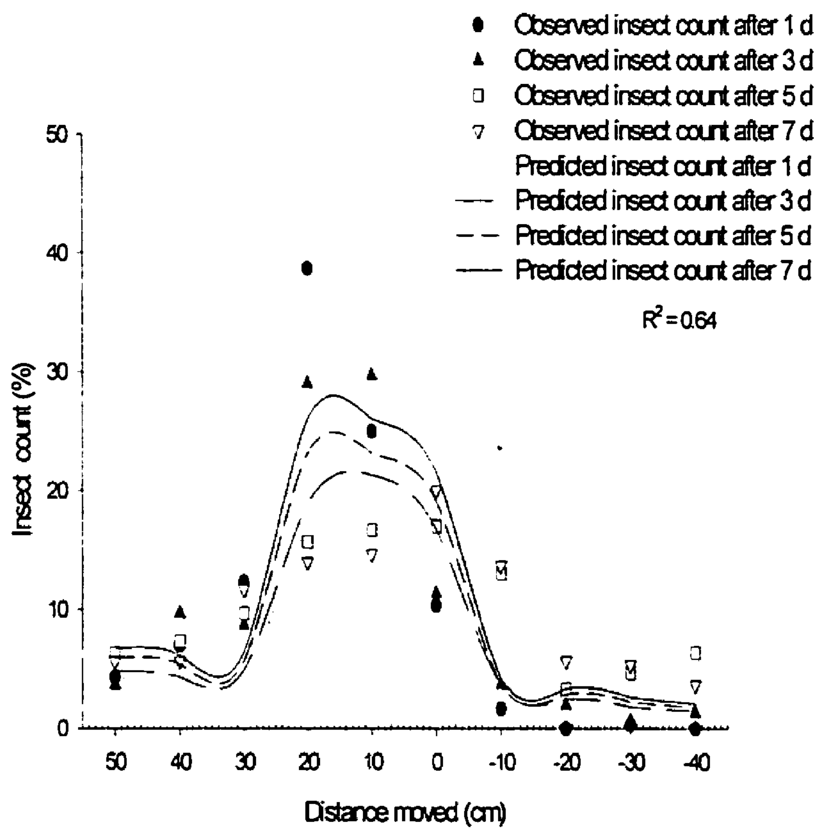


Fig. A1 Observed and predicted movement of adult *Cryptolestes ferrugineus* in pockets of high moisture content wheat.in test M17.5O13.6H1357-6. Insects introduced at 0 cm distance.

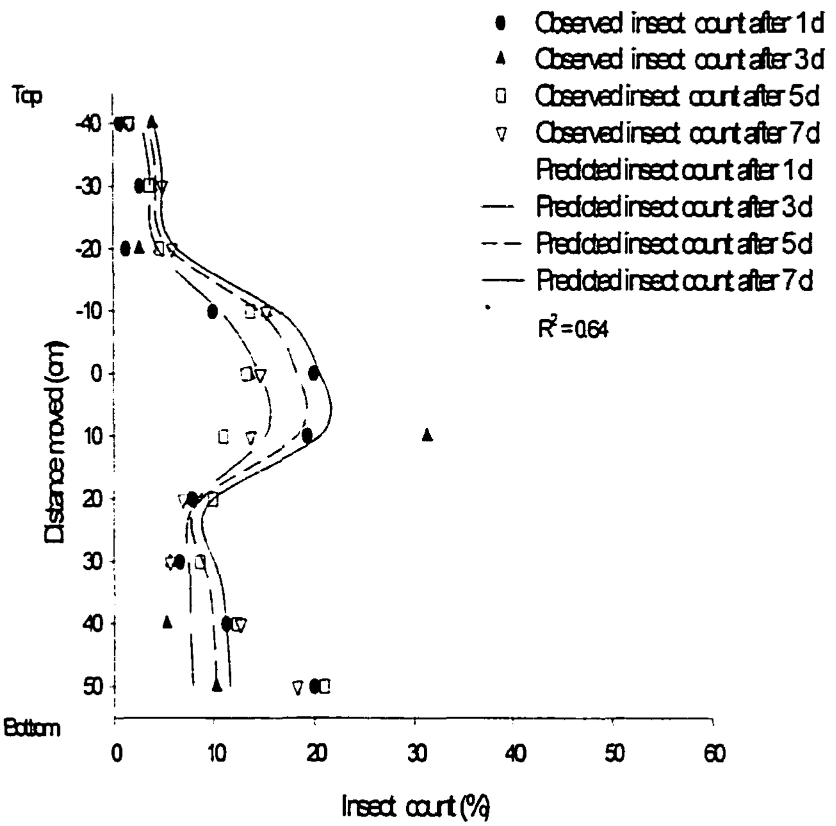


Fig. A2 Observed and predicted movement of adult *Cryptolestes ferrugineus* in pockets of high moisture content wheat in test M16.9O14.8V1357-6. Insects introduced at 0 cm distance.

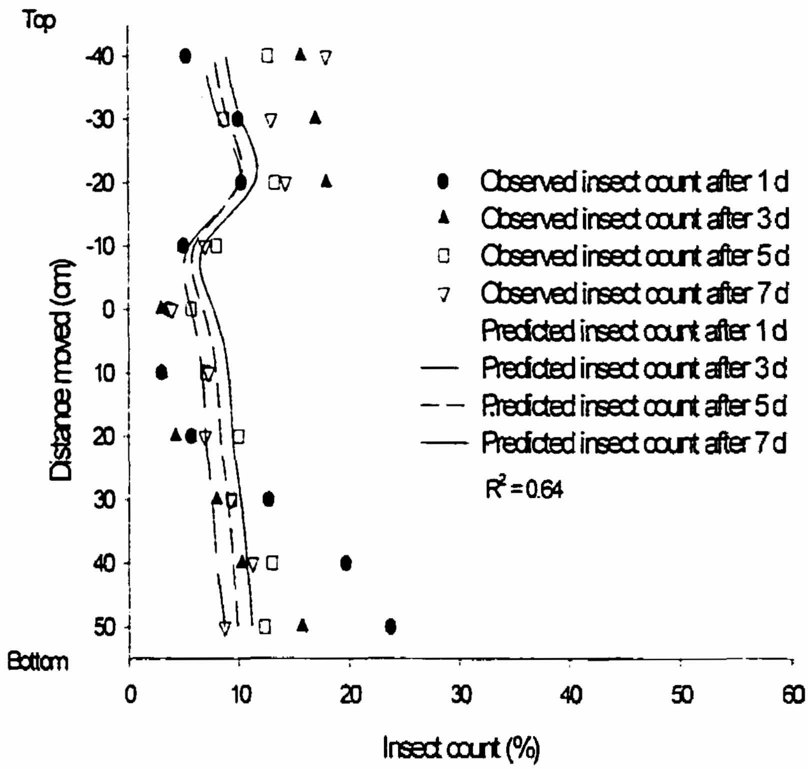


Fig. A3 Observed and predicted movement of adult *Cryptolestes ferrugineus* in pockets of high moisture content wheat in test T16.6O14.8V1357-6. Insects introduced at 0 cm distance.

