

A QUANTITATIVE MODEL FOR THE ADHESIVE LOCOMOTION OF THE TERRESTRIAL SLUG, *ARIOLIMAX COLUMBIANUS*

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SUMMARY

The foot of pulmonate gastropods adheres to the surface upon which the animal crawls, the adhesive being a thin layer of pedal mucus. The power for locomotion in these animals is provided by rhythmic muscular contractions (pedal waves) on the ventral surface of the foot, and the force due to these waves is coupled to the substratum by the pedal mucus. Movement of the animal is consequently a result of a balance of forces: if the reactive force provided by the adhesive beneath the stationary portions of the foot is sufficient to offset the frictional force caused by portions of the foot moving forward, the animal is able to crawl. Mechanisms previously proposed to explain this adhesive crawling require that the frictional resistance to forward movement be minimized by the foot being lifted during the passage of a pedal wave. This lifting, however, cannot be reconciled with the properties of the pedal mucus and epithelium, and the terrestrial slug, *Ariolimax columbianus*, is shown to not lift its foot when crawling. A new mechanism is proposed which does not require lifting of the foot and explains several difficulties inherent in previous studies. Based on this mechanism and the mechanical properties of pedal mucus, a quantitative model is constructed which predicts the frictional and reactive forces, and these predictions are tested against force measurements made under crawling slugs. The model accurately predicts the magnitude of forces under slugs crawling horizontally, but is less accurate in predicting forces under slugs crawling vertically.

INTRODUCTION

The crawling of gastropods is subject to different constraints from those of more familiar forms of locomotion. In contrast to most animals a gastropod possesses a single locomotory appendage, the hydrostatically supported, muscular foot; and the animal's range of movements is limited to those compatible with this single structure. In addition, the animal adheres to the surface upon which it crawls. This adhesion is the result of a thin layer of mucus which coats the foot, acting as a glue, and is of obvious advantage to the animal in resisting forces (e.g. gravity and drag forces) which

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tend to pull the animal away from food or shelter. Foot movements during locomotion must not disrupt this adhesion, and may themselves be limited by the strength of adhesion. Given these constraints it is interesting to examine the movements of the gastropod foot, the forces that they cause, and how these result in locomotion.

Many aquatic gastropods move by means of cilia, and it has been hypothesized (Elves, 1961) that ciliary locomotion is the primitive form of movement for gastropods. Lists of species using ciliary locomotion, and the various parameters of their movement are reported by Miller (1974). Most gastropods, however, move by means of muscular pedal waves present on the ventral surface of the foot. A variety of wave patterns are present in gastropods (Lissman, 1945*a, b*; Jones & Trueman, 1970; Jones, 1973, 1975; Miller, 1974; Gainey, 1976) and are classified according to a scheme proposed by Vlès (1907). Waves that move in the same direction as the animal are termed direct, while waves moving in the opposite direction are retrograde. A single wave may extend across the whole foot (monotaxic) or waves may be only half the width of the foot, alternating sides (ditaxic). Most species may be described by these terms though a few have waves that move diagonally along the foot or appear and disappear haphazardly. The wave pattern that has been most thoroughly studied is the direct, monotaxic (Lissman, 1945*a, b*; Jones, 1973, 1975) and these descriptions have formed the basis for the accepted model of the kinematics of gastropod locomotion (e.g. Jones, 1975; Trueman & Jones, 1977).

In spite of the amount of information available concerning the movements of the gastropod foot during crawling, the mechanism by which these movements result in locomotion has received very little attention. Lissman (1945*b*) in the principal study of this subject, proposed that locomotion using direct pedal waves is a result of the interaction of six types of force as shown in Fig. 1.

(1) An internal force acts to extend the foot. This force is probably due to hydrostatic pressure in the haemocoel.

(2) The contraction of muscles produces an internal force which compresses the foot.

(3) A frictional force exists between the substratum and the portions of the foot that are moving forward. This is the force which must be overcome if the animal is to crawl, and its magnitude is a function of the speed of movement and the mechanical properties of the pedal mucus.

(4) A reactive force exists opposing the frictional force of movement. This force acts through the mucus under those parts of the foot that are stationary.

(5) A tension may exist between adjacent stationary portions of the foot, or

(6) A thrust may exist between stationary portions of the foot.

Of these six forces, 3, 4, 5 and 6 are external forces and can be quantified by measuring the forces exerted on the substratum. Lissman (1945*b*) measured these forces, and on the basis of these measurements a qualitative model of locomotion was proposed. The basic premise of this model is quite simple and undoubtedly valid: if the reactive force (Type 4) provided by the adhesive mucus beneath the stationary parts of the foot is sufficient to offset the frictional force of movement (Type 3), the animal will move forward. This simple balance of forces was complicated, however, by the need to explain certain aspects of the force measurements. The explanation

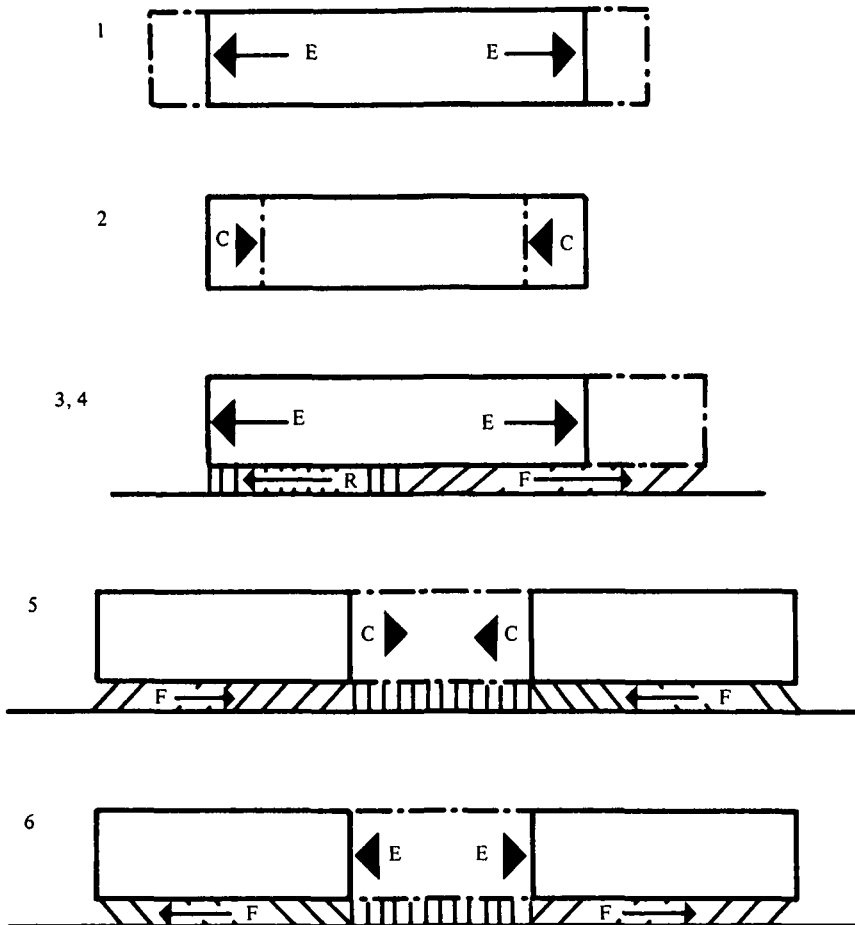


Fig. 1. The possible forces acting during gastropod locomotion (after Lissman, 1945*b*). (1) An internal force of expansion, *E*, causes the foot to elongate. (2) An internal force of compression, *C*, causes the foot to become shorter. (3) and (4) An elongating foot segment in contact with the ground will result in a frictional resistance to movement, *F*, and a reactive force resisting this movement, *R*. (5) and (6) Compression or elongation of the central of three segments will result in a frictional resistive force.

proposed requires thrusts (Type 6) between adjacent stationary areas in the anterior half of the foot, and tensions (Type 5) between stationary areas in the posterior half of the foot. The force measurements which led to the need to postulate these complicating tensions and thrusts may have been the result of the use of an inappropriate force transducer. Lissman's apparatus consists of a bridge of variable width mounted on a pendulum; the more force placed on the bridge the larger the displacement of the pendulum. The precise dimensions of the apparatus are not given, but from diagrams shown in Gray & Lissman (1938) it can be estimated that the period of the pendulum is about 0.5 to 1.0 s. If this is so, the device is incapable of accurately measuring oscillating forces with a period of one second or less, such as those associated with pulmonate crawling. Jones (1975) made force recordings beneath slugs with an isometric transducer, presumably with a high frequency response, and these recordings

differ from those of Lissman (1945*b*) both in the magnitude and direction of the force due to individual pedal waves. These differences have not, however, led to a re-interpretation of the model of Lissman (1945*b*).

