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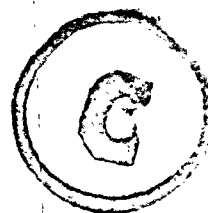
Vol. 37, No. 6

JUNE, 1960

THE INDIAN VETERINARY JOURNAL

(The Journal of the Indian Veterinary Association)

*A Monthly Record of
Veterinary Science*



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THE INDIAN VETERINARY JOURNAL

[THE JOURNAL OF THE INDIAN VETERINARY ASSOCIATION]

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A Monthly Record of Veterinary Science
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Veterinary Profession

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EDITOR

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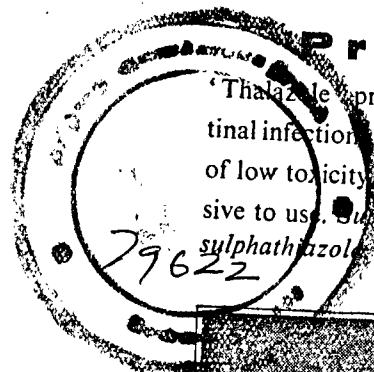
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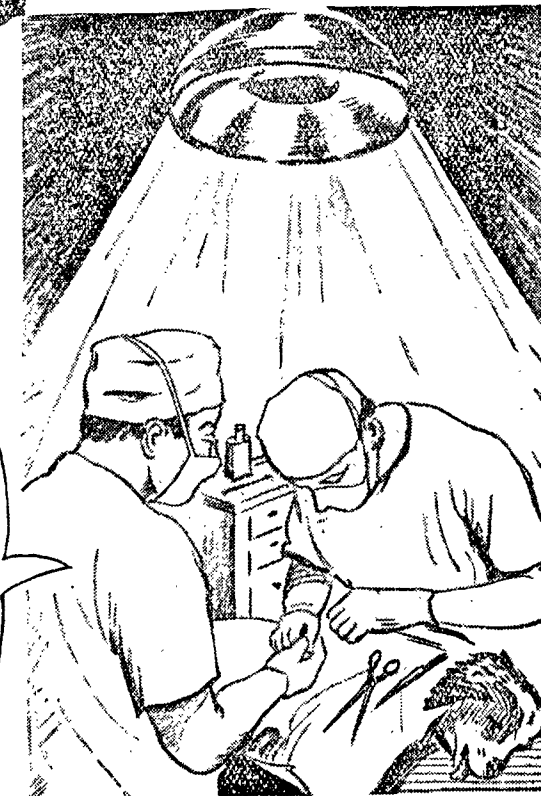
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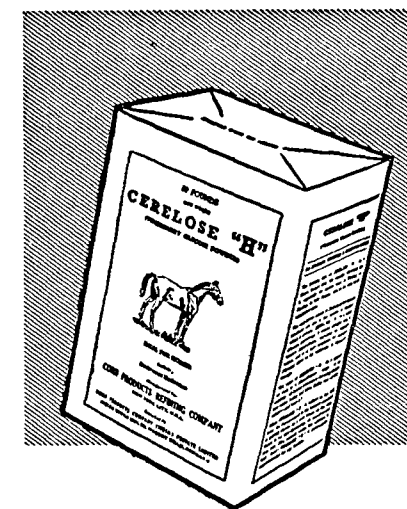
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THE Indian Veterinary Journal

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Editorial

THE INTERIM INDIAN VETERINARY COUNCIL

The above Council came into existence about four years ago and the whole profession went into ecstasy on the realisation of a long cherished ambition, though on a temporary basis to start with. Rules and Regulations were framed and representation was given to each and every State. While some of the States were quick to respond, others were absolutely indifferent and even to this day full complement of sanctioned representation is lacking on this Council. This is a very unhappy state of affairs and those who are responsible for not actively implementing the order of the Central Government on this matter deserve the condemnation of the entire profession. We say this after waiting for full four years and not in any indecent haste. In the ultimate analysis it is found that the Members of our profession in the several States are not exerting as much as they ought to, in such a matter of vital importance to the profession. We are not able to understand how we could be so blind to our own interests - both individual and collective. It is high time we realise that first things must come first. On the success of this Council, so much of the future of the profession depends. Let us make no mistake about it.

Again it is regrettable to record that the meetings of this Council are so sparsely attended. It is true the Directors of Animal Husbandry in several States are far too busy with so many meetings in these days; but the importance of the meetings of the Interim Indian Veterinary Council should be fully realised and the State Governments should be kept informed sufficiently in advance that preference will be given to such meetings. Otherwise these two factors, viz. lack of full representation of the several States and the poor attendance at the meetings, act as a drag on the progress of work of this important council.

Further a model "Veterinary Practitioners' Act" with necessary rules and regulations was prepared by this Council and circulated to be of help to the several States in drafting their Acts. But yet, many States have not yet taken up this work and we appeal to the Directors of Animal Husbandry in such States to uphold the cause of the profession without any further delay and see that the necessary Act is passed before the year is out. The Central Government are withholding the right of placing this Council on a statutory basis, because of the lack of such Councils at State level. We think it is time the Central Government allow this Interim Indian Veterinary Council to function on a statutory basis and thus hasten the States to follow suit immediately. It is arguing in a vicious cycle and it injures the interests of the profession incalculably. Such of those States in which the Act has been passed should start functioning effectively and show the way to others.

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General Articles

COMPARATIVE ANATOMY OF THE BONY SYSTEM OF THE CAMEL

(*Camelus dromedarius*)

PART II

BY

M. N. JAMDAR,

Professor of Anatomy, Veterinary College, Jabalpur (M.P.)

Bones of the Hind limb of Camel

The Os Coxae :—Both the bones are fused and the ischio-pubic symphysis is closed as in ox and horse. The tuberosity on the ventral aspect of the symphysis is present (as in the ox) but the ridge is not distinct.

The ilium :—It is comparatively shorter and wider. A curved rough line is situated in the outer quadrant of the gluteal surface near the tuber coxae and is directed downwards and slightly outwards. Laterally it meets the gluteal line which is similar to that in the ox, but it is more prominent. The bone is not so much curved (unlike in ox or horse) and consequently the gluteal face is almost flat. Ilio-pectineal line is not distinct. The psoas tubercle was found to be well developed in one specimen but was faint in the other two. The iliac crest is thicker, longer and almost straight. The lateral border is more concave and does not show the deep depression on the medial side of its acetabular end for the rectus femoris. But there is a faint pit on the lateral aspect. The medial border is less concave in its lower half. The nutrient foramen is situated on this border at the great sciatic notch. The tuber coxae does not show itself distinctly, its tuberos character being absent; it is thinner, narrower and pointed. The tuber sacrale is slightly thicker; it is separated from that of the opposite side by a wider interval than in the ox.

The ischium :—It is narrower and smaller than that of the ox. The bone meets the opposite one at a more open angle. Consequently the ischial arch is wider. The depth of the ischial arch may be said to be intermediate between that in the ox and the horse. The ischiatic spine is less in height; it is distinctly curved outwards in its posterior part. It shows distinct rough elevated vertical lines on its lateral face. The tuber ischii is not trifid; it is quadrilateral in outline with expanded ends; it resembles more the tuber coxae of the horse but is thicker and wider; its lateral angle shows a circular rough mark and

the medial one is tuberculate; ventrally there is a thick tubercular edge which may be considered as ventral ischiatic spine (as in the ox).

The pubis:—It is larger and wider than in the ox. Its pelvic surface which is comparatively more extensive, slopes downwards and forwards in its anterior part.

Note:—In one specimen a peculiar condition of the ischio-pubic symphysis was noticed on its pelvic face. Complete ossification of the symphysis on this face had not taken place. Consequently distinct wavy sutured line was evident. Anteriorly in the pubic part were two sutural lines which enclosed a $2\frac{1}{2}'' \times \frac{3}{4}''$ elliptical mass of bone and similarly posteriorly in the ischial part another mass of bone ($1\frac{1}{2}'' \times \frac{1}{2}''$) was found enclosed. In between these two masses a single median sutural line was seen. The occurrence of such masses of bone is probably akin to the presence of sutural bone found in the young skull, although the joint is a symphysis and not a suture.

The pelvis:—The pelvis of the camel for its size appears to be small. The floor of the pelvis is not so deep as found in the ox, nor is so shallow as that of the horse. The pelvic inlet is circular. The pelvic outlet is comparatively larger than that of the ox, the ischial arch being wider.

The acetabulum is wider but shallower and resembles that of the horse. Its rim is thinner which showed only one notch in all the three specimens. The acetabular fossa is shallow.

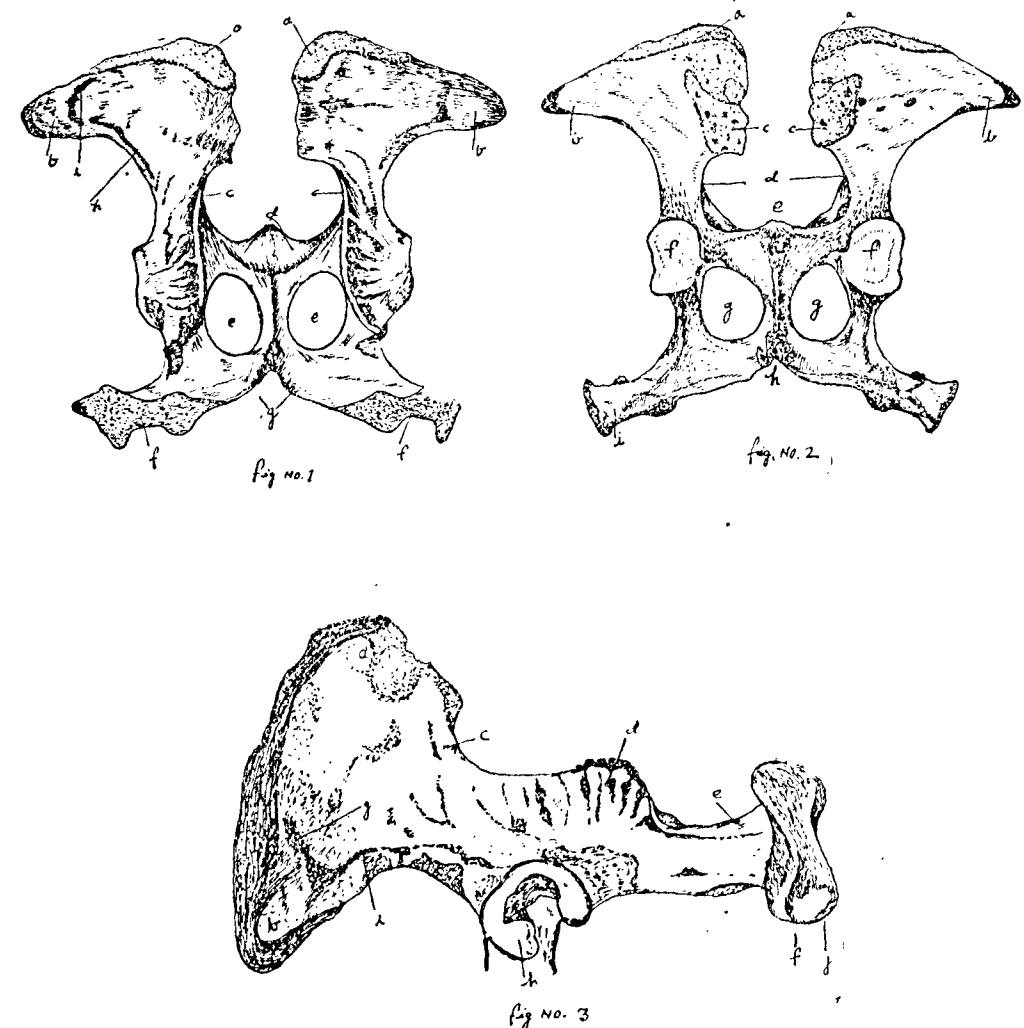
Obturator foramen:—It is oval and larger.