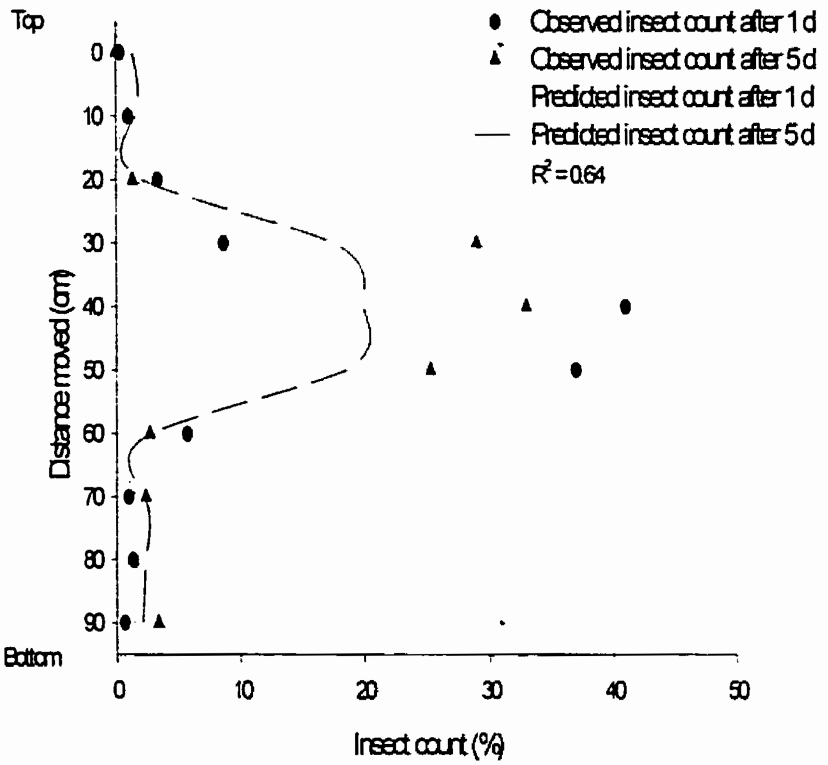


Fig. A4 Observed and predicted movement of adult *Cryptolestes ferrugineus* in pockets of high moisture content wheat in test M16.5O12V15-t. Insects introduced at 0 cm distance.

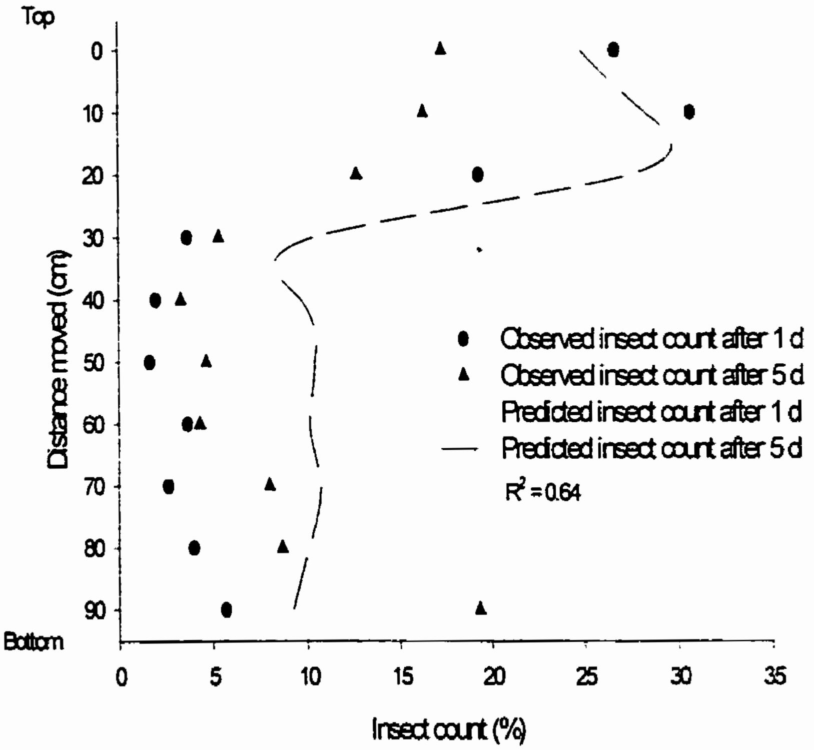


Fig. A5 Observed and predicted movement of adult *Cryptolestes ferrugineus* in pockets of high moisture content wheat in test T18.2O14.8V15-t. Insects introduced at 0 cm distance.

Appendix B

Table B1 Adult *Cryptolestes ferrugineus* count in test: U12H15-6 (under CO_2 gradients)*.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 5 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	20	27	28	22	24	23	12.2	12.2	12.2	7.8	7.7	7.7
2	24	29	26	24	14	20	12.1	12.1	12.1	7.0	6.7	6.8
3	16	27	22	18	14	13	12.1	12	12.1	6.4	6.1	6.1
4	20	7	5	13	20	13	12	12	12	5.9	5.6	5.6
5	5	4	9	7	10	10	12.1	12.1	12	5.3	5.1	5.1
6	4	5	4	5	6	6	11.9	12.3	12	4.7	4.4	4.4
7	4	1	2	1	1	1	12	12.1	11.9	3.7	3.5	3.5
8	3	0	2	2	4	6	12.1	12.2	12	3.1	3.1	3.0
9	2	0	0	3	3	3	12.2	12	12.1	2.5	2.4	2.4
10	2	0	1	5	4	5	12.1	12	12.2	1.8	1.7	1.6

Table B2 Adult *Cryptolestes ferrugineus* count in test: U12H15-6 (Control)*.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	4	4	0	18	13	14	12.1	12.2	12.2
2	4	1	4	11	13	10	12.2	12.2	12.2
3	7	8	1	11	17	12	12.1	12.1	12.1
4	1	4	10	10	13	11	12.1	12.1	12.1
5	7	3	21	7	9	14	11.9	12	12
6	17	18	17	5	13	8	11.9	12.1	12.1
7	27	18	23	12	5	5	12	12	12
8	17	22	3	13	4	8	12.1	11.9	11.9
9	12	18	8	9	10	7	12.1	12.1	12.1
10	4	6	13	4	3	11	12.2	12.1	12.1

* For description of the code, refer Table 3.2.

Table B3 Adult *Cryptolestes ferrugineus* count in test: U14.6V1735-6 (under CO_2 gradients)*.