Aside from the complications of tensions and thrusts the models of Lissman and Jones are severely limited in that they are strictly qualitative. Lacking knowledge of the properties of the pedal mucus neither Lissman (1945*b*) nor Jones (1975) could predict the magnitude of the frictional and reactive forces. While qualitative models are often useful, their inability to make quantitative predictions renders them difficult to test.

This study examines the movements of the pulmonate slug, *Ariolimax columbianus*, and on the basis of these findings proposes a quantitative model that incorporates many of the features of the models of Lissman (1945*b*) and Jones (1975). The predictions of the model are tested against forces measured under crawling slugs.

METHODS

A. columbianus were collected in the woods near the University of British Columbia, Vancouver, B.C., and were kept at 10 °C. All tests were carried out at room temperature (21–23 °C), and slugs brought into the laboratory from the cold room were allowed to come to room temperature before tests were conducted. The locomotory kinematics of *A. columbianus* were observed by allowing the slug to crawl on a glass plate, the foot being viewed through the plate. Movements of the ventral surface of the foot were recorded with a Sony television camera and videotape recorder at 60 frames per s, and taped records were played back onto a television screen. For large scale measurements a ruler taped to the glass plate near the slug allowed for the size calibration of the image on the screen. For small scale measurements the camera was mounted on a dissecting microscope (Wild M5) and the image calibrated with a 1 mm grid taped to the glass. Magnification in this case was such that a 4 mm length of foot filled the entire vertical dimension of the screen. At this magnification individual blemishes and concentrations of mucus gland cells are visible and their movements can be recorded. The spatial resolution of the system at this magnification is 20–30 μm , a factor controlled primarily by the 'jitter' of the image on the TV screen. As a consequence of this jitter a strict frame by frame analysis is not possible. It is necessary that 5–10 frames be examined before it can be stated with any certainty that a point has moved. Thus, while a movement of 20–30 μm can be detected between the start and finish of a 5–10 frame segment, the time within this segment when the movement actually occurred cannot be specified; spatial resolution being gained at the expense of temporal resolution. The structure and vertical movements of the foot during locomotion were examined using a method similar to Lissman (1945*c*) and Jones (1973). Small (3–4 cm) *A. columbianus* were placed on a strip of aluminum foil. When the slugs were actively moving the strip was dropped into liquid nitrogen, rapidly freezing the slugs. The slugs were then fixed for one week while still frozen (1 % glutaraldehyde–50 % ETOH at –20 °C), dehydrated, wax embedded, and sectioned at 8–10 μm according to standard histological techniques. Sections were stained with either Mallory's triple stain for examining the general structure of the foot, or alcian blue/eosin when the location of mucus and mucus-producing cells were to be examined. This method

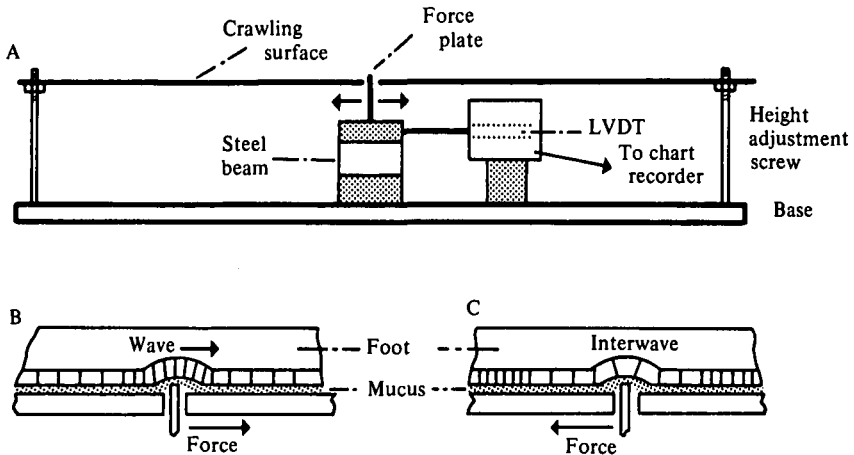


Fig. 2. An apparatus for measuring the forces beneath a crawling slug. (A) Schematic representation of the apparatus. (B) and (C) The dimensions of the force plate are smaller than either a wave or an interwave. LVDT = linearly variable differential transformer.

differs from those of Lissman and Jones in that the animals are frozen while crawling on a non-porous, relatively inflexible surface: Lissman used a copper screen while Jones used strips of filter paper.

An apparatus was constructed to measure the locomotory forces of slugs (see Fig. 2). A small (0.5×1.0 mm) force plate protrudes through a 1.5 mm hole in a large plexi-glass crawling surface, and is supported by two steel beams. A force in the plane of the crawling surface (in the direction shown in the figure) causes the plate to move; the larger the force, the larger the deflexion. The deflexion of the beam is measured by a linearly variable differential transformer, the output of this being amplified and recorded on a chart recorder. The transducer is calibrated by turning the apparatus on its side (so that the measuring beams are horizontal), and hanging accurately known weights from the force plate. Forces as small as about 5 dynes can be accurately measured, and within the range of forces encountered during testing the transducer output is linear with force. The unloaded resonant frequency of the apparatus is 100 Hz, sufficiently high to allow accurate measurement of the 1 Hz oscillatory forces associated with slug locomotion.

A test is performed by orienting the apparatus so that the crawling surface is either horizontal or vertical, and allowing the slug to crawl across the force plate along the direction in which the plate is sensitive to force. The dimension of the force plate in this direction (0.05 mm) is less than the shortest length of a pedal wave. Thus, as a slug passes over the force plate, the forces beneath the various areas of the foot can be measured. Since the area of the force plate is known, the measured forces can be expressed as force/area, i.e. stress, for comparison with the predictions of the locomotory model. It has been amply shown by Lissman (1945*b*) that the passage of a compressional wave corresponds to an anteriorly directed force. These observations were corroborated for *A. columbianus* by visually observing the ventral surface of the foot as it passed over the force plate, but were not tested further.

This apparatus is less than ideal in two respects.

(1) Because the force plate must move in order to measure force, it must protrude through a hole in the plexiglass surface. The presence of this hole, as will be shown, allows the slug to lift its foot as a wave passes over the force plate. Thus as long as the force plate is flush with the surface only the forces beneath the interwaves (where the foot is not lifted) are measured, and if the plate is slightly below the surface these are not measured accurately. This problem is remedied by raising the force plate above the crawling surface slightly ($50\text{--}150\text{ }\mu\text{m}$), as this maintains the contact between the foot and the force plate at all times. Apparently while the slug can lift its foot in the general region of the hole, it cannot lift it sufficiently in the localized region of the force plate to break contact. Once the force plate is raised sufficiently to maintain contact, the distance that it is raised, within the limits $50\text{--}150\text{ }\mu\text{m}$, does not effect the stress measured: the correlation between force plate height and overall force amplitude has a correlation coefficient of 0.059 and is not significant. The distance the plate is raised is measured with a micrometer.

(2) The second problem encountered with this apparatus is again attributable to the presence of a hole in the plexiglass surface. It was noted in a large percentage of the tests that the stress measured after the slug had passed was different from the zero stress measured before the slug first reached the plate. The magnitude and even the direction of this zero shift is unpredictable; the explanation being that solid mucus builds up in the gap either in front or behind the force plate. While this zero shift does not effect the overall amplitude of stresses measured, it makes it impossible to measure the relative amounts of this overall amplitude that are directed anteriorly and posteriorly, and as a consequence, test results are divided into two categories. The small percentage of tests where the zero stress level is shifted less than 10% of the overall stress amplitude during the passage of a slug are placed in one category and stresses are measured relative to the zero level. Tests where the zero level is shifted by a larger amount are placed in the second category and only the overall amplitude of the stress oscillation is measured.

The speeds at which the slugs walked during the tests could not be accurately measured. A slug is capable of crawling such that the anterior half of the body moves at a different speed than that of the posterior half, and measuring the speed of either head or tail does not give an accurate measure of the speed at the force plate. The presence of the force transducer precluded accurate speed measurements at the force plate. In lieu of these measurements, it is assumed that the range of walking speeds in these tests is similar to those observed in the kinematic studies, and this range is used in the calculations of the locomotion model.