Femur:—It is much longer and distinctly curved particularly in its lower part. The shaft is more rounded. It is flattened antero-posteriorly being wider at the upper third. Posterior surface shows a number of rough linear lines. Trochanter minor is tuberculate as in ox and is a little larger. The nutrient foramen in all the three specimens is found about the middle of the posterior surface of the shaft. The supra-condyloid fossa may be said to be absent; instead, there is an extensive large raised tubercular area. Medial supracondyloid crest is better defined.

The proximal extremity is wider and thinner. The head is larger and round. The fovea capitis is in the form of a deeply cut elongated notch, although it is not so deep and wide as in the horse. The head projects medially and upward to such an extent as to be higher in level than the trochanter major. The trochanter major is less massive and undivided as in ox. The trochanteric fossa, although not extensive is deep. The trochanteric ridge is absent.

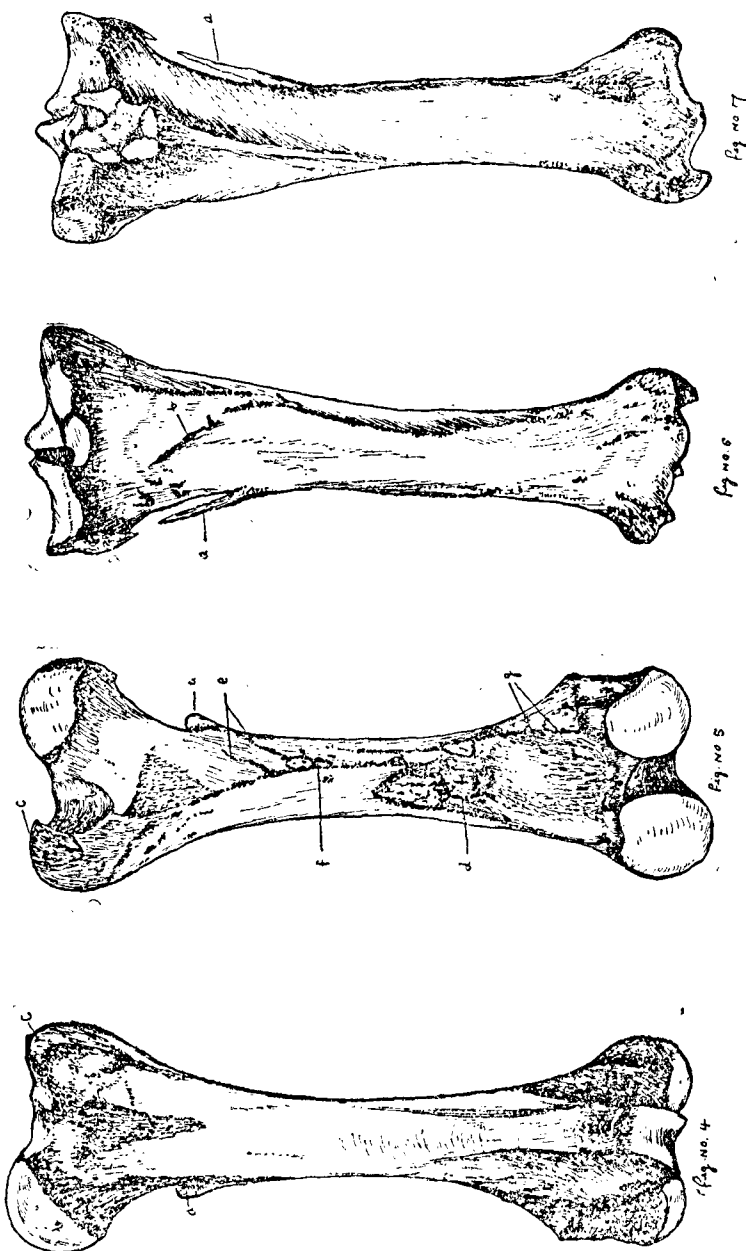
Bony System of the Camel

By
M. N. JAMDAR



Bony System of the Camel

By
M. N. JAMDAR



The distal extremity is more massive. The lateral condyle is distinctly larger. Medial epicondyle is more developed and the medial condyloid crest is thick and prominent. The trochlea is wider and the ridges are less confluent. The depression above the trochlea is large.

Patella :—It is more elongated than triangular and presents mainly two surfaces. The dorsal surface is strongly convex and rough and more regular than in ox. The articular surface is concave from above downward and does not show the ridge found in the bone of the ox. The medial long projection is much lower down and is not distinct. The articular surface at the apex is strongly convex and rounded off.

Tibia :—It is comparatively larger, being longer and wider. Tibial crest is prominent but less in length. It resembles more that of the horse. The popliteal line is very distinct and curved. The nutrient foramen is close to the lateral border as in the ox.

Proximal extremity is very massive. The lateral condyle is slightly concave and the medial one is almost flat. The lateral condyle has the proximal end of the fibula fused to it as in ox. The tuberosity of the tibia is extensive and is grooved although to a less extent than in the horse. Behind the tibial crest, this extremity shows a larger depressed area than in the horse.

The distal extremity resembles that of the ox. The articular areas are almost sagittal or very slightly oblique. There is an articular area laterally for the lateral malleolus.

Note :—(i) There was evidence in all the three specimens of the ossification of the shaft of the fibula in segments. These segmental parts of the fibula were found fused as thin rods along the lateral border of the tibia.

(ii) In one specimen the shaft of the fibula appeared to be ossified for greater part of its length leaving only a small gap of about half an inch between the upper fused extremity and the shaft.

Fibula :—Its upper end is fused with the lateral condyle of tibia (as in ox) and projects downward in the form of bony spicule. The lower extremity is separate forming the external malleolus which articulates with the tibia and resembles very much that found in the ox. The shaft of the fibula appears to undergo ossification with advancing age, remnants of which were found to a smaller extent in two specimens and to a greater extent in one.

Tarsal bones :—The tarsus closely resembles that of the ox in form and shape of the component bones. The central and fourth tarsal bones are not fused (unlike that in the ox); consequently there are six bones in the tarsus of the camel.

Tibial tarsal:—It has a typical ruminant astragalus shape being in the form of a thick quadrilateral piece. The proximal trochlear articular surface is just like the one in the ox but in the camel the lateral ridge of the trochlea is higher than the medial forming a distinct prominence. Moreover, the ridges show laterally greater articular surface. At the lower end of the lateral ridge, there is a salient bony projection extending outwards (which is absent in ox). The distal articular surface is more in the form of two condyles, medial and lateral. The lateral one is smaller of the two. This surface is traversed by two sagittal grooves and one sharp sagittal ridge. The latter is not found in ox.

Fibular tarsal: This resembles that of the ox, but is more massive. The sustentacular process is thicker and shorter and does not form a projection behind as noticed in the ox. The body is laterally much depressed. The tuber calcis is longer and thicker. The groove in front of the summit for the gastrocnemius tendon is not distinct; instead, there is a tubercle. The plantar aspect of the summit of the tuber calcis is not grooved being almost flat.

Central tarsal: (Scaphoid) is not fused to the fourth tarsal. It resembles the central component of the ox's bone but is slightly wider and the posterior upward projection is thicker and not pointed. It articulates by one small anterior facet and two slightly larger posterior facets with the fourth tarsal.

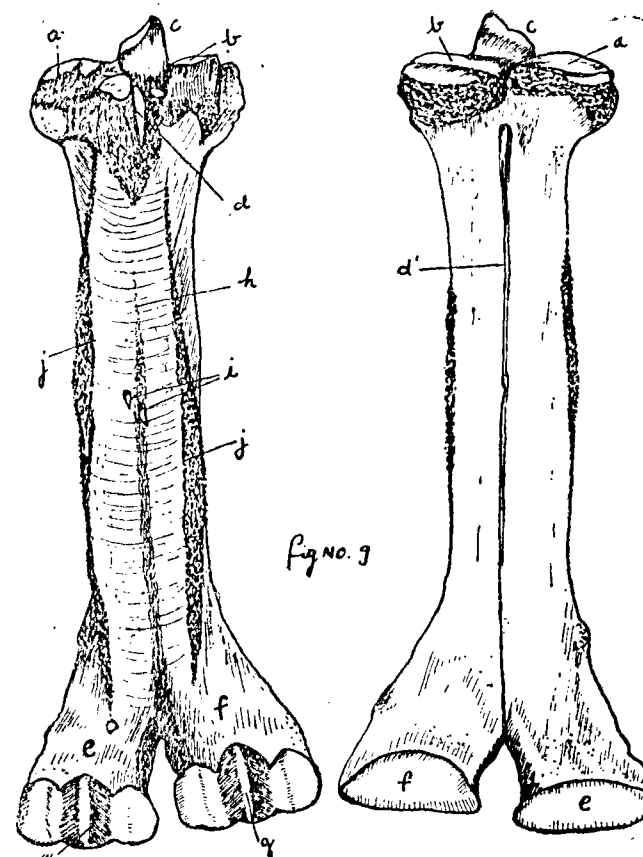
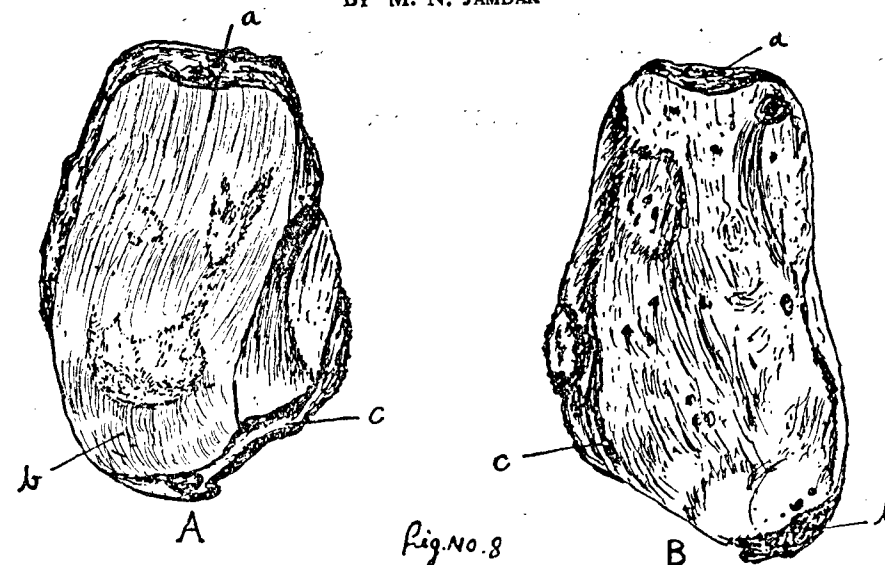
First tarsal:—It is much larger than the corresponding bone of the ox. It is in the form of a narrow, tall, four sided piece. Its anterior and posterior surfaces are narrow. The posterior face shows a large nodule. The medial and lateral surfaces are wider, rough and of equal extent. Lateral face shows an elevated rough area. Facets are similar to those in ox.

Second and third tarsals are fused to form one piece which is similar to that in ox but is much thicker (higher). It shows a large oval sloping facet on the anterior part of its lateral face, for articulation with the fourth tarsal.

Fourth tarsal: It is one third in size of the tarsal bones of the camel. It is a thick, irregularly quadrilateral with a central proximal projection. It resembles in form the fourth tarsal component of the ox's bone but for the dorsal projection referred to above. The medial concave facet on the proximal surface ascends posteriorly on the said projection. The lateral facet for the fibular tarsal is wider and does not slope so much as in the ox. On the medial surface it has three facets for the central tarsal (one anteriorly and two posteriorly) and one large irregularly oval facet (in front) for the second and third fused tarsal.