After 1 and 7 d.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 7 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	35	28	37	40	32	19	14.6	14.5	14.6	7.8	7.7	7.7
2	13	16	27	16	18	14	14.6	14.6	14.5	7.0	6.7	6.8
3	14	13	7	10	6	11	14.5	14.5	14.4	6.4	6.1	6.1
4	10	12	3	10	7	9	14.3	14.6	14.4	5.9	5.6	5.6
5	10	13	9	7	8	11	14.4	14.3	14.4	5.3	5.1	5.1
6	6	5	7	5	6	10	14.6	14.6	14.6	4.7	4.4	4.4
7	3	5	5	4	3	7	14.6	14.6	14.4	3.7	3.5	3.5
8	2	1	4	2	5	4	14.4	14.5	14.3	3.1	3.1	3.0
9	2	4	0	2	7	6	14.4	14.5	14.4	2.5	2.4	2.4
10	5	3	2	4	8	9	14.6	14.7	14.6	1.8	1.7	1.6

After 3 and 5 d.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 3 d			After 5 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	25	25	20	38	34	32	14.3	14.5	14.6	7.6	7.8	7.7
2	13	16	14	18	19	13	14.6	14.5	14.6	7.1	6.8	6.8
3	14	13	13	17	10	14	14.5	14.5	14.3	6.3	6.2	6.1
4	16	12	10	13	7	8	14.5	14.4	14.3	5.8	5.6	5.7
5	7	13	11	3	8	8	14.4	14.6	14.4	5.3	5.1	5.1
6	9	5	7	3	6	4	14.6	14.6	14.7	4.4	4.4	4.7
7	3	5	9	2	3	7	14.4	14.6	14.6	3.7	3.5	3.5
8	4	3	8	3	5	3	14.5	14.6	14.5	3.2	3.1	3.1
9	3	4	4	1	3	6	14.4	14.4	14.7	2.7	2.4	2.2
10	5	6	4	2	5	5	14.4	14.6	14.6	1.5	1.8	1.8

* For description of the code, refer Table 3.2.

Table B4 Adult *Cryptolestes ferrugineus* count in test: U14.6V1735-6 (Control)*.

After 1 and 7 d.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 7 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	32	31	25	15	15	9	14.5	14.6	14.6
2	8	19	14	9	17	10	14.6	14.6	14.6
3	10	9	11	10	9	6	14.5	14.7	14.7
4	5	12	9	15	12	11	14.6	14.6	14.6
5	11	10	12	2	5	11	14.5	14.5	14.5
6	11	7	8	9	14	13	14.6	14.6	14.6
7	6	1	6	8	5	11	14.7	14.7	14.7
8	3	0	8	13	9	10	14.5	14.6	14.6
9	8	3	5	12	8	10	14.6	14.6	14.6
10	6	7	2	7	5	9	14.6	14.6	14.6

After 3 and 5 d.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	14	26	13	15	13	11	14.5	14.5	14.5
2	27	25	9	19	10	15	14.6	14.6	14.6
3	5	18	8	8	11	13	14.5	14.5	14.5
4	6	5	18	9	6	8	14.6	14.6	14.6
5	14	12	18	8	9	7	14.5	14.5	14.5
6	6	4	9	4	11	14	14.6	14.6	14.6
7	10	3	7	9	6	8	14.7	14.7	14.7
8	6	4	4	11	10	12	14.5	14.5	14.5
9	2	2	7	9	12	7	14.6	14.6	14.6
10	9	0	7	8	12	5	14.6	14.6	14.6

* For description of the code, refer Table 3.2.

Table B5 Adult *Cryptolestes ferrugineus* count in test: U12InV15-6 (under CO_2 gradients)*.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 5 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	17	17	16	17	14	10	12.2	12.2	12.2	8.990	8.139	8.057
2	11	10	16	10	14	17	12.1	12.1	12.1	8.571	7.012	7.190
3	21	17	14	15	12	14	12.1	12	12.1	8.263	6.244	6.311
4	8	9	14	4	12	5	12	12	12	7.990	5.742	5.720
5	14	5	14	12	7	8	12.1	12.1	12	7.722	5.314	5.292
6	5	5	6	11	8	6	11.9	12.3	12	6.941	4.736	4.709
7	14	9	1	11	6	3	12	12.1	11.9	6.032	3.544	3.535
8	2	9	9	6	4	15	12.1	12.2	12	4.318	3.133	3.108
9	5	6	2	5	14	6	12.2	12	12.1	2.680	2.473	2.498
10	3	12	7	9	9	15	12.1	12	12.2	1.873	1.748	1.731

Table B6 Adult *Cryptolestes ferrugineus* count in test: U12InV15-6 (Control)*.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	3	7	6	3	4	7	12.1	12.1	12.2
2	9	5	6	10	5	5	12.2	12	12.1
3	4	4	6	3	8	6	12.1	12.2	12
4	1	11	3	9	9	3	12.1	12.1	12
5	4	3	6	7	7	5	12	12	12.2
6	10	9	19	4	8	7	12.2	11.9	12.1
7	10	15	12	7	12	13	12.1	12	12.1
8	16	14	22	13	13	18	12.2	12.1	12
9	18	19	10	20	14	16	12.1	12	12.1
10	24	13	9	24	20	20	12	12	11.9

* For description of the code, refer Table 3.2.

Table B7 Adult *Cryptolestes ferrugineus* count in test: U12V15-t (under CO_2 gradients)*.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 5 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	26	18	19	28	28	20	12.2	12.1	12.2	7.8	7.7	7.7
2	14	12	9	18	15	16	12.1	12.1	12.1	7.0	6.7	6.8
3	6	6	22	4	10	18	12.2	12	12.1	6.4	6.1	6.1
4	14	25	8	12	8	14	12	12	12	5.9	5.6	5.6
5	8	6	6	9	8	6	12.2	12.1	12	5.3	5.1	5.1
6	5	5	8	8	4	9	11.9	12.3	12	4.7	4.4	4.4
7	12	6	7	10	7	8	12.1	12.1	11.9	3.7	3.5	3.5
8	5	3	8	5	8	3	12.1	12.2	12	3.1	3.1	3.0
9	4	8	7	2	6	4	12.2	12	12.1	2.5	2.4	2.4
10	6	11	6	4	6	2	12.1	12	12.2	1.8	1.7	1.6

Table B8 Adult *Cryptolestes ferrugineus* count in test: U12V15-t (Control)*.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	15	13	13	25	21	19	12.1	12.1	12.2
2	14	10	12	14	13	17	12.2	12	12.1
3	3	8	9	6	15	12	12.1	12.2	12
4	8	6	8	10	6	8	12.1	12.1	12
5	10	9	9	6	9	12	12	12	12.2
6	10	7	7	4	8	4	12.2	11.9	12.1
7	11	13	12	19	10	9	12.1	12	12.1
8	7	13	9	5	9	8	12.2	12.1	12
9	11	13	14	5	3	7	12.1	12	12.1
10	8	7	7	6	5	4	12	12	11.9

* For description of the code, refer Table 3.2.

Table B9 Coefficients, F-statistics and R^2 in regression models fitted to the data on movement of adult *Cryptolestes ferrugineus* under CO_2 gradients in grain columns.