Simultaneous pressure and force measurements were conducted using a force plate modified as shown in Fig. 3. The essential modification consists of replacing the force measuring plate with a hollow tube cut from a 20 gauge hypodermic needle, this being connected by a rigid plastic tube to a pressure transducer. All tubing is filled with degassed water. As the slug crawls over this new force plate any force directed dorso-ventrally is detected by the pressure transducer. Unfortunately, the compliance of the transducer does not allow for the accurate measurement of pressures associated with volume changes as small as those occurring under a pedal wave. While the magnitude

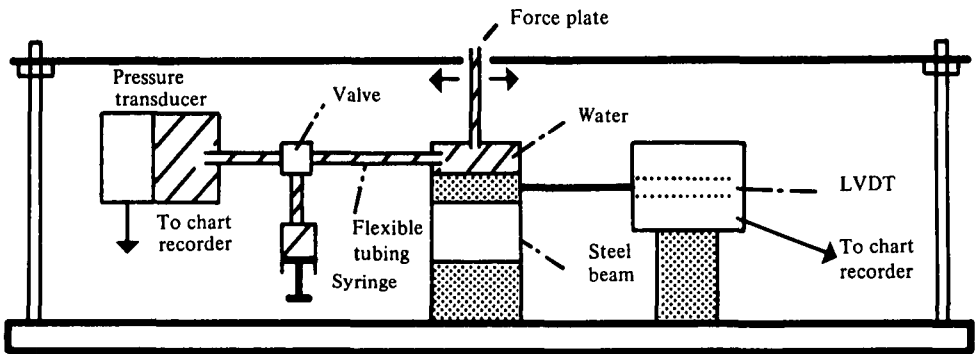


Fig. 3. A schematic drawing of the apparatus for simultaneously measuring antero-posterior forces and dorso-ventral forces beneath a crawling slug. The syringe and valve are used to adjust the level of the degassed water in the system.

of pressure measured by this apparatus is thus an underestimate of the actual pressure, the apparatus does accurately measure the direction of dorso-ventrally directed forces. In addition to pressure measurements the apparatus can still measure antero-posteriorly directed forces.

Kinematics

Gross foot movements

A stationary slug shows no evidence of pedal waves. The foot is dark tan, darkest near the centre of the foot where the digestive gland shows through the thin layer of pedal musculature. As the slug begins to move waves first appear $1/4$ to $1/3$ of the foot length back from the anterior end of the foot. As the pedal musculature and epithelium are compressed in the wave they mask the digestive gland and the wave appears lighter than the surrounding areas. As the initial waves move forward the area of wave formation moves posteriorly until waves are present along the entire foot as shown in Fig. 4. Thirteen to 17 waves are present on the foot, waves alternating with interwaves. The wave pattern occupies the central $2/3$ of the foot, waveless areas forming a 'rim'. The areas of the waves, interwaves, and rim have been measured and values for a typical (15 g) slug are shown in Fig. 4.

The overall movements during locomotion are similar to those described by Jones (1973) for *Agriolimax reticulatus*, and are best understood by following a single wave. After the full wave pattern is established each wave is formed near the posterior end of the foot by a contraction of the pedal musculature. This contraction causes an area of the foot to compress antero-posteriorly, and the tail moves forward. From video recordings of pigmented areas on the foot the extent of this compression was measured on three *Ariolimax columbianus*, 3–4 waves being measured on each slug. The ratio of extended length to compressed length (the compression ratio) ranges from 1.43 to 2.03 with an average of 1.69, again in agreement with values measured on *Agriolimax reticulatus* (Jones, 1973).

Each wave of compression is passed along the foot as muscles ahead of the wave contract and muscles behind relax. When the wave reaches the anterior end of the foot, muscles are no longer available to keep the wave compressed and it returns to its extended length. The force for this expansion is presumably provided by hydrostatic

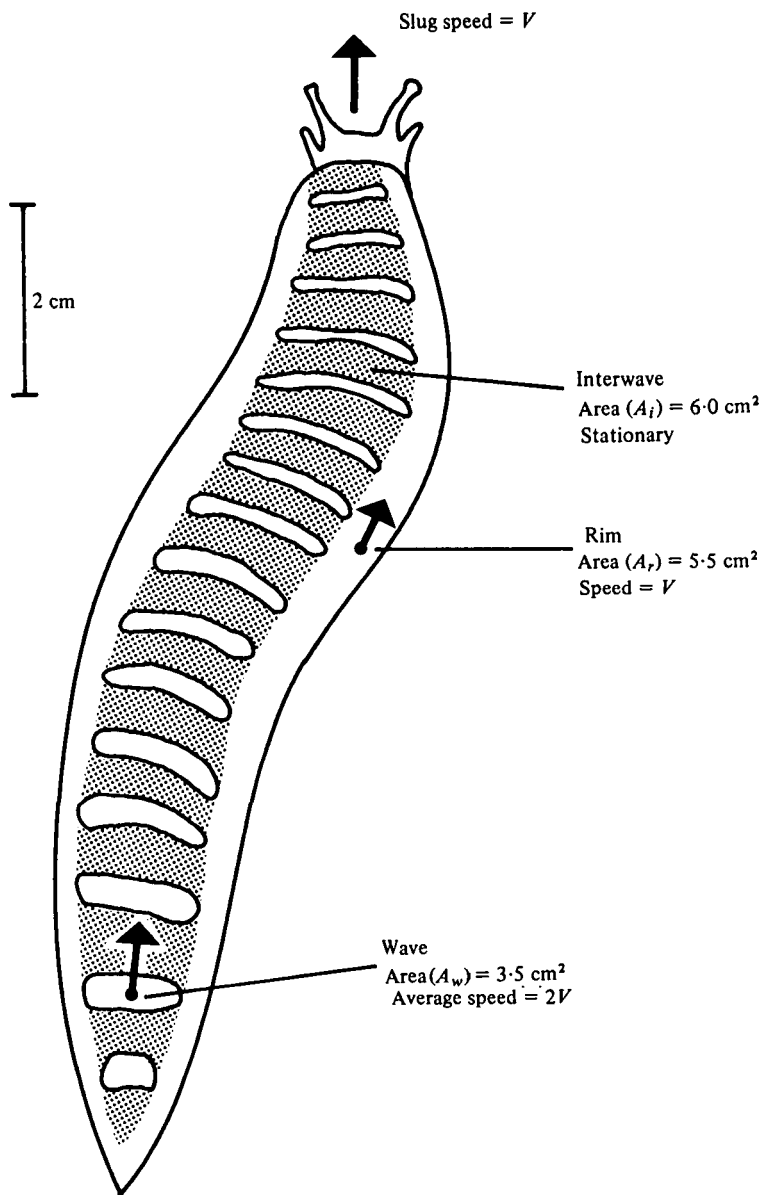


Fig. 4. A schematic representation of the ventral surface of the foot of a typical *A. columbianus*, showing the relative positions, movements, and areas of the rim, waves, and interwaves. The length of the arrows is proportional to speed.

pressure within the foot and will be discussed later. As a consequence of the re-expansion of this portion of the foot, the anterior end of the foot is moved forward. Thus each wave constitutes one step; the distance the tail is pulled forward when the wave forms being transferred to the head when the wave of compression re-expands. *A. columbianus* advances about 1.0 to 1.5 mm with each wave.

The three areas of the foot move at different rates: The rim moves at the same constant speed as the slug as a whole; portions of the foot contained in waves move forward more rapidly than the whole slug, while interwaves are stationary relative

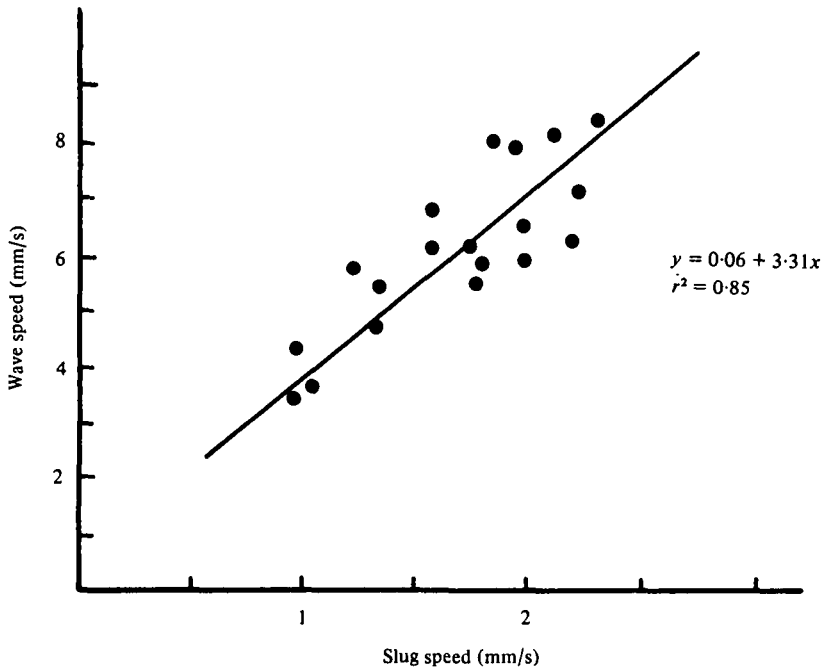


Fig. 5. The relation between the speed of the pedal waves and the overall speed of the slug. The waves move more than three times as fast as the slug, and the slug controls its crawling speed by varying the speed of pedal waves.

the ground. The average speed of the central portion of the foot must equal the speed of the whole slug, consequently the speed at which a point travels while in a wave depends on the relative periods of time spent moving and stationary. Measurements from 22 video recordings of crawling *A. columbianus* show that points on the foot spend approximately equal periods of time in and out of pedal waves. Thus the average speed of a point in a wave (the point speed) must be twice that of the whole slug.