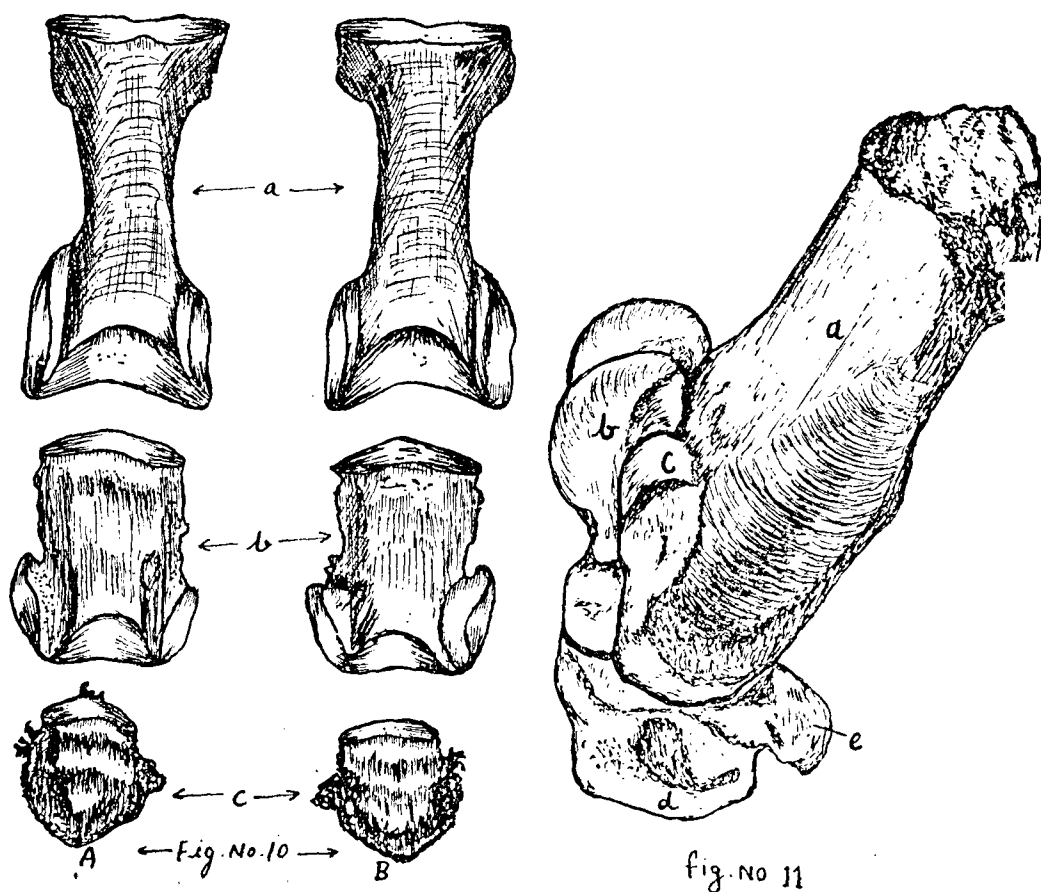
Bony System of the Camel

By M. N. JAMDAR



Bony System of the Camel

By
M. N. JAMDAR



Posteriorly the bone is narrowed down and bears a large tubercle on the inferior aspect of which is a small oval facet for articulation with a facet found on the projection on plantar aspect of the proximal extremity of the large metatarsal.

Metatarsals:—There are only two metatarsals fused to form one large metatarsal. The large metatarsal resembles very much the large metacarpal being four sided and about an inch or so longer. But the four sided character is more pronounced in the metatarsals; the shaft is more compressed from side to side so that the metatarsal looks narrower when viewed from front or behind. Lateral surfaces are slightly wider than in metacarpal. Transverse canals are absent. Vertical vascular groove on the dorsal face is slightly wider than the one in the corresponding bone of the fore limb. The plantar surface forms a narrower groove than that in the metacarpal. The collateral edges of posterior surface are thick, rough and everted; the lateral edge is thicker and more elevated. The nutrient foramina are two and are placed a little above the middle of the plantar surface one on either side of the median line but at slightly different levels.

The proximal extremity shows two large but unequal facets in front and two small facets behind. One of the small facets is placed on a bony prominent projection (directed upward) which is found on the middle of the posterior border of this extremity. This bony projection forms a salient feature of this bone. The proximal and distal extremities are distinctly narrower than those of the metacarpal. Condyles of the distal end are also correspondingly smaller. Otherwise they resemble those of the metacarpal, the medial condyle being slightly shorter and smaller than the lateral.

Digits of the hind limb:—These resemble those of the fore limb except that the first phalanges are about half an inch shorter, slender and narrower than the corresponding ones of the forelimb; the second phalanges are slender, narrower and very slightly (about one fourth inch) shorter than those in the fore limb; the third phalanges do not show any difference.

Comparison

In Camel (1) Femur is the longest bone in the hind limb, but the difference between its length and the length of tibia (2nd longest bone) is significantly greater than that between the corresponding bones of the ox and when compared to that of the horse, the difference is negligible with a slight increase in the horse. (2) Tibia is the second longest bone. (3) Large metatarsal is third in the order of length. (4) In the pelvic bone the conjugate and the vertical diameters exceed the transverse diameter—The vertical diameter being the largest. (5) The dimensions of the pelvis in the camel are almost the same as those in the ox, except for the significantly greater transverse diameter in the former. (6) The greater transverse diameter

Bony System of the Camel

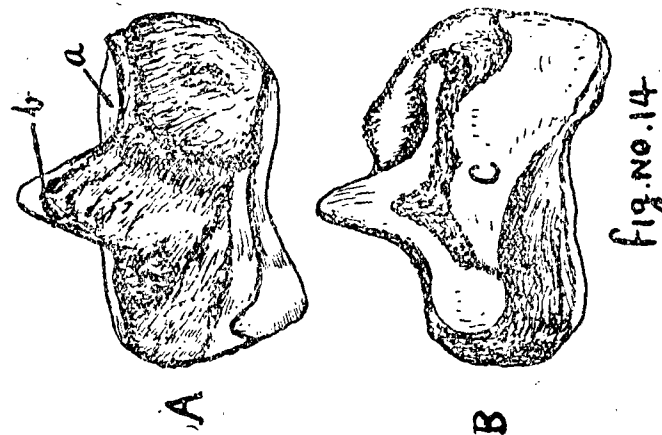
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fig. no. 14

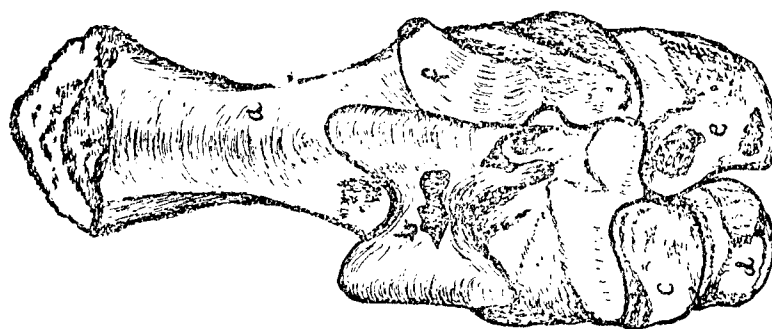


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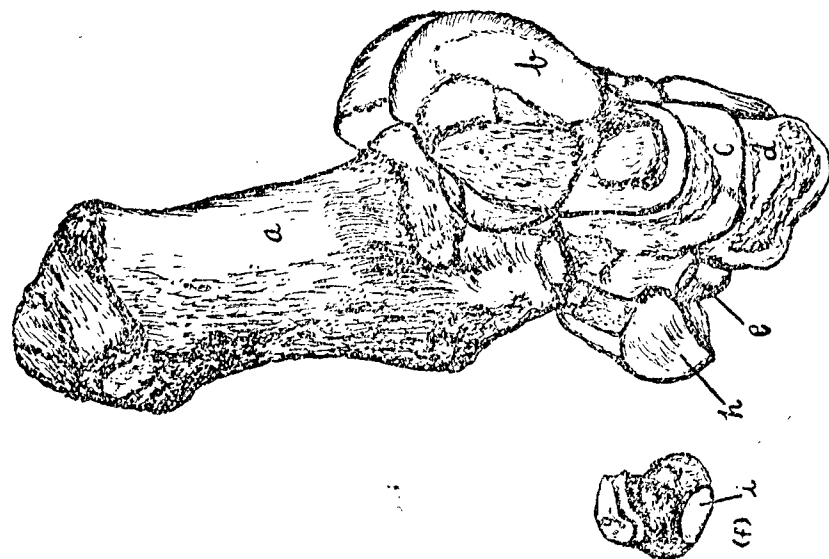


fig. no 12

in the camel is a character of the pelvis of the horse. (7) From comparison of the pelvic measurements of the pelvis of the camel and the ox, size for the size the pelvis of the camel is smaller.

In Ox (1) Longest bone in the hind limb is the femur. (2) Tibia is next in length (3) Metatarsal is smaller and third in order of length. (4) In the pelvic bone the conjugate and vertical diameters are found to exceed the transverse diameter. The conjugate diameter is the largest and the transverse diameter the smallest, except in one case where vertical diameter is the largest. (5) Width between the ischiatic spines is smaller than any of the foregoing diameters except in one case.

In Horse (1) Femur is the longest bone. (2) Tibia is next in length. (3) Large metatarsal is third in order of length. (4) In the pelvic bone transverse diameter and the vertical diameter are more than the conjugate diameter except in one case where the transverse and conjugate diameters were found to be the same. Vertical diameter, although more than the conjugate diameter is less than the transverse diameter. i.e. transverse diameter is the longest and conjugate diameter is the smallest. (5) The width between the ischiatic spines is smaller than any of the before mentioned dimensions except in one case.

Key to the Figures (Hind Limb)

- Figure No. 1. Pelvic Bone Viewed from above and behind.
 (a) Tuber Sacrale.
 (b) Tuber Coxae.
 (c) Greater Sciatic Notch.
 (d) Wide bevelled portion of the pelvic face of the pubic bones.
 (e) Tuber ischii.
 (f) Notch formed by the posterior borders of the two ischial bones.
 (g) Gluteal line.
 (h) Curved rough line.
- Figure No. 2. Pelvic Bone viewed from front and below.
 (a) Tuber Sacrale.
 (b) Tuber Coxae.
 (c) Auricular facet for sacrum.
 (d) Lateral borders of pelvic inlet.
 (e) Anterior tuberculate end of the pubic symphysis.
 (f) Acetabulum.
 (g) Obturator foramen.
 (h) Notch formed by the posterior borders of ischii.
 (i) Tuber ischii.
- Figure No. 3. Pelvic bone-left lateral view.
 (a) Tuber Coxae.
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Same as in Part I of this series.

CATIONS AND BACTERIA

By

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The antagonistic effects of cations have been recognised from the days of Ringer (1883), Loub (1900) and Flexner (1907). Since then the influence of metal ions on bacterial metabolism has formed a topic for advanced research. It has been established that apart from Na and K, most bacteria require Mg. These ions are necessary for division and growth. So much so that their sub-optimal concentrations induce either a longer lag phase or morphological changes. Apart from these, cations have been found to play important roles in respiration, pigment formation, protein assimilation and in all enzymatic reactions. Some ions act as activators, others as suppressors and still others as protectors. In the *Lactobacilli*, Mg and Mn are essential [MacLeod and Snell (1947) Svec (1948) Carlson (1952) Bard (1955) Goucher (1955)]. Ca has been found to be necessary for acid production of *Lactobacilli* [Merril (1952), Young (1944)]. Likewise *Azotobacter*, *Nitrosomonas* and *Pseudomonas* have been found to be requiring Ca. (Hellinger 1951). The compensating effect of Mg by Cobalt has

been established (Hewitt 1951). Many workers have reported compensation of Mg with Mn. Cu has been found to induce resistance by Minagawa (1955), and this resistance is attributed to the change in ribonucleic acid.