Parameters	Coefficients			
	$RegCO_25$*	$RegCO_23$	$RegCO_24$	$RegCO_2$
Distance	0.002	0.002	0.002	0.034
Time	0.012	0.012	0.012	0.063
CO_2	***	0.097	-0.044	-9.60
Intercept	0.538	0.352	0.635	17.80
CO_2^2	****	****	0.015	1.937
CO_2^3	****	****	****	-0.087
F-statistics (at P<0.01)	61.75	56.17	46.24	48.04
R^2	0.65	0.63	0.66	0.72

Table B10 Standard error of adult *Cryptolestes ferrugineus* count and relative percent error for the tests on insect movement under CO_2 gradients in grain columns.

Test	$RegCO_25$		$RegCO_23$		$RegCO_24$		$RegCO_2$	
	SE	e (%)	SE	e (%)	SE	e (%)	SE	e (%)
U12H15-6	5.1	83	5.4	84.6	5.3	82.9	4.4	84.3
U14.6V1735-6	3.8	21.4	4.5	24.4	3.7	20.8	3.9	20.7
U12InV15-6	5.8	37.5	4.3	32.3	6.5	38.9	5.7	41.5
U12V15-t	2.7	21.4	2.8	23.8	3.1	22.3	3.0	24.9

* $RegCO_25$ denotes a regression model examined for the movement of adult *Cryptolestes ferrugineus* under CO_2 gradients in grain columns.

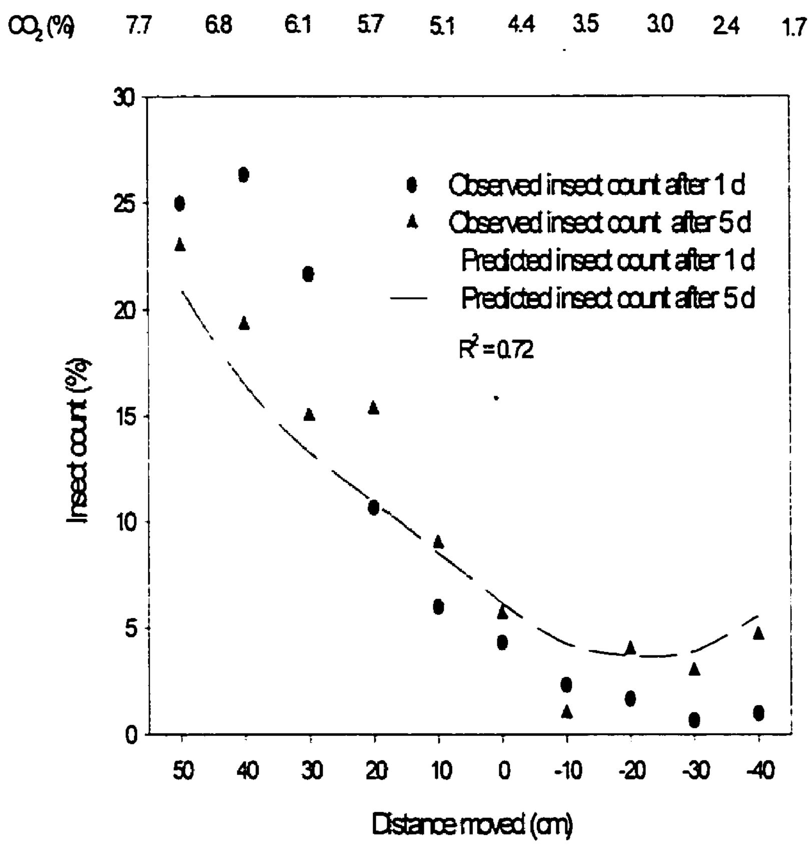
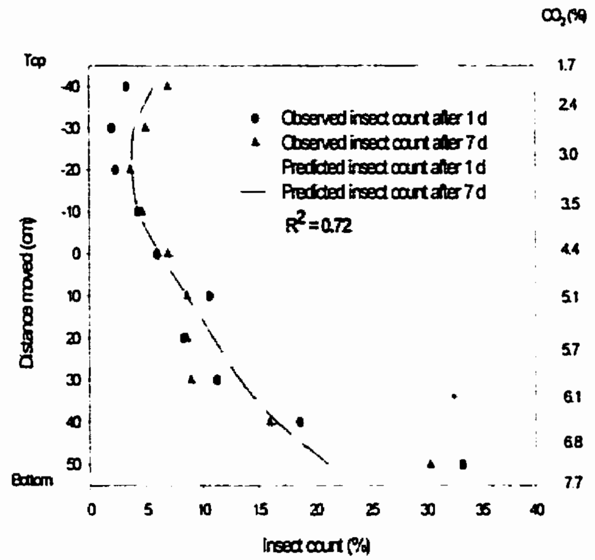


Fig. B1 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients in test U12H15-6. Insects introduced at 0 cm distance.



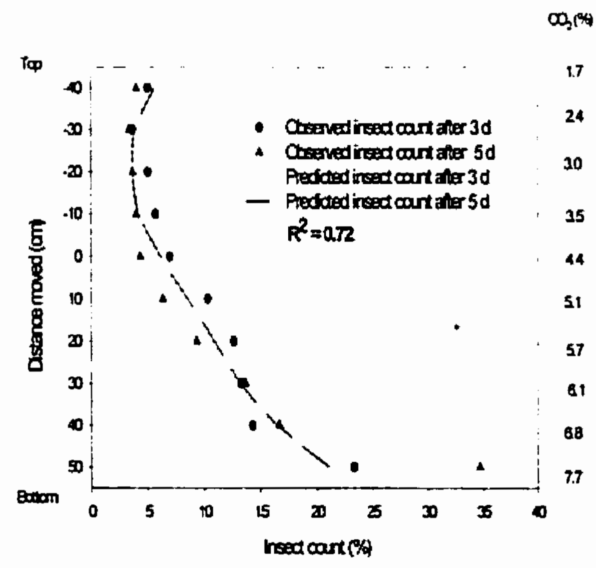


Fig. B2 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients in test U14.6V1735-6. Insects introduced at 0 cm distance.