Fig. 5 is a plot of wave speed and overall slug speed for 18 measurements from 8 slugs. The ratio of wave speed to slug speed is about 3.3, a fact which may be used as a check on the value of the compression ratio. Since wave speed is 3.3 times the overall slug speed, a wave will advance into a stationary interwave at 3.3 times the slug speed. Points moving in a wave, however, are themselves travelling at 2 times the slug speed or at a speed relative to the wave of $2/3.3 = 0.61$. Since points in a wave move slower relative to the wave than points in interwave, the antero-posterior dimension of a wave must be less than an interwave by a ratio $1/0.61 = 1.65$, if equal time is to be spent moving and stationary. The calculated value of compression ratio is thus 1.65 which compares closely to the measured average of 1.69.

The information of Fig. 5 shows that *A. columbianus* is similar to other pulmonates in controlling its locomotory speed by controlling the speed of the pedal waves (Jones, 1975; Crozier & Pilz, 1924).

The observations reported here for *A. columbianus* are an accurate description of the cross movements of the foot during locomotion. However, the forces which propel the

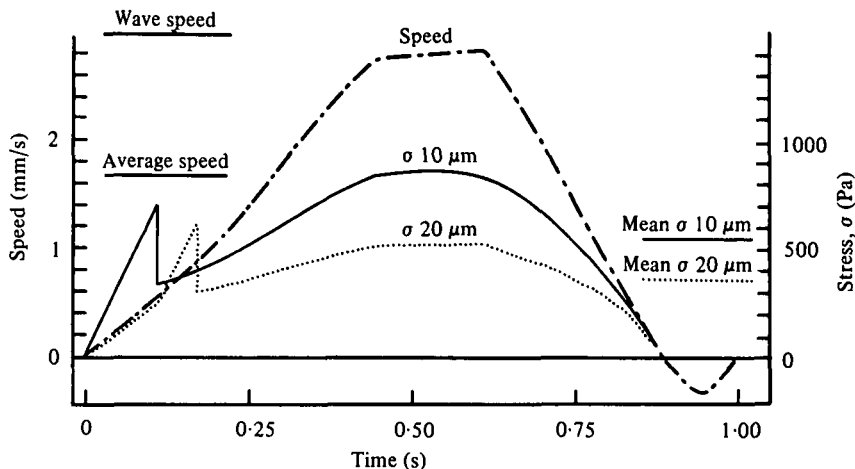


Fig. 6. The speed and stress profiles of an average pedal wave. The velocity curve represents values averaged from 22 separate crawls by two *Ariolimax columbianus*. Stress is calculated for the speed profile shown and for mucus layer thicknesses of 10 and 20 μm .

slug are due to the interaction of *individual points* on the foot with the substratum via the pedal mucus. Fig. 6 shows an average velocity profile for a point on the foot during the passage of a pedal wave. As a wave overtakes a point, that point is gradually accelerated to a peak speed considerably greater than the average slug speed and approaching the wave speed. The peak speed is maintained for a short time before the point is decelerated back to zero. It is sometimes found that deceleration continues past zero, i.e. there is some backslip after the wave has passed. This backslip is small, amounting at most to 30–50 μm . As a consequence of this small amount of backslip the time course of backslip is uncertain, as explained in the methods. It is certain however that it occurs in the first 0.008–0.17 s (5–10 frames) after zero velocity is reached.

In sagittal or parasagittal sections of the foot taken from frozen slugs the waves can be seen (Fig. 7). These sections suggest that the slug does not lift its foot during the passage of a pedal wave when walking on a nonporous, inflexible surface. Instead the pedal wave consists entirely of antero-posterior compression, as seen by examining the shape of epithelial cells. In the extended portion of the foot the cells are long antero-posteriorly, and short dorso-ventrally; these dimensions being reversed in the compressed areas of the foot. The compression ratio measured on the section shown in Fig. 7 is about 2, in rough agreement with values calculated by other means. In many sections the pedal mucus layer had become dislodged during fixation and embedding, but in those cases where the mucus layer was apparently intact it was found to be 10–20 μm in thickness.

The foot structure of *A. columbianus* is similar to that of other slugs (Barr, 1926, 1928). Cilia are present and form two distinct areas. The extreme edges of the foot are densely covered with cilia approximately 4 μm long. The direction of the effective stroke of these cilia is such that mucus is propelled posteriorly. The sole of the foot is also ciliated, but the cilia are shorter and more sparsely distributed. The direction of the effective stroke of these cilia was not determined, but Barr (1926, 1928) found that pedal cilia in other slugs transported mucus laterally and posteriorly. The pedal mucus

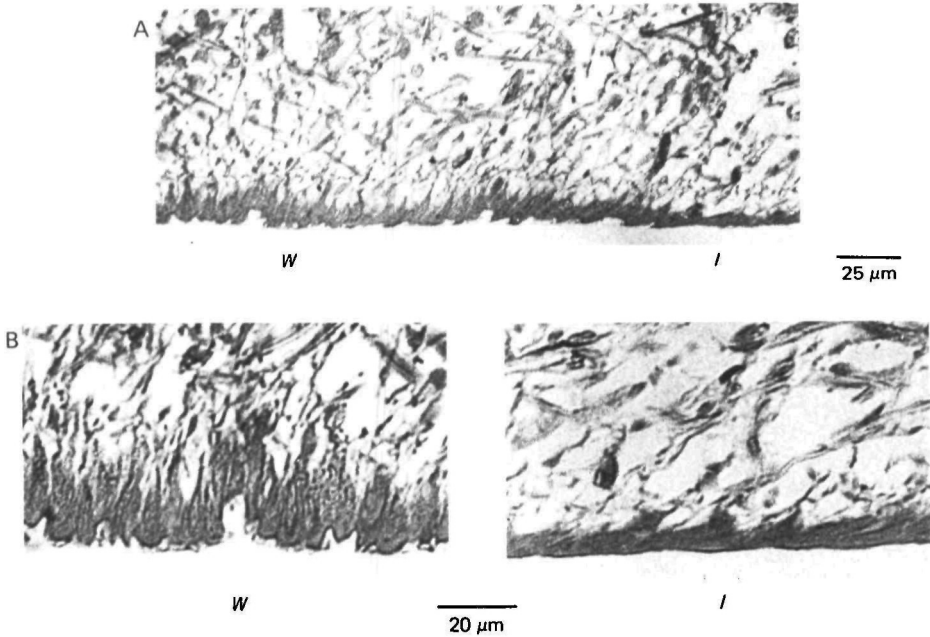


Fig. 7. A micrograph showing the pedal epithelium of a crawling slug. (A) A sagittal section showing the transition between a wave (*W*) and an interwave (*I*). Notice that the foot is not lifted in the wave. (B) A higher magnification of the epithelium under a wave. The epithelial cells are compressed antero-posteriorly. (C) A higher magnification of the epithelium under an interwave. The epithelial cells are extended antero-posteriorly. The pedal mucus layer has become dislodged and is not seen in these sections.

■ *A. columbianus* is produced by the suprapedal mucus gland and exuded from a pore beneath the mouth. Several authors (Barr, 1926, 1928; Jones, 1973) have reported the presence of a watery pedal mucus in addition to that of the suprapedal gland, presumably produced by the pedal epithelium. This watery mucus was not observed in *A. columbianus*.

The results reported here differ substantively from those of Lissman (1945*a*) working with *Helix aspersa* and *pomatia* and Jones (1973) working with *Agriolimax reticulatus* only in that the foot of *Ariolimax columbianus* is not lifted during the passage of a pedal wave, while those of *Helix* and *Agriolimax* are. This difference is important in that it bears directly on the mechanism of locomotion. In order for a slug to move forward the force required to move the waves and rim against the frictional resistance of the mucus and substratum must be less than the total resistance to movement provided by the interwaves. It has been assumed (Lissman, 1945*a, b*; Jones, 1973) that this difference between forwards and backwards resistances is a result of the foot being lifted during the passage of a pedal wave, as observed for *Helix* and *Agriolimax*. It is possible that the different pedal wave shape reported here is simply due to species differences. Another explanation, however, appears more likely. Regardless of the attractiveness of a lifted wave mechanism for crawling, this explanation suffers from three major problems, all related to the mucus sandwiched between the foot and the substratum.

(1) It has been assumed (Lissman, 1945*a, b*; Jones, 1973; Jones & Trueman, 1970) that when the foot is lifted the space beneath the lifted portion is filled with either mucus or some fluid exuded by the foot. As the wave travels forward this volume of mucus travels with it and should be deposited in front of the foot. This process has never been observed. Jones (1973) speculates, but without evidence, that this mucus is somehow either reabsorbed into the foot, or prevented by ciliary action from travelling with the wave.