The effect of ions on phosphatases and esterases has been widely worked with individual enzymes and their specific substrates; Mg has been found essential for phosphorylating enzymes by Trevelin (1952) and Malstrom (1953). The production of A. T. P. is dependent on Mg. [Kachmar and Boyar (1953) Liebecq (1953)]. Ca has been found to suppress phosphorylations by Slater and Cleland (1952). Most of the enzymes in the Krebs cycle require specific metallic ions for activation Wolfe (1954). It has also been declared that many enzymes have metallic ions functioning as bridges between the prosthetic group and the coenzyme. The antibiotic production of Bacteria and Fungi is dependent on cations and their requirement for a particular cation is specific. Ching Tsang Tsang (1955) has worked on the kinetics of glucose breakdown and considers that the metallic ions determine the rate of breakdown. Rockford and Mendalin (1953), Webb (1953), & Maxwell and Nickel (1954) have noted the formation of long chains and filamentous forms in *Diplococcus pneumoniae*. The significance of Mg in the Gram's reaction has been described by Henry and Stracey (1946). Though literature is replete with the effect of deficiencies of various metal ions on the morphological or metabolic activities of bacteria there has not been much systematic work to assess the minimum, maximum and optimum concentration of specific metallic ions on definite species of bacteria. The reason for this has been the difficulty in developing a suitable media and de-ionising it, and for want of adequate methods to exactly determine the precise amount of ions required at very low concentrations. These difficulties were overcome in the following experiments and the study was made to assess the optimum requirements of various metallic ions for the catalase negative *Streptococcus* sp. belonging to Lancefield's classification Group N on which these tests were conducted.

Procedure

Two synthetic media were developed. Medium A and Medium B. The former contained 10 grams of Tryptone, 10 grams of Yeast extract, 5 grams of K_2HPO_4 , 5 grams of $Na_2HPO_4 \cdot 7H_2O$, 1 litre of distilled water, 2 grams of Tween 80 - pH 6.8.

Medium B: 24.4 grams yeast extract, distilled water: 1.5 litres. Tween 80: 4.5 grams.

Medium B was rendered ion free by passing it through resin KSN 159 (Bayer) in a 50 cms long 3 cms wide jacketed column kept at 32°C



Fig. 1. X 1,000

Between 12-18 gamma Mg ++/ml. level Show a tendency to form long chains. Individual cells more elliptical and smaller. Certain cells have taken the counterstain.

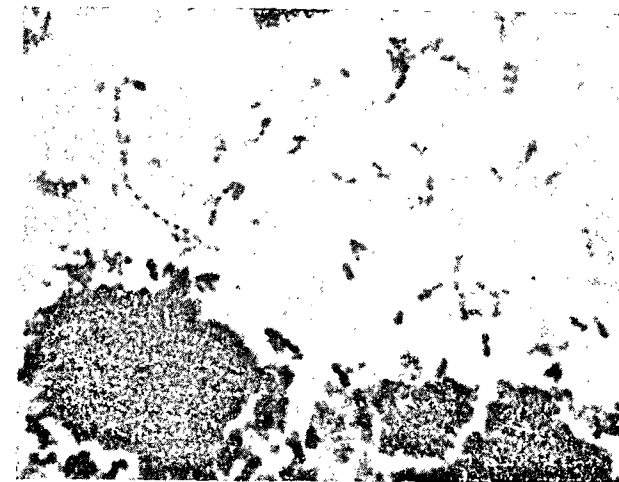


Fig. 2. X 1,000

At 10 gamma Mg. ++/ml. Single chain Shows mother cells Gram positive and daughter cells. With each division the daughter cell is smaller and takes more of the counter stain.

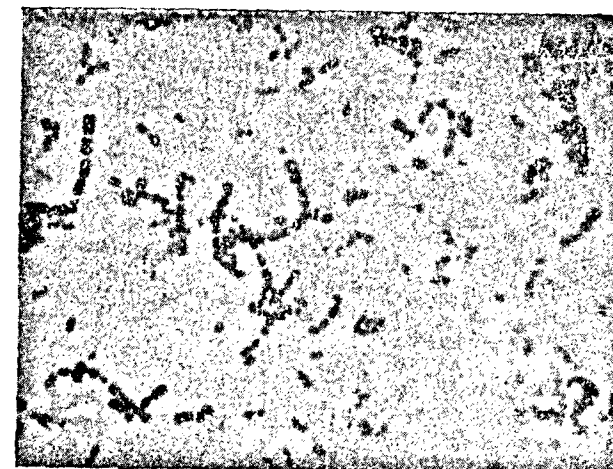


Fig. 3. X 1,000

At 9 gamma Mg. ++/ml. The chain shows a tendency towards knotting. Usual straight chains are less. Gram positive are present side by side with Gram negative chains. Individual cells in Gram negative cases are definitely smaller than the Gram positive cells.



Fig. 4. X 1,000
At 8.5 gamma Mg. ++ /ml.
the knotting is clearly seen.

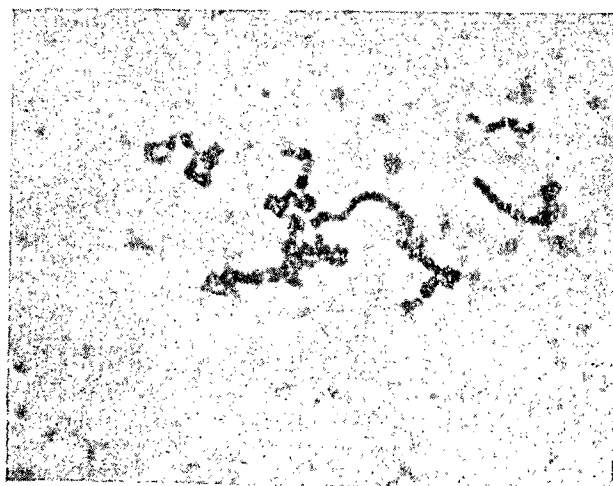


Fig. 5. X 1,000
At 8 gamma Mg. ++ /ml, the
knotting becomes still pro-
nounced.



Fig. 6. X 1,000
At 7.5 gamma Mg. ++ /ml.
the chains have become smaller.
Individual cells show a signet
ring Gram positive area; the
rest of the cell taking the
counter stain.

by circulating hot water. The medium was passed a number of times to completely de-ionise it and then standardised and sterilised. Medium B was supplemented with 3 grams of L-Lysin, L-Histidine and DL-Methionine. To both media glucose sterilised by filtration of 20% solution was added to a final concentration of 1%. The growth in Medium A was used as positive control and experiments were done in Medium B containing various concentrations of metallic ions and their growth tested after 36 hours by a spectrophotometric colorimeter at constant wave length. The metallic ions were prepared in 1,000 to 2,000 gamma concentrations and added in quantities to produce the various dilutions. All the glassware used in experiments were rendered ion free by freshly prepared $K_2Cr_2O_7$ and H_2SO_4 . The ion free media was also tested for its content of Mg and other ions before use. The first set of experiments were conducted to determine the amount of Na and K needed. Subsequently experiments were made with individual ions or with concentrations of 2 or 3 metal ions keeping Na and K levels at the optimum.

Results

Mg was found to be essential for the growth of the bacteria in addition to Na and K. The optimum concentration of Na and K was found to be 960 and 720 gamma respectively. While suboptimal concentration of Ca suppressed growth, the effect of suboptimal Na was to prolong the lag phase. With single metallic ion added, Mg at 25 gamma produced maximum growth at 72% of growth in Medium A. Higher concentrations produced suppression which was offset by the presence of any other divalent ion even in minimum concentrations. Cobalt toned up growth whereas Cu and Zn completely suppressed growth. Neither Mn or Fe or any other metal could replace Mg though Cobalt to a certain extent could compensate this ion. Addition of third bivalent ion had no visible beneficial effect.

Having determined the optimum concentrations of metallic ions necessary it was decided to test out the morphological changes that resulted from growing bacteria for 36 hours in decreasing concentrations of Mg in the presence of optimum Na and K.

The normal bacteria grown in Medium A was again taken as standard. Growth in 12-18 gamma Mg: Some parts of the chain show smaller sized organisms which are Gram negative, chain length decreased. Growth in 10 gamma: Shows cells gradually reducing in size more Gram negative shorter chains. The effect of growing bacteria on 5 gamma Mg shows cells becoming once more larger and their periphery becoming undefined. Stepwise reduction of Mg content and its effect is shown in the photographs with Mg level at 12.18 gamma, 10 gamma,

9 gamma, 8.5 gamma, 8 gamma, 7.5 gamma, 7 gamma, 6.5 gamma 6 gamma, 5 gamma, under 5 gamma and revival of cells grown at 6 gamma and subcultured in 25 gamma. These pictures show the gradual reduction in chain length after initial spurt, smaller cell size, knuckling of bacteria, the retention of a polar Gram positive signet ring, and finally a Gram positive point. This stage is followed by the cell wall rupturing and releasing the cell debris. The cells could be revived till the stage of 5 gamma but the revived cells do not assume the same morphological structure as the original till considerable number of subcultures are made at optimum Mg levels.

Oxidative reaction in cells grown at optimal levels of Mg were found to be less well marked than under higher concentrations. In the specific chain of reactions $\text{CH}_3\text{COCOOH} \rightleftharpoons \text{CH}_3\text{CHOHCHOH} \rightleftharpoons \text{CH}_3\text{COCHOH} \rightleftharpoons \text{CH}_3\text{CHOHCOOH}$, the velocity of reaction 2 to 3 was found to be higher at lower Mg concentration. This indicates the importance of metallic ions on cell surface enzymes and suggests the possibility that electron transfer takes place through absorbed cations across the cell wall.

The estimation of Mg and Ca were done by the method of Levine and Cummings (1955) by the Complexometric titration and spectrophotometric measurements which are accurate to 0.2 gamma.

Conclusion

Morphological changes induced by metallic ions particularly Mg in suboptimal concentrations indicate that certain ions are essential in optimum concentrations for morphological stability and enzyme activities, as indicated by transfer of electrons particularly cations across the bacterial cell wall.

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Fig. 7. X 1,000

At 7 gamma Mg. ++ /ml. Growth is suppressed considerably and the few short chains have no more tendency towards knotting. The signet ring shaped Gram positive area of the previous illustration is further reduced till in some cases only a Gram positive dot (polar body) is seen in the margin of the cell.

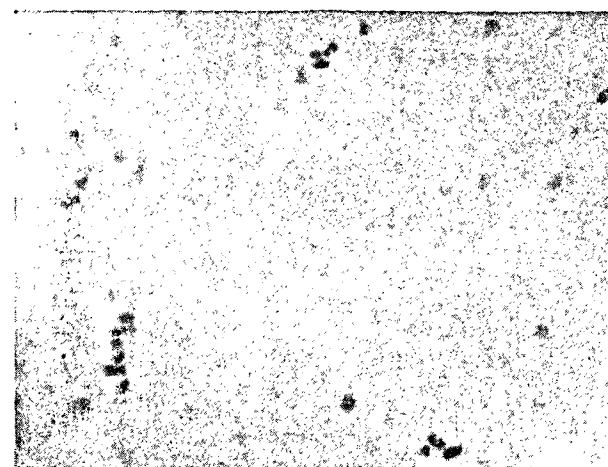


Fig. 8. X 1,000

At 6.5 gamma Mg. ++ /ml. The Gram positive dot is clearly visible.