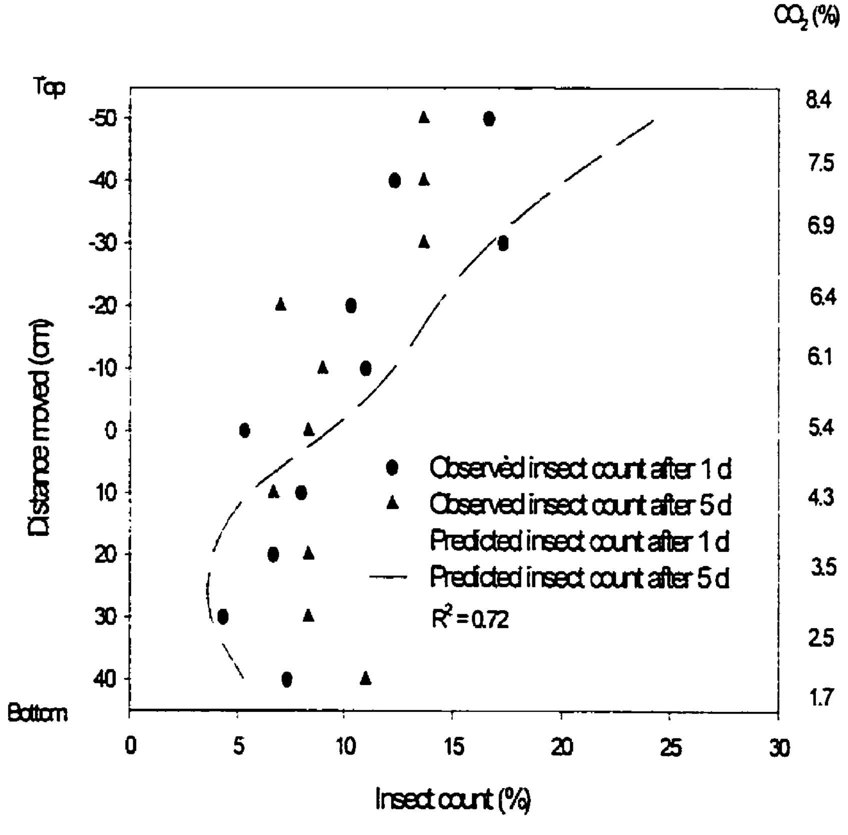


Fig. B3 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients in test U12InV15-6. Insects introduced at 0 cm distance.

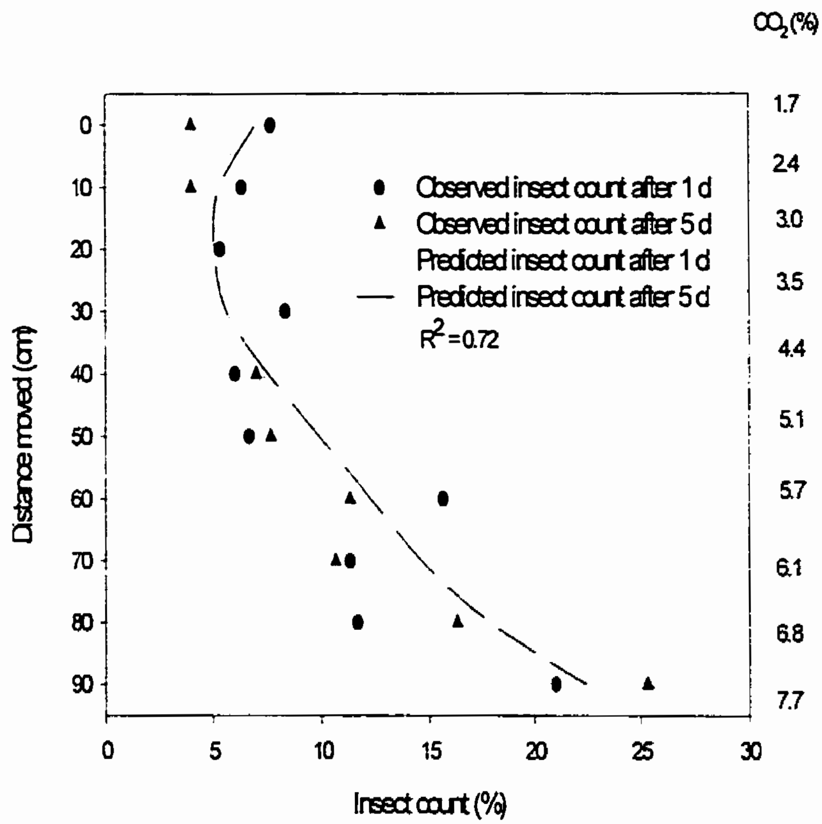


Fig. B4 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients in test U12V15-t. Insects introduced at 0 cm distance.

Appendix C

Table C1 Adult *Cryptolestes ferrugineus* count in test: OE16.6O14.4H15-6 (under CO_2 gradients)*.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 5 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	17	17	14	14	18	14	14.2	14.3	14.3	8.4	8.5	7.2
2	20	14	32	20	21	21	14.3	14.3	14.2	8.0	8.1	6.9
3	16	33	15	11	19	12	14.4	14.2	14.4	7.8	7.9	6.3
4	20	26	15	14	12	11	14.3	14.4	14.3	7.5	7.7	6.0
5	16	3	17	6	3	7	14.3	14.4	14.2	6.9	7.3	5.4
6	4	4	0	5	1	4	14.4	14.3	14.3	6.4	7.0	5.0
7	0	0	2	0	7	5	14.7	14.6	14.7	5.8	6.8	4.6
8	2	1	2	7	5	6	16.2	16.6	16.7	5.3	6.2	4.1
9	5	1	3	16	5	8	16.8	16.6	16.4	4.8	5.6	3.9
10	0	1	0	7	9	12	16.8	16.7	16.9	4.0	5.0	3.2

Table C2 Adult *Cryptolestes ferrugineus* count in test: OE16.7O14.3H15-6 (Control)*.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	0	0	4	9	0	9	14.3	14.3	14.3
2	0	2	4	2	0	8	14.4	14.4	14.2
3	4	3	0	4	4	3	14.2	14.3	14.4
4	5	5	0	2	4	1	14.3	14.3	14.3
5	12	10	2	6	2	3	14.2	14.4	14.4
6	10	5	14	3	5	3	14.2	14.5	14.3
7	10	10	7	9	12	7	14.0	14.4	14.5
8	15	17	18	8	19	16	16.6	16.7	16.3
9	28	40	30	32	43	22	16.7	16.7	16.7
10	16	8	21	25	11	28	16.8	16.8	16.8

* For description of the code, refer Table 3.3.

Table C3 Adult *Cryptolestes ferrugineus* count in test: T16O14.1V17-6 (under CO_2 gradients)*.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 7 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	37	41	44	15	15	19	14.1	13.9	14.1	8.210	8.100	8.057
2	14	21	18	9	5	7	14.2	13.8	14	7.890	7.664	7.880
3	17	13	12	8	12	8	14.1	14	13.8	7.460	7.290	7.530
4	7	7	9	4	9	8	14	14.1	13.9	7.128	7.030	7.378
5	4	4	2	4	5	5	14	14.2	14.1	6.788	6.810	6.940
6	2	4	3	5	7	6	13.9	14	14.2	6.409	6.510	6.542
7	4	1	2	4	2	5	14.6	14.7	14.7	6.181	5.948	6.220
8	3	4	3	13	10	13	16.2	15.8	15.8	5.690	5.607	5.948
9	3	3	2	22	28	17	16.1	16.1	16	5.380	5.225	5.648
10	9	2	5	16	7	10	15.9	16.2	16.1	5.180	4.825	4.960

Table C4 Adult *Cryptolestes ferrugineus* count in test: T15.9O14.1V17-6 (Control)*.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 7 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	35	24	35	15	11	14	13.9	14.2	14
2	17	6	13	13	4	8	14.1	14.1	14
3	14	16	10	1	5	6	14.2	14.1	13.8
4	11	16	10	4	4	2	14.1	14.1	14.1
5	5	10	6	2	3	2	13.9	14.2	14.2
6	1	6	3	1	3	7	14.2	14.3	14
7	0	4	5	4	8	6	14.7	14.8	14.8
8	6	0	8	20	11	20	15.9	15.9	15.7
9	8	15	5	16	27	20	16.1	15.8	15.9
10	3	3	5	24	24	15	16.2	16	16.1

* For description of the code, refer Table 3.3.