(2) Once the foot is lifted in a wave it must be returned to the substratum to which it will adhere during the interwave. Mechanisms proposed to date (Lissman 1945*b*; Jones, 1973; Jones & Trueman, 1970) to account for the lifting and subsequent lowering of the foot require two processes to occur simultaneously. (1) The contraction of muscles which raise the foot pulls upwards and forwards on the mucus beneath the anterior half of a wave, creating an area of low pressure. It is this low pressure which, if it exists, would cause fluid to move out of the foot to fill the space beneath the wave. (2) At the same time the hydrostatic pressure within the foot (presumably responsible for forcing the foot back to the substratum) is pushing downwards and forwards on the mucus beneath the posterior half of the wave, creating an area of high pressure. However, unless the wave were to move at a very great speed, thereby creating a large dynamic pressure, or a rigid partition were to separate the two halves of the wave (and no such partition has been found) the downwards and upwards forces will cancel and all the mucus beneath the wave will be at essentially ambient pressure. Jones & Trueman (1970) however measure a substantial negative pressure (i.e. an upwards force) under the retrograde pedal waves of the limpet *Patella vulgata*, and Jones (1973, 1975) reports a similar force for *Agriolimax reticulatus*.

(3) It can be calculated that the force necessary to lift the foot from the substratum

as the animal first begins to move is beyond the capability of the slug's body structure and musculature. If the values for pedal wave dimensions reported by Jones (1973) are used as an example, a pedal wave is 0.05 cm long, 0.02 cm high at its highest point, and roughly 0.2 cm wide for a volume of 1×10^{-4} ml. The fluid to fill this volume must come from either the pedal haemocoel or the mucus beneath surrounding areas of the foot, and must be drawn in within half the time of passage of a single wave, about 1/2 s. If the fluid comes from the haemocoel it must thus come across the pedal epithelium with a flux of 1.56×10^{-2} ml/cm².s. Machin (1966) reports the transfer coefficient of *Helix aspersa* dorsal epithelium as 47 mg/cm².h.mmHg (9.63×10^{-8} ml/cm².sec.Pa). If the slug pedal epithelium shows a similar value, the stress which must be placed on the pedal epithelium to provide the necessary flux is 162 kPa. If a slug is sectioned parallel to the foot surface muscle fibres are found to account for only about one quarter to one third of the area, and Jones (1973) reports that these fibres run at an angle of 20–30° to the surface. Thus the stress on the muscle fibres required to lift the foot is 1400–1900 kPa. Several molluscan muscles have been found to contract with a maximum isometric stress of 450–510 kPa (Schmidt-Nielsen, 1979), and less stress would be produced if the muscle shortens during contraction as it would in this case. Thus it seems unlikely that a slug could pull hard enough on the ventral surface of its foot to force fluid out at the rate required. Further, if these estimates of muscle strength are inaccurate, and the muscles can actually provide the necessary stress, it seems unlikely that the very flexible body of the slug is capable of supporting such stress; the contraction merely serving to deform the body rather than lifting the foot.

A similar argument applies if the fluid to fill the wave space is supplied from the mucus of surrounding areas. Crisp (1973) calculates the stress required to separate two discs immersed in a viscous fluid, and has applied these calculations to the adhesion of barnacles. The same equation will be used here to estimate the force required to overcome mucus resistance:

$$\text{stress} = 1.5 r^2 \eta (dy/dt) y^{-3},$$

where r is the radius of the disc, η is the viscosity of the fluid and y is the initial thickness of the fluid layer. A pedal wave is not circular but as a first approximation one half the length of the wave ($= 0.025$ cm) may be taken as a typical radius. The initial thickness of the mucus is about 15 μ m and the viscosity is about 40 poise (Denny & Gosline, 1980). The foot must be lifted 0.02 cm in 1/2 s thus $dy/dt = 0.04$ cm/s. The stress is thus 4.4×10^{10} Pa, a value which obviously cannot be supplied by the muscles or supported by the body. This calculation certainly overestimates the true adhesive capability of the mucus (for example, even under ideal conditions the water in the mucus would cavitate at a stress of less than 10^{10} Pa) but does not alter the fact that the forces of adhesion are very strong compared to the capabilities of the muscles and body. For all of the above reasons it seems unlikely that a slug will lift its foot with each pedal wave when it is crawling on a nonporous surface.

A model for slug kinematics

The results of this study suggest a mechanism that both avoids the problems of lifting the foot and explains the results of previous authors. It is essentially an altera-

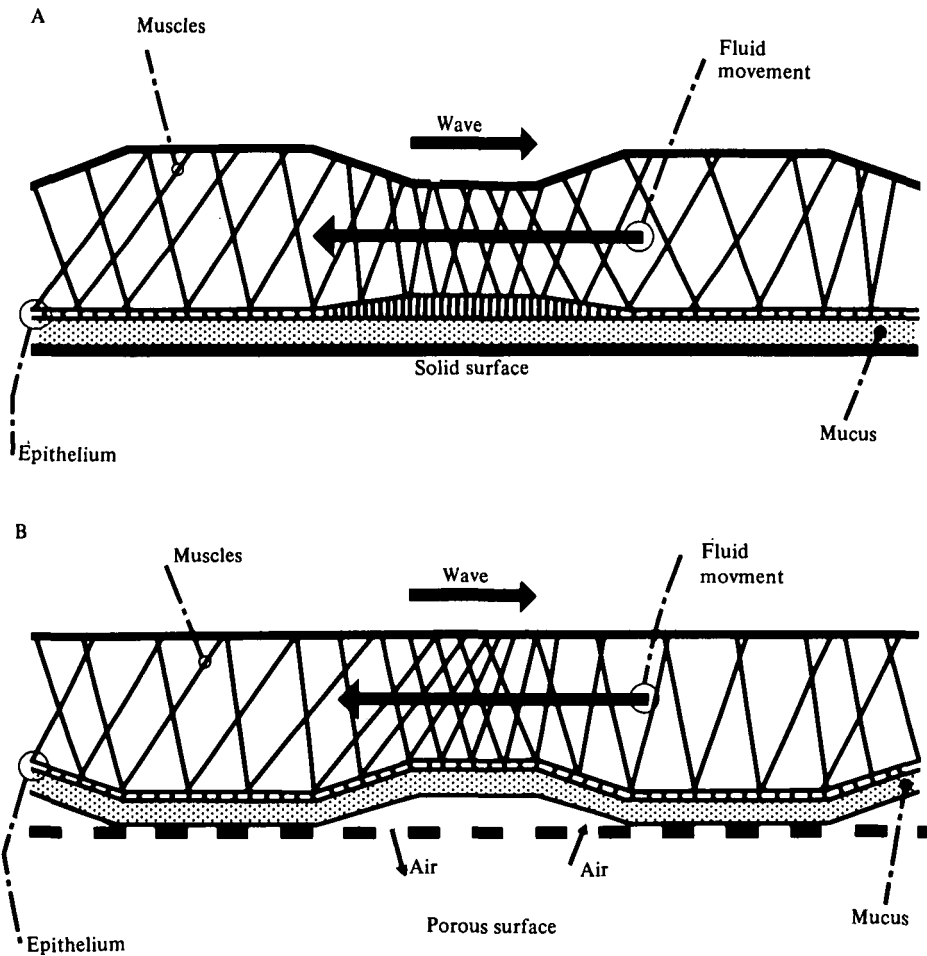


Fig. 8. A model for the movement of the foot of *Ariolimax columbianus* on a solid substratum (A) and a porous or flexible substratum (B). The forward movement of the wave and the decreased thickness of the foot result in a high hydrostatic pressure in the haemocoel ahead of the wave.

tion of the mechanism proposed by Jones (1973) and is shown schematically in Fig. 8. When the animal is crawling on a nonporous surface the muscles and connective tissue of the foot are incapable of lifting the foot from the substratum, as explained above. Unable to lift the foot, the oblique muscles instead act to pull the body down, thereby narrowing the spaces of the pedal haemocoel. The force required to pull the body down is counteracted by a force pulling upwards on the mucus beneath the foot. This upwards force appears as a negative pressure to a sensor on the substratum. Without a better understanding of the internal kinetics of the foot the magnitude of this upwards force cannot be predicted, however it should be possible to detect if it is present. A series of six tests on three *A. columbianus* (using the apparatus of Fig. 4) confirmed that there is a dorsally directed force acting under a pedal wave. The record from one test is shown in Fig. 9. As the wave moves forward, the fluid in the