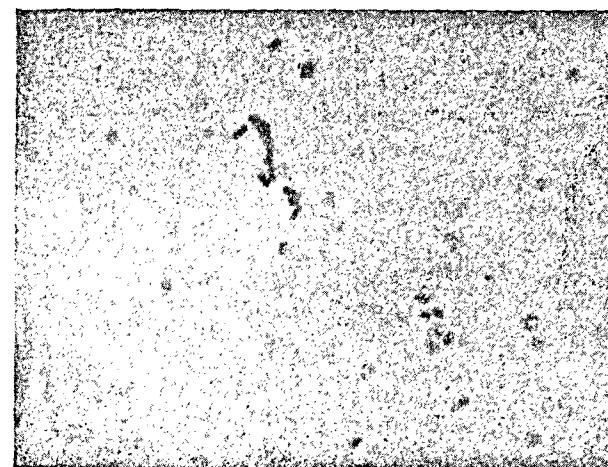


Fig. 9. X 1,000

At 6 Gamma Mg. ++ /ml. The cells have become nearly completely Gram negative and the individual cells attain the smallest size observed. Some cells reveal a hazy tendency.

CATIONS AND BACTERIA

By U. K. MENON

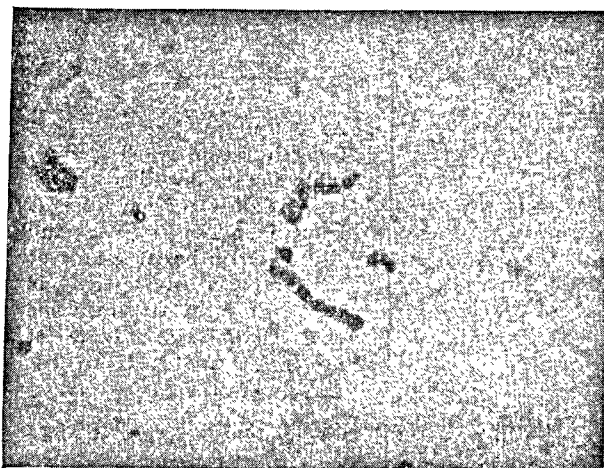


Fig. 10. X 1,000

At 5 gamma Mg. ++ /ml. The cells are slightly larger fully Gram negative and the cell wall has given way in some places making the boundary less sharply differentiable.

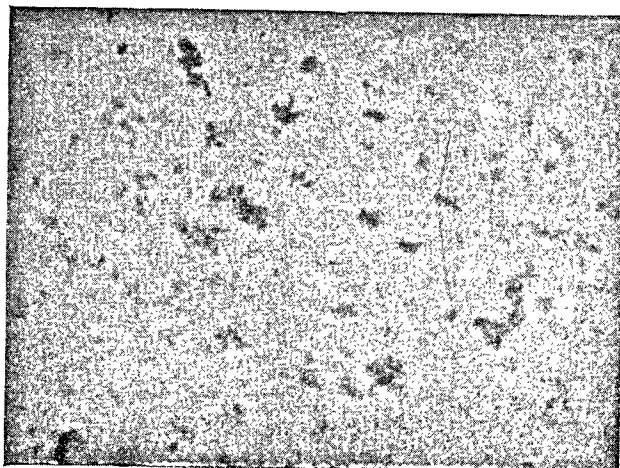


Fig. 11. X 1,000

Shows the Gram Negative cell debris.

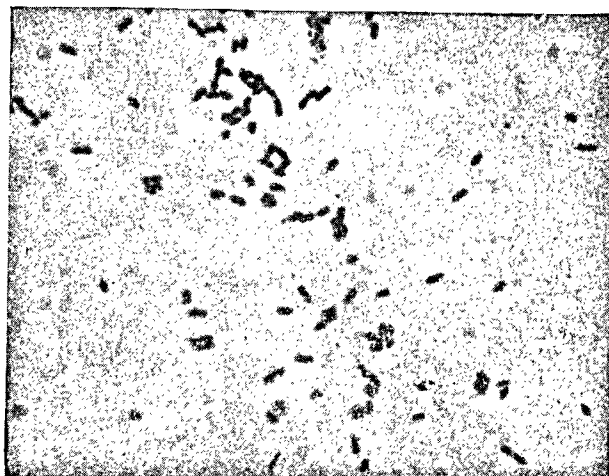


Fig. 12. X 1,000

Shows cells grown in 6 gamma Mg. ++ /ml. as subcultured for 24 hrs. and revived in 25 gamma Mg. ++ /ml. Cells have become Gram positive. Short chains and diplococci predominating. Individual cells are still smaller-Cocci-bacillary in appearance.

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CULTIVATION OF SHEEP-POX VIRUS

By

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Work done in the earlier times seems to have been influenced by the presumed inclusion of *Variola ovina* in *Variola vaccinia* group of Viruses. Justification for such a classification is now open to doubt, particularly since its distinct host specificity, and antigenic structure with no immunological relationship with *variola-vaccinia* groups of viruses have now been established.

Attempts to cultivate sheep-pox virus without adversely affecting its immunogenic property by passage through other species of animals have been made from time to time. Kasai (1929) claims that it is transmissible through goats and becomes sufficiently attenuated to be used as a safe and effective antigen. Kai and Kasai (1927) claimed that *Vaccinia* gets transmuted by continuous "Ovination" into sheep-pox, which nevertheless, constitutes a very weak antigen to be of any practical use. Gin and Kunert (1927) have asserted that even unaltered or "non-ovinated" *Vaccinia* Virus imparts solid immunity against *variola ovina* in sheep. Roa (1938) claimed to have adopted sheep-pox on chorio-allantoic membrane, of developing chick embryo, which however, failed to impart any degree of immunity against sheep-pox. Ortenzi *et al* (1954) failed to cultivate it on chorio allantoic membrane. However during the first passage there was an initial though transitory multiplication of Elementary Bodies. Bridre (1931) states that sheep-pox is transmissible to horse, rat, guineapig, doreas gazelle, and barbery sheep. Bennet, Horgan and Haseeb¹ have failed to:

1. transmit sheep-pox to animals most commonly used in pox work, viz., rabbit, monkey and calf.
2. establish any immunological relationship between the virus of sheep-pox and vaccinia or sheep-pox and goat-pox.
3. to confirm that sheep-pox can be modified by either "Caprinisation" or "Ovinisation".

Aygun (1955)¹¹ claims to have propagated sheep-pox virus in roller tube cultures of lung and skin tissue of embryo.

The author made four separate attempts to check on the possibility of adapting sheep-pox virus on other species of animals using improved method of freeing pox lesions from bacterial material by incorporation of streptomycin.

Seed Virus

Two strains of Virus maintained in 50% Glycerine saline at refrigeration, were used in these studies:

1. Laboratory strain isolated from field in the year 1945 and maintained by regular periodic passage through sheep by percutaneous inoculation.
2. Recently isolated field strain which had shown high thermal reaction and typical generalisation. The scabs were well macerated in N.S.S., to make 1:10 dilution and subjected to 15 minutes centrifugation at 3000 r.p.m. The supernatant was treated with streptomycin and tested for sterility. It was passed through sheep before being used as seed virus.

Since in the absence of any tissue reaction at the site of inoculation, the multiplication or destruction of Virus can only be ascertained by titrating back tissue suspension prepared from the site of inoculation into susceptible hosts, it was first necessary to determine the quality of virus per gram (M.I.D) of inoculum. And any decrease or increase in titre was presumed to be associated with corresponding destruction and multiplication of Viral bodies.

Titration of Virus in Suspension

Virus rich tissue harvest from a donor of 100% "Takes" was macerated into a fine emulsion in N.S.S., using Waring blender and titrated in progressive dilutions by inoculating 0.5 c.c. subcutaneously of each on a well demarked area shaved and sterilised on abdominal surface of susceptible sheep. 6 to 8 different dilutions could comfortably be titrated on a single animal with distinct isolated reaction. The surface is so divided that the area inoculated with lower concentration of virus is opposite that of higher concentration, thus avoiding any confluency of reaction. The use of progressive dilution on a single animal rules out or reduces any possibility of variation in local reaction due to individual susceptibilities. The shaved abdomen is sterilised with rectified spirit before inoculation, and kept covered with a sterilised cloth, changed daily, during the entire period of observation. At least three animals are used for each titration.

The highest dilution which precipitates well defined zone of congestion, subsequently followed by swelling, hot and painful, was considered as the M.I.D. Ulceration was not considered necessary as the presence of live virulent virus was evidenced by local reaction and pyrexial response.

It is observed that :

1. The reaction may not be visible from 4 to 6 days with dilutions less than 1/20,000 and for 6 to 8 days with higher dilutions; and that no reaction will appear after 10 days.
2. The same maximum was reached whether the concentration of the original inoculum was 1/100 or 1/160,000. As can be expected the greater the concentration of virus used, the sooner was the maximum reached.
3. The severity of the reaction did not depend upon quantity, but on the concentration of inoculum.
4. With laboratory strain maintained by percutaneous passage or freshly isolated field strain, titre of the virus with the primary passage was 1:40,000. With the third passage onwards, the titre reached the maximum of 1:80,000 and then onwards remained constant.
5. The yield of Virus rich tissue by s/c inoculation ranges from 300 to 500 grams per animal as compared with 5-7 grams with Scarification technique.
6. The titration of one batch of virus suspension constitutes the production of another batch of virus material. It is a continuous chain reaction.
7. The virus, even after the first s/c passage produces poor "Takes" when subsequently grown by scarification.

Animal Inoculation

The animals chosen were goats, rabbits, and guineapigs, for every experiment. The sites of inoculation were the back region of rabbits and guineapigs and abdominal region of goats. The site was thoroughly shaved, cleaned by scrubbing well with soap and distilled water and sterilised by application of rectified spirit or absolute alcohol. Each animal was inoculated by Scratch method on one half of the abdomen and by S/c method on the other half. Three linear scratches were made running parallel to the longitudinal axis of the body and placed equi-distant from one another. Multiple S/c inoculations were done, three for rabbits and guineapigs, and four for goats, using 0.5 ml. per inoculum of 10^{-1} dilution of viral suspension in N. S. S. treated with 10 mgm. per ml. of streptomycin and tested for sterility. The number of M.I.D.'s per gram of viral tissue was previously determined. The viral suspension was found to be infective in 1:80,000 dilutions. Therefore, the rabbits and guineapigs received 12,000 M.I.D.'s (1.5 ml. of 10^{-1} suspension) and goats 16,000 M.I.D., (2 ml. of 10^{-1} suspension) each, positively planted by S/c route. Besides an unknown number of M.I.D.'s was inoculated by scratch

method, where positive retention of viral bodies could not be guaranteed due to their accidental washing off by the exuded lymph or blood from the seat of scratch. While the rabbit and guineapig were caged without any further treatment, the inoculated abdominal areas of goat was kept covered with sterile cloth. No topical application of any bactericidal or bacteriostatic agent to the inoculated part was practised, as it had been found by practice that incorporation of streptomycin with the inoculum keeps all the subsequent tissue reactions completely free from extraneous contamination. The sterile bandages were changed everyday; and only female goats were chosen so that the infected parts can be prevented from being soiled with urine. Three female sheep were used in each experiment as control and the virus suspension was titrated on them to have check on the drop in titre, if any.

Local reaction was observed from the 4th day onwards while the temperature was recorded daily.

None of the infected goats, rabbits and guineapigs developed either local or general reaction, while all the controls manifested typical local and thermal reactions. The first rise in temperature was observed on the 4th day and the local reaction from the 6th day onwards. The highest dilution which gave local reaction was 1:80,000 thus duplicating the titration.