Table C5 Adult *Cryptolestes ferrugineus* count in test: B16O13.9InV15-6 (under CO_2 gradients)*.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 5 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	11	9	8	9	8	9	13.7	13.8	13.9	8.440	7.83	8.31
2	10	7	7	4	4	5	13.8	13.9	13.7	7.790	7.59	7.71
3	6	5	3	5	6	4	13.9	14	13.8	7.380	7.24	7.33
4	2	2	2	1	5	5	13.7	13.9	13.7	7.180	6.89	6.87
5	3	5	8	2	6	6	13.8	13.8	13.9	6.480	6.51	6.54
6	2	7	5	3	3	6	14	14.1	14.1	6.020	6.12	6.27
7	5	5	8	5	8	5	14.1	14.2	14.2	5.790	5.74	5.78
8	12	23	10	10	9	12	15.9	15.9	16	5.310	5.4	5.47
9	27	23	24	28	20	25	16	15.9	16.1	4.820	5.09	5.14
10	22	14	25	33	31	23	16.3	16	16.2	4.010	4.52	4.46

Table C6 Adult *Cryptolestes ferrugineus* count in test: B15.9O13.9InV15-6 (Control*).

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	1	1	1	3	1	2	13.8	13.9	13.8
2	0	2	0	3	1	1	13.8	13.7	13.7
3	1	1	0	4	0	3	13.9	14.1	13.8
4	0	1	0	4	2	2	13.7	13.9	13.7
5	0	0	2	1	1	0	13.8	13.8	13.9
6	5	2	2	2	1	9	14.1	14.1	14.2
7	5	3	0	6	7	7	14.1	14.2	14.1
8	10	7	11	15	18	17	15.8	15.8	15.9
9	23	24	30	35	35	28	16	15.9	16.2
10	55	59	54	27	34	31	16.1	16.1	16.1

* For description of the code, refer Table 3.3.

Table C7 Adult *Cryptolestes ferrugineus* count in test: T17O12.8InV15-6 (under CO_2 gradients)*.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 5 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	15	22	18	30	20	29	17.1	17.2	16.8	8.210	7.937	8.057
2	34	18	40	27	30	27	17.2	17.3	17.2	7.860	7.664	7.880
3	18	19	20	23	29	26	17.2	17.1	17	7.450	7.290	7.616
4	7	8	5	11	10	7	13.8	13.5	12.4	7.220	7.010	7.378
5	1	7	2	3	4	4	12.8	12.9	12.9	6.620	6.810	6.993
6	4	1	2	2	2	2	12.6	12.8	12.7	6.280	6.426	6.542
7	6	10	3	2	1	1	12.5	12.6	12.8	6.030	5.948	6.180
8	10	6	6	1	1	2	12.7	12.7	12.6	5.710	5.607	5.948
9	4	5	3	1	1	1	12.5	12.5	12.8	5.320	5.225	5.648
10	1	4	1	0	2	1	12.7	12.4	12.7	5.010	4.825	4.960

Table C8 Adult *Crytolestes ferrugineus* count in test: T17.2O12.8InV15-6 (Control)*.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	9	4	8	19	14	16	17.3	17.2	17.2
2	13	11	11	22	17	33	17.1	17.1	17.3
3	21	16	21	27	38	14	17.3	17.2	17.4
4	7	13	6	15	13	15	13.6	13.6	13.8
5	6	4	3	1	2	3	12.8	12.9	12.8
6	6	3	8	1	2	2	12.9	12.8	12.9
7	7	15	5	3	3	1	12.8	12.6	12.8
8	6	15	10	1	1	1	12.7	12.6	12.7
9	6	16	9	1	8	1	12.7	12.7	12.6
10	19	3	19	10	2	14	12.6	12.8	12.7

* For description of the code, refer Table 3.3.

Table C9 Adult *Cryptolestes ferrugineus* count in test: M15.5O12.1V15-t (under CO_2 gradients)*.

Section	Insect count (%)						Moisture content (%)			CO_2(%)		
	After 1 d			After 5 d								
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	3	8	3	5	2	6	12.2	12.2	12.1	8.265	7.937	8.057
2	2	6	1	3	2	3	12.1	12.3	12.1	7.964	7.664	7.880
3	3	3	0	1	3	2	12.1	12	12.1	7.526	7.290	7.616
4	6	6	0	6	6	4	12.4	12.3	12.5	7.128	7.010	7.378
5	31	32	39	32	31	26	15.4	15.5	15.6	6.788	6.810	6.993
6	30	27	28	28	34	40	15.5	15.4	15.7	6.409	6.426	6.542
7	19	12	21	22	17	17	15.5	15.7	15.8	6.181	5.948	6.180
8	3	3	7	0	4	2	12.3	12.4	12.4	5.768	5.607	5.948
9	2	2	0	1	1	0	12.2	12	12.1	5.414	5.225	5.648
10	1	1	1	2	0	0	12.1	12	12.2	5.236	4.825	4.960

Table C10 Adult *Cryptolestes ferrugineus* count in test: M15.5O12.1V15-t (Control)*.

Section	Insect count (%)						Moisture content (%)		
	After 1 d			After 5 d					
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
1	0	0	0	3	1	2	12.1	12.1	12.2
2	0	0	0	2	2	2	12.2	12	12.1
3	0	0	0	2	0	0	12.1	12.2	12.3
4	0	2	2	6	7	2	12.4	12.4	12.6
5	19	19	11	29	27	31	15.6	15.5	15.4
6	16	23	34	26	28	29	16	15.6	15.5
7	19	30	14	12	30	27	15.4	15.8	15.6
8	29	25	22	11	5	6	12.5	12.5	12.3
9	8	0	9	6	0	0	12.1	12.1	12.1
10	9	1	8	3	0	1	12	12	11.9

* For description of the code, refer Table 3.3.

Table C11 Coefficients, F-statistics and R^2 of regression models fitted to the data on the movement of adult *Cryptolestes ferrugineus* under CO_2 gradients and in pockets of high moisture content wheat in grain columns.