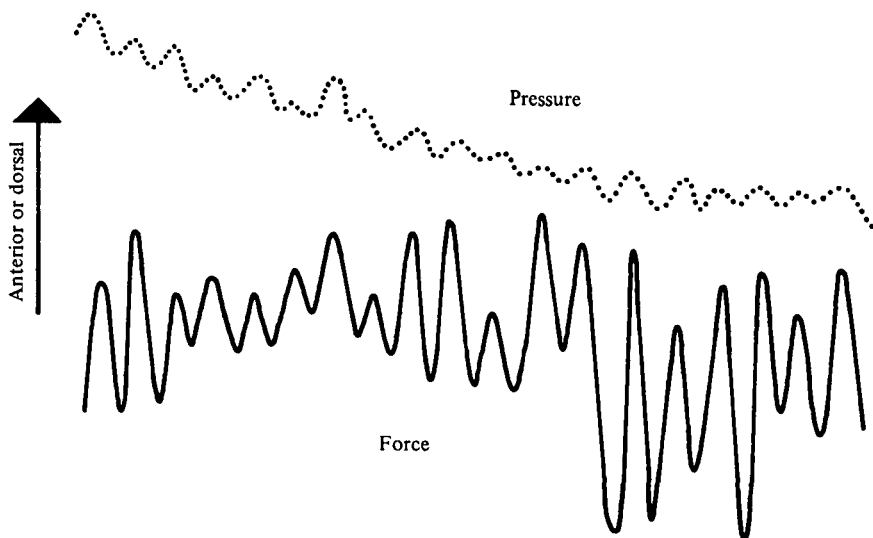


Fig. 9. An example of simultaneous force and pressure measurements. The dorsally directed force peaks at the same time as the anteriorly directed force. The overall slope of the pressure trace is due to temperature drift in the transducer.

pedal haemocoel is forced to flow through the narrowed channel in the area of muscular contraction, raising the pressure of the haemocoelic fluid ahead of the wave. Since the pressure required to force fluid through a tube at a given rate is an inverse fourth power function of radius, it is only necessary for the spaces of the haemocoel to be narrowed slightly to create a substantial pressure in the haemocoel ahead of the wave. The haemocoelic spaces in the lateral part of the foot are typically smaller than those in the central portion of the foot, so that hydrostatic pressure will not leak around the edges of the wave. It is this haemocoelic hydrostatic pressure which is responsible for re-extending the foot at its anterior end. This model does not require that mucus or fluid beneath the foot be transported forward with each wave, and since the foot is never lifted there is no problem with getting it back down.

When the slug is crawling on a porous or flexible surface the situation is changed only in that the oblique muscles are then capable of either pulling the foot away from the substratum or deforming the substratum itself. In the case of a porous surface the ability to detach the foot is a consequence of the surface's porosity which allows air to replace the displaced mucus easily. It is much easier for air (viscosity = 1.8×10^{-4} poise) to move into this space than it would be for mucus (40 poise). The lowering of the foot is accomplished by the hydrostatic pressure in the haemocoel, created as explained above. In order for the foot to be forced down only the air beneath the wave must be displaced, a process again facilitated by the porosity of the surface. In nature slugs are required to crawl over surfaces of many types. While most of these are porous or flexible there are surfaces such as the bark and leaves of some trees on which the slug would not be able to lift its foot, and the model described here for a nonporous surface would operate.

While this model can account for the movements of the foot it would seem to pose a problem as to the mechanism of locomotion. If the foot is not lifted in a pedal wave the frictional resistance to movement must be lowered sufficiently by other means to

low for locomotion. It thus seems likely that in circumstances where the foot is not lifted a change in the properties of the pedal mucus must occur beneath the waves. Such a change has been reported for *A. columbianus* pedal mucus by Denny & Gosline (1980), and these properties can be combined with this kinematic model to produce a quantitative model of locomotion.

Locomotion model

The basic premise of this model is the same as that of Lissman (1945*b*): a gastropod will move forward if the *reactive force* beneath the stationary parts of the foot (the interwaves) is sufficient to offset the *frictional force* due to the movement of the waves and rim. The prediction of these two important forces is considerably simplified if the following constraints are imposed.

(1) The model proposed here is designed to predict the forces operating under conditions such that the foot is not lifted from the substratum during the passage of a pedal wave.

(2) The slug as a whole is assumed to move at a constant rate, and the model does not take into account forces due to the acceleration or deceleration of portions of the foot. The masses and accelerations in pedal movements are small, and each part of the foot that is accelerating is offset by an equivalent part decelerating so that even the small forces present should tend to cancel.

(3) The model does not allow for the presence of tensions or thrusts between interwaves, in essence requiring that the length by which the foot contracts on entering a wave be simultaneously offset by the foot re-extending on leaving a wave.

(4) No account is made for the action of those cilia present on the pedal epithelium.

(5) The pedal mucus is assumed to have the mechanical properties reported as typical by Denny & Gosline (1980).

The model, incorporating these simplifying assumptions, will be tested against actual force measurements, and the validity of these assumptions can then be examined.

Waves

As a point on the foot is overtaken by a pedal wave it begins to move forward, shearing the mucus beneath it. The shear strain, γ , is equal to the distance moved by the foot divided by the thickness of the mucus layer (see Fig. 10*a*). The force necessary to shear the mucus increases linearly with γ until at a strain of 5 to 6 the mucus is sheared beyond its yield point and it flows (Denny & Gosline, 1980). The magnitude of the yield stress, σ_y (in Pa), is a function of the strain rate, $\dot{\gamma}$, such that:

$$\sigma_y = 6.9 \dot{\gamma} + 313. \quad \text{eqn. 1 (Denny \& Gosline, 1980)}$$

After the mucus has yielded, as long as the foot moves forward it moves over a viscous fluid. The viscosity of the fluid and the strain rate determine the flow stress, σ_f :

$$\sigma_f = 2.3 \dot{\gamma} + 199. \quad \text{eqn. 2 (Denny \& Gosline, 1980)}$$

From these relationships the stress operating under each point in a moving wave can be calculated. The calculations are graphed in Fig. 6 for a typical wave and two values of mucus thickness. The overall force of moving waves forward, F_w , is the average stress under the wave (as shown in Fig. 6) times A_w , the total wave area (see Fig. 10).

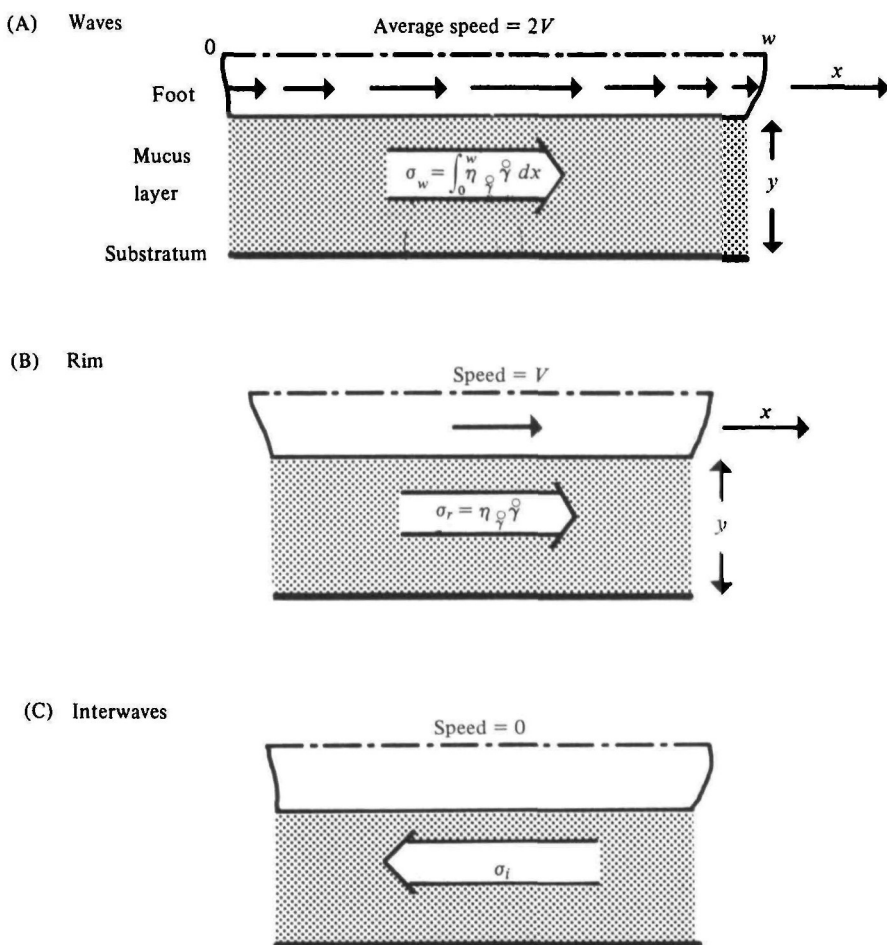


Fig. 10. The forces present under a moving slug. (A) Waves. The stress at any point is equal to the shear rate, $\dot{\gamma}$, times the viscosity at that shear rate, $\eta\dot{\gamma}$. The sum of all stresses under a wave is the wave stress (σ_w). The wave stress times the wave area equals the wave force, F_w . (B) Rim. The rim stress is equal to the shear rate under the rim times the viscosity at that shear rate. The rim stress (σ_r) times the rim area equals the rim force, F_r . (C) Interwaves. In order for locomotion to occur

$$F_i = -(F_r + F_w),$$

where F_i is the interwave force. Thus the interwave stress (σ_i) equals F_i divided by the interwave area.

This wave force places an equal and opposite force on the stationary portions of the foot, the interwaves.