The inoculated rabbits, guineapigs and goats were sacrificed on the 14th day and skin at the site of inoculation was incised to an extent of $\frac{1}{2}$ " radius. The incised portions were separately macerated into a fine emulsion in N.S.S. to make a 1:10 suspension, treated with streptomycin and titrated on sheep using three sheep for each batch. For each titration two control sheep were used with sheep-pox material from donor of 100% "Takes". None of the sheep titrated with skin emulsion from rabbits, guineapigs and goats developed any lesion while all the controls manifested typical lesions.

Propagation of Sheep-Pox Virus in C.A.M.

Following the encouraging results of Goodpasture, Woodruff and Budding' (1932) who demonstrated profuse growth of vaccinia on developing chick embryo periodic attempts have been made to adopt this technique with sheep-pox virus, with the object of producing a specific, bacteria free cheap antigen of altered virulence. Roa (1938) claims to have adopted it on chorio-allantoic membrane, with luxuriant growth, but regrets having lost it in subsequent passages. Ortenzi and Tiecco (1954) were unsuccessful in their attempts to adapt it on 8 day old chick embryo. Yuan Ching-Tze *et al* (1957) succeeded in adapting the virulent Tai-yuan strain on chorio-allantoic membrane of developing chick embryo, which after 90 successive passages is reported to have become sufficiently attenuated to be used as an antigen.

Sabban (1957) has adapted the virulent Cairo S. Strain on developing chick embryo by 21 alternating passages between sheep and chick embryo reaching 10^{-5} and 10^{-6} titre respectively without loss of virulence for sheep.

Repeated attempts of the author over a period of the years have proved futile. The procedure adopted is detailed as follows :

Preparation of inoculum

1 : 10 dilution of virus suspension treated with 10 mgm. per ml. of streptomycin, and used in the transmission experiments with guinea-pigs, rabbits and goats, was further subjected to centrifugation for 15 minutes at 3,000 r.p.m. to remove any gross tissue particles which might create any traumatic injury to the chorio-allantoic membrane. The supernatant was again treated with streptomycin, and tested for the presence of virus by inoculating into the susceptible sheep.

Method of inoculation

A continuous supply of eggs was maintained from the vigorous healthy breeding flock of Government composite Livestock Farm, Hessarghatta. Only clean white shelled eggs were preferred as determination of vigour, activity, and viability of developing embryos candling was easier. The eggs were incubated at 37.5°C . and about 60% relative humidity. They were turned twice daily. Only live, vigorous and active embryos were chosen. The egg to be inoculated was thoroughly cleaned with rectified spirit or absolute alcohol, and a drop of melted paraffin was put on the area, marked just above the embryo. The shell was cut through the paraffin drop to an extent of 1×2 cm. with a dental motar and an abrasive disc, keeping the shell membrane intact. The area was again mopped with absolute alcohol and a fine opening made with a sterile needle. A counter opening was made on the air sac and membrane dropped by evacuating the air through it. The dropped condition of the membrane was confirmed by candling before inoculation. After inoculation both the openings were sealed with paraffin, and eggs transferred to incubator. Candling was done daily to determine the impaired development or death of the embryos, if any. The period of post inoculation incubation was 4 and 5 days.

7, 9, 11, 12, 14 and 16 day old eggs were inoculated with varying doses of inoculum (from 0.1 ml. to 0.5 ml. per dose) containing 80,000 M.I.D's, per ml. The post inoculation incubation temperature was 37.5°C .

No macroscopic metastatic lesions were observed other than a traumatic lesion at the place where inoculum was placed.

Membrane from the 1st four passages were macerated in N.S.S. to a fine emulsion and 1 : 10 suspension of this treated with strepto-

mycin failed to produce either a local or general reaction amongst sheep. Animals so inoculated, when subsequently challenged with virulent virus developed typical lesions.

Having failed to demonstrate the presence of virus either by macroscopic lesions on the chorio-allantoic membrane or immunity response in susceptible host, it was decided to continue egg to egg passage through chorio-allantoic membrane irrespective of demonstrable lesions or virus, and to test for the presence of virus every 10th passage. Besides absence of any tissue response both in the chorio-allantoic membrane and susceptible sheep even with the 30th passage virus, no immunity was evinced in sheep vaccinated with 1 : 10 suspension of chorio-allantoic membrane in N.S.S. at 1st, 10th, 20th and 30th passage.

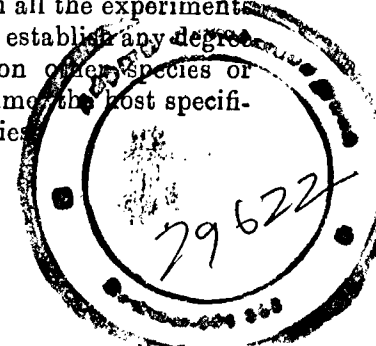
Discussion

While vaccinia has an extremely wide range of infectivity, the sheep-pox virus is practically host specific and so far all attempts to adopt it in other species of animals or on chorio-allantoic membrane have completely failed. Wherever success has been claimed, it can reasonably be presumed that the lesions so produced were due more to the presence of contaminants than the virus itself. Use of chloroform, acetone, ether or chemical dyes like Gentian violet, practised before the advent of antibiotics, was not a complete success to keep off the contaminants. And wherever suspension of Elementary bodies has been used, it has not been possible to adopt or grow this virus on any species of animals other than sheep. Roa's (1938) claim to have obtained a luxuriant growth on chorio-allantoic membrane with this virus needs serious analysis. It has immense potentialities. But it must not be forgotten that these transmission experiments were conducted in lymph Institute where intrusion of vaccinia is notoriously possible. This inference may be quite justifiable in view of his own conclusions which are briefly as follow :—

1. The egg adapted sheep-pox virus is easily transmissible to sheep.
2. The sheep vaccinated with this ovinised virus were highly susceptible to virulent strain of sheep-pox virus, but, on the other hand, were solidly immune against vaccinia.

Since the immunological relationship between two different viruses is the acid test of their identity and since in all the experiments conducted by the author, it has not been possible to establish any degree of immunity amongst sheep with virus "grown" on chorio-allantoic membrane, it is reasonable to presume the host specificity of sheep-pox strain maintained in our laboratories.

3L
KZ m2, N24
N60.37-6



Encouraging results have been reported by Yuan Ching-Tze *et al*¹ (1957) and Sabban (*loc. cit*) with certain strain of sheep-pox virus in their adaptability to chorio-allantoic membrane of developing chick embryo and their immunological behaviour. The strain maintained in our laboratories has however, failed to give any growth response on species other than sheep.

Scab has been extensively used as the source of material, and as is now recognised by workers on variola-astrim-vaccinia groups of infection, it forms a poor source of virus even when fresh. Besides it is apt to be very heavily contaminated with extraneous organisms. The scarification technique so far adopted has proved unreliable even with pure suspension of E.B. The chances of virus being washed off from the site of sacrifice, and site getting contaminated resulting in false reaction are increased. With the incorporation of antibiotic, it has been possible to get viral suspension-completely free from extraneous organism. And with s/c inoculation it has been possible not only to get a definite reproducible reaction with known strength of viral suspension, but also to evolve a technique of titrating the virus. Besides it forms a source of producing a rich quantity of virulent virus completely free from extraneous contamination.

Summary

Method of producing and titrating pure suspension of viral bodies of sheep-pox has been discussed.

The non-pathogenicity and non-adaptability of sheep-pox virus to guinea pig, rabbits, goats and chorio-allantoic membrane has been dealt with.

Acknowledgment

The author is grateful to Dr. K. S. Shetty, Director of Animal Husbandry Services in Mysore and Dr. G. A. Hardikar, Deputy Director, Mysore Serum Institute for their guidance and encouragement.

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ON A HITHERTO UNRECORDED OBSERVATION REGARDING THE SHEEP NODULAR WORM INFECTION

BY

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The species of the genus *Oesophagostomum* Molin, 1861 that have so far been specifically recorded from the Indian goats are : *O. asperum* Railliet and Henry, 1913 [Bhalerao, 1933 ; Thapar, 1956] ; *O. venulosum* (Rudolphi, 1809) Bailliet, 1885 [Bhalerao, 1932 ; Mudaliar, 1943 ; Thapar 1956 and Sinha, 1958] ; *O. columbianum* (Curtice, 1890) Stossich, 1899 [Bhalerao, 1935 ; Pande, 1942 ; Thapar, 1956 ; Sinha, 1958 and Sood, 1960].

The life cycle and pathogenesis of *O. columbianum* in sheep have been extensively studied notably, among others by Veglia (1923, 1928), Fourie (1936) and Sarles (1944) while the life history of the bovine nodular worm was worked out by Anantaraman (1942). Veglia's important work in 1923 on the sheep nodular worm was followed by the report in 1928 on oesophagostomiasis in sheep and Sarles (*loc. cit.*) described the effects of an experimental infection in this host. Essentially on such work, the information contained in the compilations of Newson (1952), Morgan and Hawkins (1953) and Lapage (1956, 1956 a) is based. The parasitic third stage larva, after penetrating the intestinal epithelium in the submucosal part, lives inside a cyst/nodule and, after undergoing the third moult, the fourth stage is reached within a week, and it subsequently returns to the lumen. Surrounded by mucus, the fourth moult is completed on the mucosal surface and in about a month's time after infection the adult stage is reached. The development of nodules of varying sizes and character in the submucosa have been attributed to a number of factors e.g., secondary bacterial infection, immunological responses etc. It has also been mentioned that the span of the larval life in the intestinal wall in lambs and in mature animals may be of a fairly long duration extending from one to three months and in the majority of cases the larvae may not emerge out of the nodules but become encapsulated and later destroyed. It has also been suggested that the larvae, after leaving the nodules, may pierce the serous coat and wandering between the muscle fibres reach the lymph nodes, liver, peritoneal cavity etc. The younger worms found lying in the intestinal lumen have been associated with a certain degree of mucosal irritation leading to diarrhoea and the adult worms, not known to attach themselves to the

mucosa, also cause production of mucus probably because of the secretion from their greatly developed cervical glands. Sarles (*loc. cit.*) reporting on several ulcerated areas near some of the worms noted lying embedded between the mucosal folds, deeply scattered red patches which were probably due to petechial haemorrhage on the mucosa.

During the present study on certain aspects of helminthiasis in Indian goat, a number of serially cut stained sections of the nodules in the submucosa revealed in its peripheral zone, in addition to the fourth larval stage, the fifth stage consisting of younger juvenile which exhibited the developing gonads, *viz.*, the ovarian and oviductal tubes, fully formed and coiled round the intestinal tract though the latter was without any lumen. (Plate 1, a). This stage was found coiled with its different regions including the oral leaf crown cut in various planes, and evidently had secondarily entered the nodule from the lumen. The mechanical damage done to the intestinal lining was distinctly visible in this case. The normally occurring fourth stage larva, however, shows its characteristically well developed buccal capsule with the oesophageal tooth and the prominently developed cervical gland. This unusual behaviour of secondary migration of the younger juveniles was also encountered in respect of the adult worms, which as described by Sarles (*loc. cit.*) were found to have penetrated rather deeply into the mucosa with portions of bodies protruding into the lumen. In some of these cases the anterior region in the mucosal part had reached as far as the muscularis mucosae while in others this part was in the lumen while the middle and hind regions were contained in the mucosa (Plate I, b).

Sectioning of such patches with the contained parts of the adult worm have yielded an interesting information about the biology of this species in relation to its adult stages which though known to penetrate at times, however, showed significant pathological changes consisting of erosion and desquamation of the surface epithelium with development of necrotic areas in the mucosal region - the marked degree of necrosis observed consisting of a cellular infiltration with a preponderance of lymphocytes and eosinophiles and accompanied by prominent presence of debris arising out of the degenerated mucosal glands. The lumen of the glands in the affected part was obliterated to a varying extent in consequence of the inflammation of the gland cells which too were infiltrated with lymphocytes.