Parameters	Coefficients			
	RegMCCO$_2$-5a*	RegMCCO$_2$-4a	RegMCCO$_2$-2a	RegMCCO$_2$-3a
Distance	0.001	0.004	0.004	0.003
Time	0.005	0.005	0.005	0.005
m.c.	-13.530	1.540	1.486	1.591
CO_2	0.178	-0.078	0.070	0.08
Intercept	59.781	-11.80	-11.860	-12.69
Distance^2	****	****	****	0.00001
m.c.^{2}	0.988	-0.046	-0.044	-0.047
$CO_2{}^2$	****	0.011	****	****
m.c.^{3}	-0.023	****	****	****
F-statistics (at P<0.01)	19.35	18.77	22.7	18.87
R^2	0.56	0.54	0.54	0.55

*RegMCCO$_2$ denotes a regression model with both high moisture content (MC) and CO_2 being present in the columns for studying movement of adult *Cryptolestes ferrugineus*.

Table C12 Standard error of adult *Cryptolestes ferrugineus* count and relative percent error for the tests on insect movement under CO_2 gradients and in pockets of high moisture content wheat in grain columns.

Test	RegMCCO$_2$-5a		RegMCCO$_2$-4a		RegMCCO$_2$-2a		RegMCCO$_2$-3a	
	SE	e (%)	SE	e (%)	SE	e (%)	SE	e (%)
OE16.6O14.4H15-6	6.8	164.5	6.9	190.3	6.7	184.7	7.0	187.3
T16O14.1V17-6	9.7	70.9	9.6	61.5	9.4	63.1	9.6	63.6
B16O13.9InV15-6	10.4	49.8	10.7	57.0	10.4	56.5	10.6	55.2
T17O12.8InV15-6	5.4	32.3	4.7	31.2	4.3	30.8	4.8	32.0
M15.5O12.1V15-t	8.0	41.8	8.4	36.0	7.7	34.7	8.3	38.3

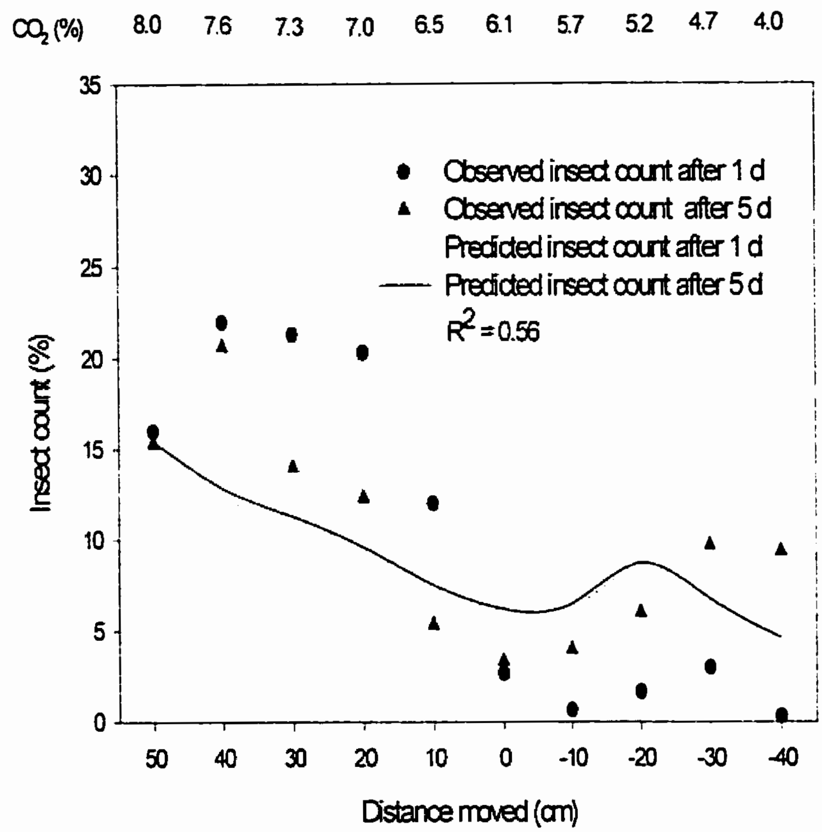


Fig. C1 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients and in pockets of high moisture content wheat in test:OE16.6O14.4H15-6. Insects were introduced at 0 cm distance.

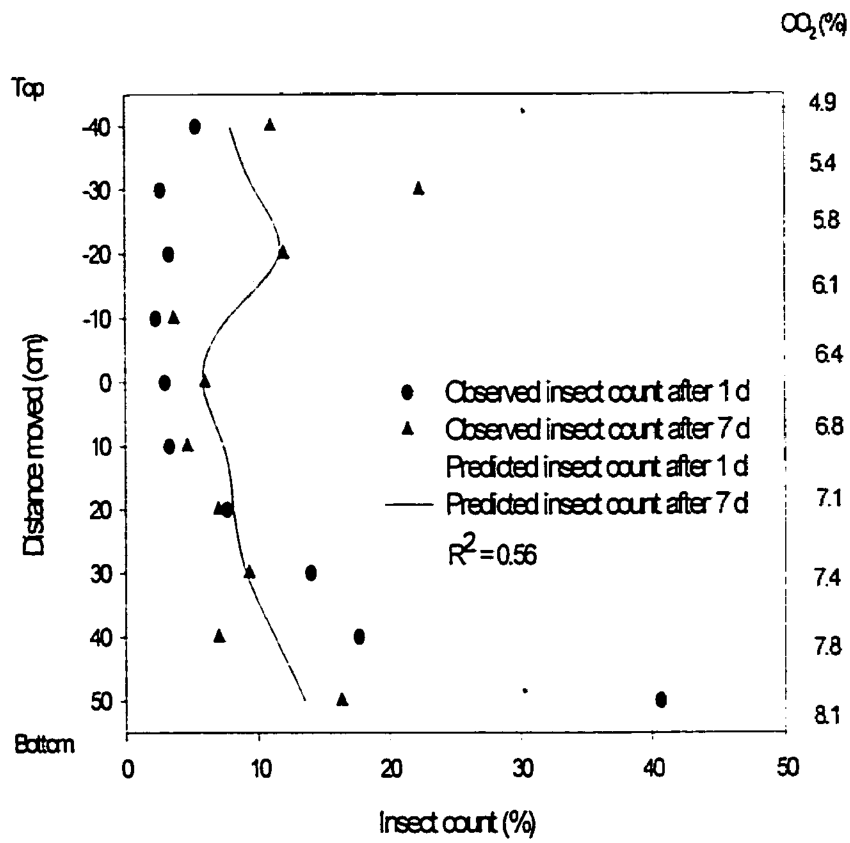


Fig. C2 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients and in pockets of high moisture content wheat in test: T16O14.1V17-6. Insects were introduced at 0 cm distance.