Rim

Once sheared beyond its yield point, and for as long as the slug continues to move, the mucus beneath the rim remains in its fluid form. The rate of movement of the rim and the viscosity of the fluid mucus determine F_r , the resistance to movement:

$$F_r = \sigma_r A_r, \quad \text{eqn. 3 (for } \sigma_r \text{ see eqn. 2)}$$

where A_r is the area of the rim. The force required to move the rim forward places an additional force on the mucus beneath the interwaves.

Table 1. Predictions of the locomotion model

Slug Weight 15 g Foot area				
Rim		$5.5 \times 10^{-4} \text{ m}^2$		
Waves		$3.5 \times 10^{-4} \text{ m}^2$		
Interwaves		$6.0 \times 10^{-4} \text{ m}^2$		
		0.85 mm/s		2.00 mm/s
Slug speed				
mucus thickness	10 μm	20 μm	10 μm	20 μm
Shear rate (rim)	85/s	42.5/s	200/s	100/s
Shear stress (rim)	395 Pa	297 Pa	659 Pa	429 Pa
Force (rim)	0.217 N	0.163 N	0.363 N	0.236 N
Average shear stress (waves)	542 Pa	365 Pa	1059 Pa	610 Pa
Force (waves)	0.193 N	0.127 N	0.371 N	0.213 N
Total force (waves + rim)	0.410 N	0.290 N	0.724 N	0.449 N
Shear stress (interwaves) (horizontal)	682 Pa	485 Pa	1222 Pa	749 Pa
Overall stress amplitude (waves + interwaves) (horizontal)	1232 Pa	850 Pa	2281 Pa	1359 Pa
Overall stress amplitude (vertical up)	1482 Pa	1100 Pa	2531 Pa	1609 Pa
Overall stress amplitude (vertical down)	970 Pa	600 Pa	2031 Pa	1109 Pa

Interwaves

As a wave moves away from a point on the foot the point decelerates until it is stationary. As soon as the point is stationary the mucus beneath it begins to 'heal': the glycoprotein network of the mucus reforms, and the mucus becomes more solid as time passes (Denny & Gosline, 1980). The healing process occurs quickly so that most of the interwave rests on solid mucus, and this solid mucus holds the interwave stationary against the combined force of the waves and interwaves. The stress on the mucus is the force of forward motion ($F_w + F_r$) divided by A_i the interwave area.

This model thus predicts stress values under waves, rim, and interwaves as a function of $\dot{\gamma}$. The strain rate is in turn determined by the speed of the slug and the thickness of the pedal mucus layer. *A. columbianus* crawls at speeds typically ranging between 0.85 and 2.00 mm/s, with mucus thicknesses between 10 and 20 μm . These ranges in speed and mucus thickness have been used to calculate the approximate range in stresses that would occur under slugs crawling horizontally, and these values are shown in Table 1. If the slug is to move, the mucus under the interwaves must be able to resist the stress placed on it without yielding. Are the stress values calculated here less than the yield stress of pedal mucus as reported by Denny & Gosline (1980)?

The ability of the solid mucus beneath an interwave to withstand this stress without yielding is dependent on the shear rate that this stress causes. It has been shown here that upon entering an interwave, a point on the foot may slip backwards a distance of 30–50 μm in a time which, though not accurately measurable, is less than 0.08–0.17 s. The mucus in the interwaves is thus sheared at a rate which is at least 18–72/s, and the actual value may be higher. For the range of stresses calculated by the model it is necessary that shear have been applied at a rate of 60–120/s in order for the mucus not to yield. Thus, while technical limitations have not allowed precise measurement of actual shear rates, it is at least consistent with these observations that the mucus under the interwaves is sheared at a rate sufficiently high to withstand the calculated stresses.

Another question can be asked of these calculations. After the quick initial appli-

Table 2. *The measured and predicted forces of Ariolimax columbianus locomotion*
34 tests on 19 different slugs

	Stress (Pa)	
	Predicted	measured
Rim	297-659	201-512
Waves	365-1059	336-526
Interwaves	485-1222	591-1171
Overall	850-2281	780-2280 ($\bar{x} = 1493$)

cation of force the solid mucus under the interwaves should creep as the interwaves continue to be stressed, but this creep was not observed. What magnitude of creep is expected? While it is not strictly valid to equate stress relaxation with creep, stress relaxation data may provide an educated guess as to how far mucus creeps in the one second that it is stressed while under an interwave. It takes solid mucus about 100 seconds to relax to half of its initial stress value (Denny & Gosline, 1980). Consequently it may be expected that mucus would require about the same period to creep to twice its length, or a creep of $0.1 \mu\text{m/s}$ for a $10 \mu\text{m}$ mucus layer. This amount of creep is far too small to have been detected by the video tests.

Vertical crawling

These predictions for the stresses operating under a slug are valid only for slugs crawling on a horizontal surface. If the slug is crawling vertically up, the force due to its weight acts over the area of the interwaves to place an additional stress on the interwaves. For a slug walking vertically down, this stress is in direct opposition to the stress caused by the forward movement of the foot, and subtracts from the stress imposed on the interwaves. Values predicted for vertical crawling are tabulated in Table 1.

The values tabulated in Table 1 provide a range of predictions for slugs under various crawling conditions. The accuracy of these predictions is tested by comparing this range of values to the range of stresses actually present under a crawling slug.

Horizontal tests

The average overall amplitude of the stress measured under the central portion of the foot for 34 tests on 19 different slugs was 1493 Pa with a range from 780 to 2280 Pa. This is in reasonable agreement with the predicted values for overall stress amplitude which range from 830 Pa to 2281 Pa as shown in Table 2. The values for five measurements of rim stress are similarly close to the predicted values. The records obtained from three slugs had sufficiently small zero shifts to allow for the measurement of anteriorly and posteriorly directed stresses. The record of one of these tests is shown in Fig. 11. Again these values are close to those predicted by the model.

Vertical tests

Seventeen tests on nine slugs were conducted to compare the stress values between crawling horizontally and vertically up. The mean of the total stress amplitude for these horizontal tests was 1383 Pa. Each horizontal test was followed by orienting the

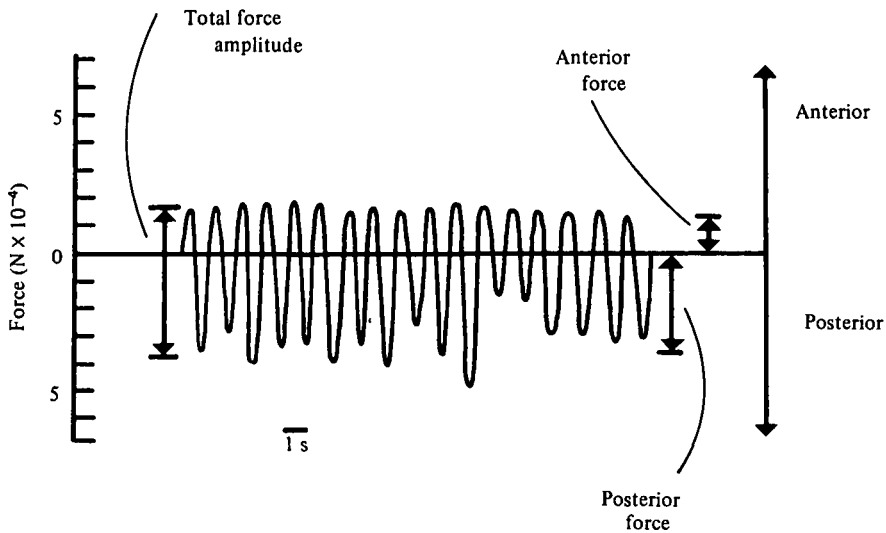


Fig. 11. An example of the record of forces measured beneath a crawling slug, with terms defined. Amplitudes are measured for each wave and are averaged for each slug.