This picture of the hitherto unrecorded secondary migration at times resorted to by both the juvenile and adult stages may be a temporary and occasional phase due to unknown factors but due to it has a bearing on the pathogenesis of the sheep nodular worm and has also to be borne in mind in the programme of therapeutic control.

ON A HITHERTO UNRECORDED OBSERVATION REGARDING THE SHEEP NODULAR WORM INFECTION

By S. M. Sood

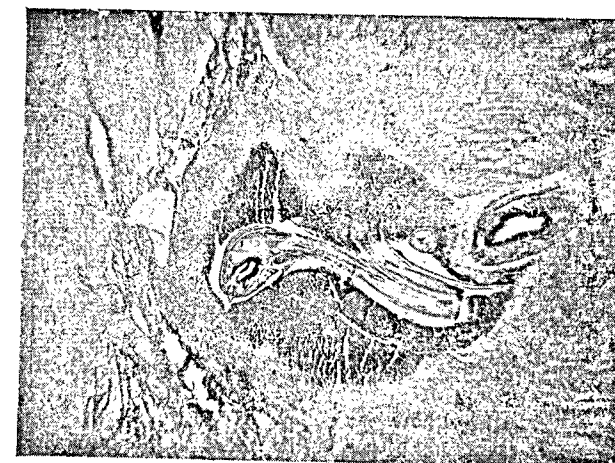


Fig. 1 (a)

Photomicrograph of a section of a submucosal nodule showing the fifth stage (younger juvenile) cut. X 25

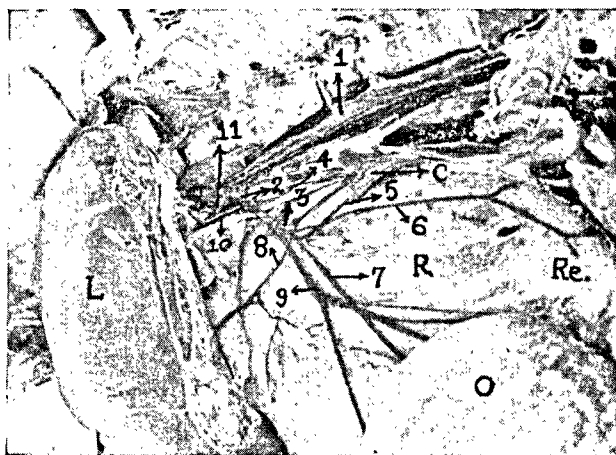


Fig. 1 (b)

Photomicrograph of a section of intestinal wall showing tunnelling and erosion in mucosal lining with a part of adult female worm cut. X 30

DEVIATION IN THE BRANCHING OF COELIAC ARTERY IN BUFFALO

By J. P. KUKRETI AND S. B. L. SAXENA



Photograph showing:

1, Aorta; 2, Coeliac artery; 3, Splenic artery; 4, Left phrenic artery; 5, Right phrenic artery; 6, Reticular artery; 7, Left ruminal artery; 8, Right ruminal artery; 9, Omaso-abomasal artery; 10, Hepatic artery; 11, Anterior mesentric artery; L, Liver; R, Rumen; Re., Reticulum; O, Omasum; C, Right Crus of diaphragm.

ANCONEUS MUSCLE IN CATTLE

By J. P. KUKRETI AND S. B. L. SAXENA



Photograph showing:

1, Anconeus muscle; 2, Long head of triceps brachii, (reflected); 3, Lateral head of triceps brachii (reflected); 4, Distal end of humerus; 5, Olecranon.

Acknowledgment

The author is greatly indebted to Professor, Dr. B. P. Pande, for his constant guidance and help in this work.

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DEVIATION IN THE BRANCHING OF COELIAC ARTERY IN A BUFFALO

By

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Introduction

The abnormality described here was noted during dissection of a buffalo in the Department of Anatomy, U. P. College of Veterinary Science and Animal Husbandry, Mathura. On referring to literature it appears that deviation in the course of coeliac artery of Buffalo has not been recorded so far.

Observations

In the present specimen of a male Buffalo aged about three years, the coeliac artery originated from the aorta and gave off the following main branches in a serial order:—

1. Left phrenic artery.
2. Hepatic artery.
3. Right ruminal artery.
4. Splenic artery.
5. Left ruminal artery.
6. Omaso-abomasal artery.

The reticular artery (*A. reticularis*) has been described in some detail here since it differed in its origin from that given in the standard text books of Anatomy. The reticular artery in this case was found to be originating from the omaso-abomasal artery (vide photograph)-6 as a medium sized vessel which continued in a forward and somewhat downward direction on the upper part of the visceral surface of the dorsal sac of rumen to the left of the cardia. Here it divided into two branches—one curved towards the left face of rumen and supplied the region there while the other passed downward and forward to reach the reticulum and at the ruminoreticular junction it again bifurcated into right and left branches. The left branch curved to reach the left face of rumen to anastomose with branches of the left ruminal artery; while the right branch ran downward to anastomose with a branch of the omaso-abomasal artery.

The left phrenic artery in the same specimen, originated directly from the coeliac artery as a delicate vessel which passed forward and somewhat dorsally to enter the left crus of the diaphragm.

The right phrenic artery, on the other hand, was found originating from the left ruminal artery about two inches from its origin. It

ran dorsally and forward under the Reticular artery over the dorsal sac of rumen nearly parallel to the left phrenic artery to supply the right crus of the diaphragm.

Discussion

Sisson and Grossman (1955) described the coeliac artery of ox as a vessel about 4 to 5 inches in length descending ventrally and forward between the rumen and pancreas on the left and the right crus of the diaphragm and the posterior venacava on the right and giving off the following five chief branches.

1. The Hepatic artery.
2. The right ruminal artery.
3. The left ruminal artery.
4. The Omaso-abomasal artery.
5. The Splenic artery.

Regarding the left ruminal artery (*A. ruminalis sinistra*), the authors described this vessel as descending on the anterior part of the right face of rumen on the anterior furrow, passing from right to left, and then continuing posteriorly to be in the left longitudinal groove and joining with the branches of right ruminal artery. It supplied mainly the anterior part of the parietal surface of rumen.

The reticular artery has been described by them as a small vessel arising from the ruminal artery about 1 to 2 inches from its origin. Further, it is stated that this vessel passes on the dorsal curvature of rumen and turns downward in the rumino-reticular groove where it continues around ventrally to the right side. It is also stated to give a branch which passes to the left of cardia and along the lesser curvature of reticulum to the neck of omasum. The branches of the reticular artery anastomose with the Omaso-abomasal and the left ruminal arteries.

Chauveau (1891) also has mentioned about the reticular artery as originating from the left ruminal artery in cattle.

It would thus be seen that the origin of the reticular artery in the present specimen differs to great extent from that what has commonly so far been described in the literature. However, no significant deviations were observed in this case in the distribution of the other branches of the coeliac axis.

Variations in the origin of phrenic arteries are frequently met with. Sissons & Grossman (1955) have mentioned variability in their origin from the aorta or coeliac or left ruminal or an intercostal or lumbar arteries. In the present case, however, in addition to variations noted so far, the two phrenic arteries took origin as the left phrenic from the coeliac and the right phrenic from the left ruminal artery.

Summary

Deviation in the origin of the reticular artery in the case of Indian buffalo has been recorded. Variations in the origin of the left and right phrenic arteries have been reported.

Acknowledgment

Our thanks are due to Sri C. V. G. Choudary, B.Sc., M.R.C.V.S., Principal, U. P. College of Veterinary Science & Animal Husbandry, Mathura for the guidance and for affording the necessary facilities.

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ANCONIUS MUSCLE IN CATTLE

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The presence of Anconeus Muscle in cattle has been described in the text books. For instance, Sisson (1955), Bradley (1946), McFadyean (1922) have described its presence in horse and Gray (1949) in human beings.

Ellenberger and Baum (1932) have considered the three heads of Triceps Muscle as the extensors of elbow joint and the flexor of shoulder joint. He has further designated the three heads as M. Anconeus longus, Anconeus lateralis and Anconeus medialis. The description of muscle Anconeus (parvus) with its attachments has been described by them to be the same as in horse.

Recently, however, Grossman (1959) has pointedly emphasised the absence of this muscle in cattle. This led us to reinvestigate the occurrence of this muscle in buffalo and ox and the presence of Anconeus muscle in these animals was confirmed. It originates in the upper third of the posterior surface of humerus and is inserted on the lateral surface of olecranon process.

Our finding confirms the presence of Anconeus Muscle in cattle and its description by authors mentioned above and is against the assertion of Grossman.

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STUDIES ON BRUCELLOSIS

By

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To eliminate Brucella abortion at the State Live-stock Farm, Chak Ganjeria, Lucknow, agglutination tests have been carried at this farm at yearly intervals. The incidence of abortions had never been heavy but a regular trickle of abortions was observed. During the course of investigation it was observed that a number of animals with history of abortion failed to show any sero-agglutinin titres against *Br. abortus* antigen. As a result of this it was found difficult to establish the cause of majority of abortions that had occurred in the herd. To find out the role of *Br. abortus* in the etiology of abortions in the herd, microscopic, cultural and serological examinations of the material from animals aborted during the year 1958 were conducted and the observations are recorded in the paper under report.

Further as many abortors failed to show seroagglutinin, and the behaviour of the non abortor reactor animals was rather unpredictable, therefore, an effort was made to make a comparative study of virulence and antibody producing behaviour of the locally isolated strain 137/23, virulent McEwen strain 544, and avirulent Cotton strain 19.

Methods and Material

Methods of husbandry:—The herd maintained at Chak Ganjeria comprises of Murrah buffaloes and Sahiwal cows which are kept in four spacious sheds with cement concrete flooring and satisfactory arrangements for cleaning and washing. The inheat females are artificially inseminated with semen from non-reactor bulls maintained on the farm premises. The expectant females are removed to confinement boxes about two months prior to the expected date of parturition. The animals

are kept for a week in the confinement box after normal calving and are then mixed up with the lactating herd. The animals that show any parturient abnormality are segregated till they are apparently cured. The aborted animals are kept in segregation for a month.

Technique of agglutination test :—Sera from all the animals in the herd was tested by tube agglutination test with the antigen issued by Indian Veterinary Research Institute, Mukteshwer. The readings were taken after 28 to 48 hours of incubation at 37°C. Sera giving a titre of 1/40 and above were considered as positive.

Isolation of *Br. abortus* :—The placental cotyledons of the aborted cows were collected in sterile glass containers and brought to laboratory immediately or were kept in refrigerator at 4°C till they could be brought for cultural examination. The cotyledons were triturated with sterile sand and suspended in sterile normal saline. Of the suspension 0.02 c.c. was spread on the surface of 1 : 1,00,000 Gentian violet liver agar plates. The plates were examined after three days incubation at 37°C in an atmosphere of 10% Carbon dioxide. The colonies were then picked up for further examination. The plates in which no *Brucella* like growth appeared were further incubated for 3 days before finally discarding them.

McEwen strain 544 and Cotton strain 19 were kindly supplied by the Head of Division of Pathology and Bacteriology, Indian Veterinary Research Institute, Mukteshwer.

Preparation of suspension of *Br. abortus* for inoculation.

Cultures of *Br. abortus*, McEwen strain 544, Cotton strain 19 and local strain 137/23, were grown separately on liver infusion agar for 3 days in an atmosphere of 10% CO_2 . The growth was suspended in sterile normal saline. The opacity of the suspensions was adjusted to No. 1 of the Brown's scale. Then 0.5 c.c. of a 10^{-5} dilution of these suspensions containing about 10,000 organisms was made and injected subcutaneously in male non reactor guinea pigs. Inoculum was used immediately after preparation. Two guinea pigs were used for each different strain.