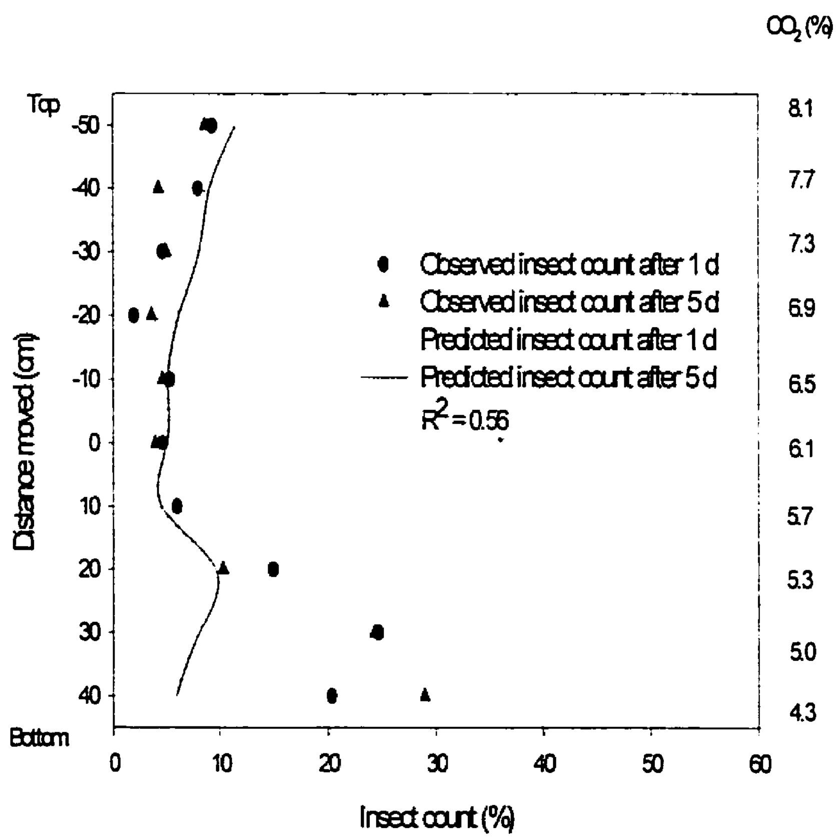


Fig. C3 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients and in pockets of high moisture content wheat in test: B16O13.9InV15-6. Insects introduced at 0 cm distance.

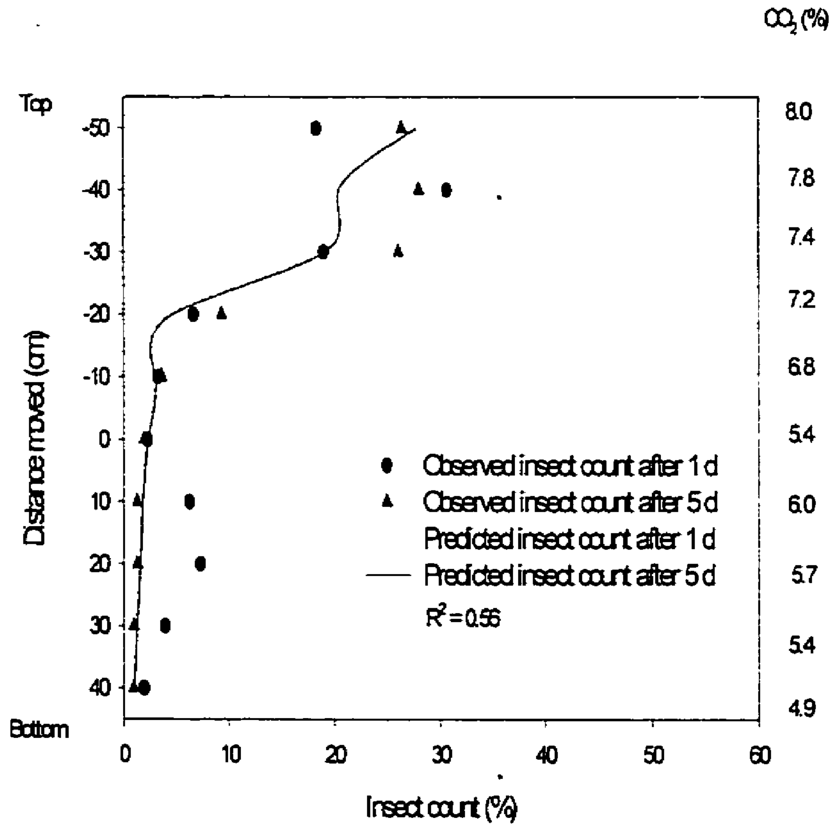


Fig. C4 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients and in pockets of high moisture content wheat in test: T17O12.8InV15-6. Insects introduced at 0 cm distance.

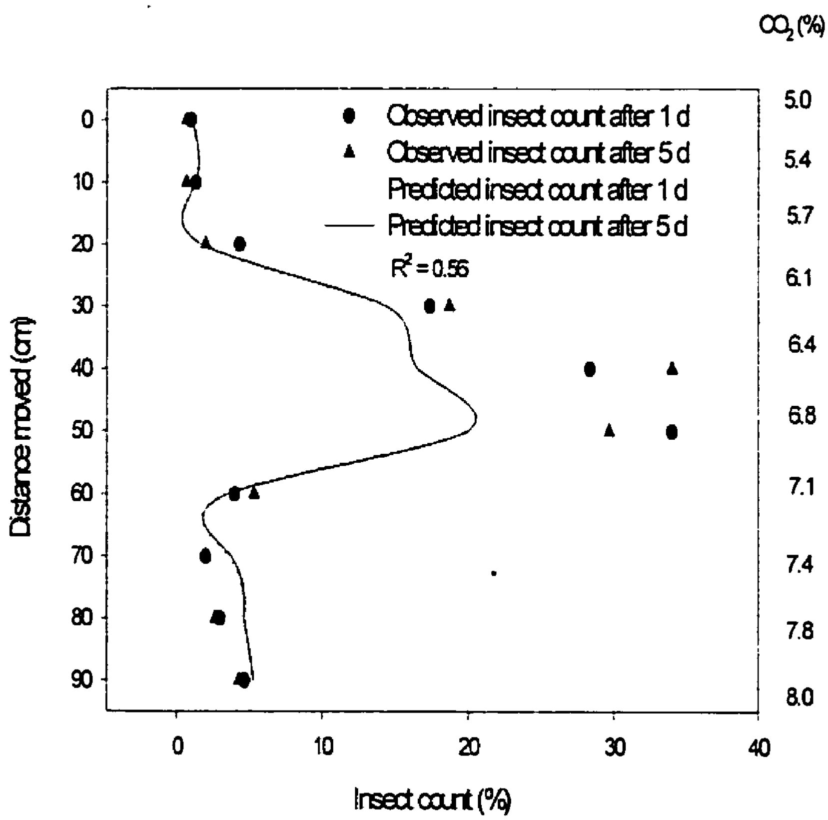


Fig. C5 Observed and predicted movement of adult *Cryptolestes ferrugineus* under CO_2 gradients and in pockets of high moisture content wheat in test: M15.5O12.1V15-t. Insects introduced at 0 cm distance.