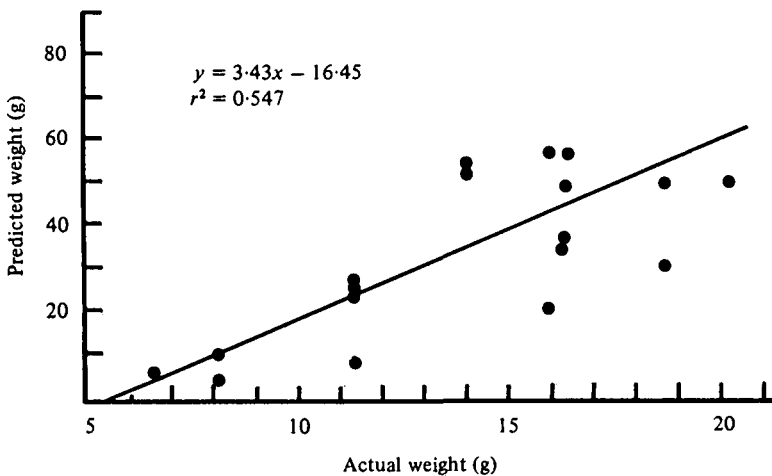


Fig. 12. A plot of weight as predicted from the model versus actual weight for all vertical crawls. While predicted weight is proportional to actual weight the slope of the relationship is greater than expected.

crawling surface vertically, and allowing the slug to crawl over the force plate again. The average value for slugs crawling vertically up was 1991 Pa, significantly greater ($P < 0.01$) than the mean for the horizontal tests when compared by a one way analysis of variance. The overall foot area of each slug was measured and multiplied by $6/15 = 0.4$ to estimate the interwave area, and each slug was weighed. The difference in stress values between walking vertically and horizontally should be equal to the force due to the weight of the slug divided by the area of the interwaves. Having obtained the difference in stress, the calculated interwave area and the slug weight, the predicted and actual values can be compared (see Fig. 12). In theory the result should

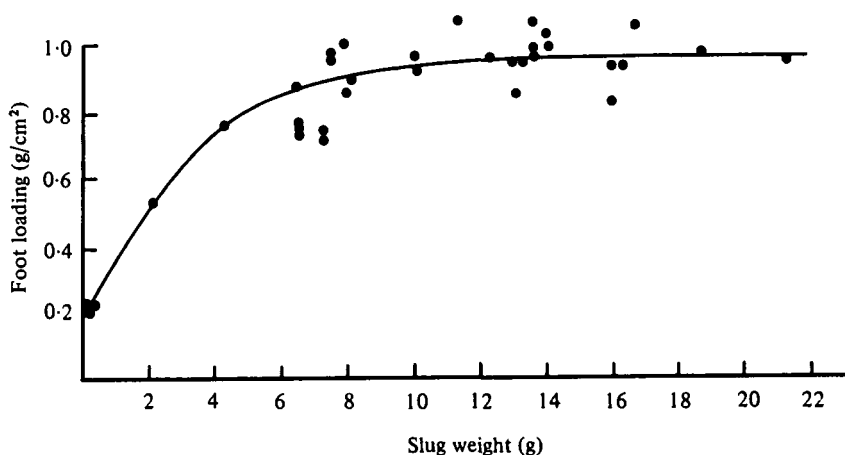


Fig. 13. The foot loading (weight/foot area) of *Ariolimax columbianus* is constant above a weight of about 5 g.

be a line passing through the origin with a slope of one. The change in stress amplitude (and therefore the predicted weight) increases as the slug's weight increases, however the change is significantly greater ($P < 0.005$) than that predicted by theory, i.e. some factor other than just the slug's weight is increasing the interwave stress required to crawl vertically. The basis for this fact is not known, though there are several possibilities.

(1) The predictions made above are for a typical slug weighing 15 g and with a foot of 15 cm². Must this model be scaled to apply to slugs of different sizes? Two factors are involved in this question. 1. The foot area determines the area of mucus over which the slug must move, and thereby the frictional resistance to movement, and if all slugs have the same shape, increases as the square of the length of the slug, L^2 . 2. For a slug crawling vertically the mass of the slug adds to the amount of stress placed on the interwaves, and for a slug of standard shape, increases as the cube of the slug's length. On this basis the foot loading (mass/foot area or L^3/L^2) is expected to increase in proportion to the slug's length, and thus would affect the stresses measured under a slug crawling vertically. Foot loading values were measured for *A. columbianus* for a variety of sizes (Fig. 13). For slugs above 5 g the foot loading remains constant at a value of about 0.95 g/cm². Evidently the slugs change shape as they grow such that weight per foot area remains constant, and the model, while calculated for a 15 g slug should directly apply to any *A. columbianus* of greater than 5 g. All slugs used in these tests weighed more than 5 g.

(2) *A. columbianus* is strongly negatively geotactic. Thus it is possible that the slugs crawl faster as a response to being tilted to a vertical position. While the speed of the slugs was not measured during these tests and this factor cannot definitely be ruled out, no visible increase in speed was noted.

(3) It is possible that the slug alters the relative areas of the various portions of the foot in response to the angle of the substratum. For the tests described above the overall foot area was measured and the various areas were assumed to occur in the

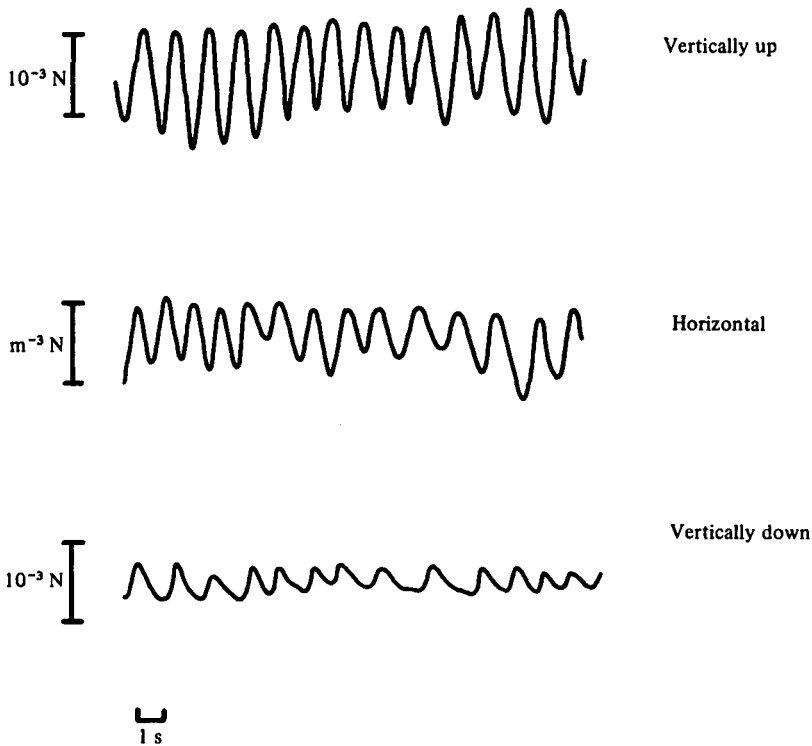


Fig. 14. A representative record of the forces measured beneath a slug when crawling vertically up, horizontally, and vertically down. The slug weighed 16.24 g and the vertically up and down records predict a weight of 34.0 and 54.4 g respectively.

average proportions observed in the kinematic study. Thus this possibility has not been tested.

(4) It is possible that the mucus layer thickness is altered when crawling vertically. Decreasing the thickness of the mucus layer would increase the force needed to move the slug upwards and thereby the interwave stress.

Further, more accurate, measurements of the various parameters of locomotion are necessary before the basis for increased stress in vertical crawling will be known.

In two tests, in addition to the slug's crawling horizontally and vertically up, the slug was induced to crawl vertically down. In this case the overall stress amplitude is predicted to be less than for horizontal crawls by an amount related to the slug's weight. Again, while qualitatively correct, the tests lead to an overestimate of the animal's weight. The stress records from one such series of tests are shown in Fig. 14.

CONCLUSIONS

These tests show the model presented here to be a reasonably accurate description of the mechanics of adhesive crawling on horizontal surfaces. The simplifying assumptions made in constructing the model would thus seem to be valid for this case.

The model is qualitatively correct but quantitatively inaccurate in predicting stresses

under slugs crawling vertically. Several possibilities for the basis of this fact have been noted, all involving changes in the parameters of the model, without altering the basic assumptions of the model. At present it appears likely that further, more accurate, measurements will reveal the basis for the model's inaccuracy, and that appropriate corrections can be made within the model's structure.

The basic principles of this model are applicable to all types of gastropod adhesive locomotion utilizing pedal waves. However, the mechanical properties of the pedal mucus for species other than *A. columbianus* have not been measured. Until these properties are known the parameters that are necessary for testing the model as it applies to the various wave types cannot be calculated.

While the model proposed here explains many aspects of the locomotion of gastropods, there is at least one aspect of *A. columbianus* locomotion for which it cannot account. If a slug is lifted off the substratum and held in mid air, it will attempt to crawl, pedal waves moving anteriorly on the foot. If chalk is dusted on the foot, the mucus layer can be seen to move posteriorly, and if a straight chalk line is drawn laterally across the foot it can be seen that the mucus in the central portion of the foot and along the extreme edges of the foot moves posteriorly faster than in the rim. It is difficult to imagine a mechanism whereby the pedal waves can account for this movement of mucus, and it seems more likely that cilia are responsible. However Litt, Wolf & Kahn (1976) have found that the effectiveness with which mucus is transported by cilia is dependent on its storage modulus. They found that mucus with a storage modulus of 1 Pa was transported most effectively by the ciliated epithelium of a frog palate, mucus with higher or lower storage moduli being transported less well. *A. columbianus* pedal mucus, with a storage modulus of 100 Pa (Denny & Gosline, 1980) would by this criterion not be effectively transported. Beyond these general observations this phenomenon has not been investigated. Until the basis for observations such as this are known, the model proposed here must be considered as only a preliminary step in explaining gastropod locomotion.

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