The guinea pigs were destroyed about six weeks after inoculation. Blood was collected for agglutination test. Spleen was weighed and triturated with sterile sand and suspended in 10 times its w/v of sterile normal saline and 0.02 c.c. of this suspension was spread on 1 : 1,00,000 Gentian violet liver agar for isolation of the organism. The serum titre of the guinea pigs was studied by tube agglutination method.

Observations

The herd statistics had been changing annually as during this period many of the uneconomical animals were culled. Since November, 1956, the agglutination test of the whole herd was conducted thrice.

TABLE I
Showing reactor and aborter animals in the herd.

Time when the test was conducted	No. of animals tested	No. of reactors		No. of aborters	
		Cows	Buffaloes	Cows	Buffaloes
Nov. 1956	731	3	5	—	1
Dec. 1957	521	8	20	3	4
Dec. 1958	462	7	11	1	1

During the first test 1.1% of the animals in the herd were reactors and 12.5% of the reactors aborted. During the second test there were 5.3% of the animals which showed positive agglutination test and 25% of the reactors aborted. In the test conducted in December, 1958, 4% of the animals were reactors and out of them 11.1% aborted. The rate of abortion among the reactors in the total herd strength was 0.13% during the first test, 1.3% during the second test, and 0.43% during the third test. Generally the uneconomical *Brucella* reactors were culled and a few economical reactors were segregated. The segregated reactor animals were not included in the subsequent tests.

During the period under report a constant trickle of abortions was also observed among the animals which showed serum titre less than 1 : 20 during these three tests.

TABLE II
Showing abortions among the non-reactors.

Time when the test was conducted	No. of animals tested	No. of non-reactors	No. of abortions among non-reactors		Total
			Cows	Buffaloes	
Nov. 1956	731	723	5	3	8
Dec. 1957	521	493	3	14	17
Dec. 1958	462	444	—	3	3

The rate of abortions among the non-reactors in the total herd strength was 1.09% during the first study, 3.2% during the second study and 0.64% during the third study. Apparently the incidence of non-reactor aborters was higher in the herd as compared with the reactor aborters. The history of the animals which aborted since August, 1955 to 1958 is given in Table III. The history of some of the cows and buffaloes which had been removed earlier could not be available and has not been included in the table.

TABLE III
Giving histories and showing results of Agglutination Tests in aborted animals.

S. No.	Brand No.	Species	Date of Birth	Pregnancy when aborted	Date of abortion	Age of foetus		Results of Agglutination tests		
						Month	Days	Aug. 56	Nov. 57	Dec. 58
1	225	Bovine	13-8-48	IV	30-8-55	6	5	—	1/20 ±	—
2	18/31	Bovine	12-9-51	I	20-11-55	5	25	—	—	—
3	46/40	Bovine	5-12-50	III	19-1-56	4	0	—	—	—
4	Heroine	Bovine	13-8-44	VI	30-1-56	5	8	—	—	—
5	189/14	Bovine	13-8-54	I	17-8-57	6	5	—	—	—
6	Murli	Bovine	21-7-47	VI	5-4-57	6	23	—	—	—
7	25/20	Bovine	26-6-50	III	1-9-57	4	12	—	—	—
8	118/30	Bovine	20-8-50	V	11-1-58	7	11	—	—	—
9	62/42	Bovine	24-10-52	II	8-3-58	7	19	—	1:80	1:80
10	105F	Bovine	1-1-49	VII	8-3-58	7	10	—	1/10	1/80
11	189/14	Bovine	13-8-54	II	12-11-58	6	0	—	1/80 +	—
12	242	Buff	25-11-41	VIII	26-8-56	7	24	—	—	—
13	277	Buff	31-3-43	VIII	12-5-56	5	2	—	—	—
14	288	Buff	27-10-42	X	2-8-56	7	18	—	—	—
15	46	Buff	30-9-40	X	15-12-56	6	0	—	—	—
16	(92)	Buff	10-10-44	VII	15-4-57	6	5	—	+	Not done? Removed
17	151	Buff	15-10-41	XII	22-4-57	4	1	—	—	—
18	161/42	Buff	21-10-52	II	25-4-57	7	28	—	—	—
19	43	Buff	10-10-42	X	14-6-57	8	25	—	—	—
20	83/49	Buff	29-11-49	IV	25-6-57	7	21	—	+	Removed
21	300	Buff	31-8-44	X	20-6-57	7	21	—	1:80	Removed
22	283	Buff	31-10-14	VII	15-9-57	8	9	—	—	—
23	70/80	Buff	12-9-50	IV	15-9-57	8	9	—	—	—
24	104	Buff	25-6-43	IX	19-9-57	8	0	—	—	—
25	157/32	Buff	5-9-52	II	30-10-57	6	2	—	—	—
26	158/33	Buff	29-9-53	II	11-11-57	8	7	—	—	—
27	171/34	Buff	30-7-54	I	11-7-57	7	0	—	—	—
28	38	Buff	30-9-41	XII	20-11-57	6	21	—	—	—
29	107/30	Buff	20-9-50	IV	20-11-57	8	15	—	—	—
30	170/34	Buff	28-4-54	V	10-5-58	4	19	—	—	—
31	324	Buff	31-3-43	X	28-4-58	6	29	—	+	1/20
					15-5-58	3	26	—	—	—

TABLE IV
Showing the results of serological and cultural examination of material from aborted animals.

Sl. No.	Brand No. of aborted animals.	Pregnancy	Date of abortion	Age of foetus		Date of test	Serum titre of the animal	Results of bacteriological examination.
				Months	Days			
1	62/42 Cow	II	3- 3-58	7	9	17-11-58	1 : 160	Not conducted
2	105F Cow	VII	3- 3-58	7	0	17-11-58	1 : 80	Not conducted
3	170/34 Buff	I	28- 4-58	6	29	16- 1-59	1/20	Cotyledon smears at the time of abortion positive for Brucella.
4	47/48 Buff	VII	9- 8-58	8	0	12- 6-59	1/10	Not conducted
5	107/30 Buff	IV	9- 8-58	7	9	12- 6-59	1/40	Not conducted
6	61/29 Buff	VI	27- 9-58	6	29	17-11-58	1/80	Cotyledon smears at the time of abortion positive for Brucella.
7	189/14 Cow	II	12-11-58	6	0	12-11-58	—	—
8	137/23 Cow	IV	26- 1-59	6	12	16- 1-59	1/320	Brucella isolated
9	99 Cow	VII	1- 3-59	5	11	8- 3-59	1/80	Brucella isolated
10	180/44 Buff	I	9- 4-59	6	0	9- 4-59	1/160	Brucella isolated from foetal stomach
11	237/45 Buff	I	8- 5-59	6	0	8- 5-59	1 : 1280	Brucella isolated
12	172 Cow	IV	13- 3-59	6	0	13- 3-59	1 : 320	Brucella isolated

It will be seen that the abortions among cows and buffaloes were generally after the completion of 4th month of pregnancy and extended to the 9th month. The abortions occurred during different pregnancies and in the different age groups. Although, agglutination tests were conducted at about yearly intervals, many of the aborters failed to show a positive serum titre. Apparently the aborted animals, the placenta or the foetus did not give indication of any other infection. Therefore, during the year 1958 it was decided to conduct microscopic, serological and cultural examination of materials from the aborted cows.

* Agglutination tests of buffalo 47/48 and 107/30 could not be conducted just after abortion.

Some of the animals described above had been tested previously on more than one occasion and their relevant history is given below.

(a) Cow 62/42 was served on 14-7-57, had a serum titre of 1 : 10 on 18-11-57, was grossly positive in February, 1958 and aborted on 3-3-58 a foetus 7 months and 9 days old.

(b) Cow 137/23 had a serum titre of 1 : 10 on 17-11-57 which was 1 : 40 (not end titre) in February, 1958. On 16-1-59 it had a serum titre of 1 : 320 and aborted on 26-1-59. Cultures of *Brucella* were isolated from the cotyledons.

(c) Cow 99 was born on 4-6-47 and aborted during its third pregnancy on 5-6-54. The cow did not exhibit positive serum titre in the tests conducted in Feb. 1957 or in Dec. 1958. It aborted again on 1-3-1959 during its seventh pregnancy. The cow showed a serum titre of 1 : 80 and *Brucella* organisms were isolated from the cotyledons.

(d) Murrah heifer 180/44 aborted on 9-4-59 during its first pregnancy. The heifer had not exhibited positive serum titres in the test conducted in March 1958. The organisms could not be isolated from the cotyledons of placenta due to advanced putrefaction but pure cultures of *Br. abortus* were isolated from the foetal abomasal fluid. The serum titre of the heifer was 1 : 160.

(e) Murrah heifer 237/45 aborted on 3-5-59 and had a serum titre of 1 : 1280 on that day.

Morphology of the organisms: The organism isolated from the cows and buffaloes formed minute, round, raised, entire, glistening, convex, translucent colonies on liver agar after 48 hours of incubation in an atmosphere of 10% CO_2 . The organism failed to grow aerobically.

Microscopic examination of the culture smears revealed short, slender bacilli with parallel borders and rounded ends. Small coccoid and oval forms were also present lying singly or in chains of two to

four. The organism gave cocco-bacillary appearance and was Gram negative. It stained red with modified Ziehl Neelsen's method.

The organism grew in the presence of 1 : 1,00,000 Gentian violet and 1 : 60,000 Basic fuchsin, but failed to grow in 1 : 50,000 Thionine liver agar. It did not ferment mannitol, raffinose, salicin, sorbitol, aesculin, glucose, maltose, lactose, sucrose or inulin. The organism was catalase +, Indole - and reduced nitrate to nitrite. Hydrogen sulphide was produced for three days.

Pathogenicity for guinea pigs:

The virulent *Br. abortus* is known to be generally excreted in the milk and also to produce high sero-agglutinin titres in the infected animals which are maintained for quite long duration.

The incidence of Brucellosis as observed in the herd under report was quite low among the reactors. The organism was being excreted in the milk of 2 out of the sixteen examined reactors, (Lall & Bakshi 1959). A perusal of Table IV also indicates that the sero agglutinin titres were not very high. Therefore, a study on the comparative virulence and antibody producing behaviour of the locally isolated strain 137/23, McEwen strain 544 and Cotton strain 19 was made. About 10,000 organisms of the particular strain were inoculated subcutaneously in guineapig on the left thigh. Due to certain limitations only two guineapigs could be used for each strain. Daily morning and evening temperature of the guineapigs was recorded.

Guineapigs No. 20 & 21 inoculated with Cotton strain 19:

Guineapigs No. 20 & 21 were kept under observation for a period of 45 days. The highest temperature of 103.6°F was recorded in guineapig no. 21 on 25th day. Both the guinea pigs remained active during the period of observation.

Guineapigs No. 22 & 23 inoculated with McEwen strain 544:

Guineapigs No. 22 & 23 were kept under observation for 51 days. The first rise of temperature to 104°F was recorded in guineapig No. 22 on the 7th day and 103.6°F in guineapig No. 23 on 25th day.

Guineapigs No. 26 and 27 inoculated with local strain 137/23:

Guineapig No. 26 showed a temperature of 104.2°F on the 19th day and guineapig No. 27 a temperature of 103.6°F on the 18th day.

All the guineapigs were destroyed by severing the neck and bleeding. No gross lesions were observed in the internal organs of any of the guineapig. There was no perceptible swelling of spleen or testis. No *Brucella* organisms could be isolated from the spleen of any of the guineapig.

The blood was collected from the blood vessels of the neck after severing it for determining the sero-agglutinin titres of the guinea pigs.

TABLE V
Showing the serum titres of the guineapigs.

Organisms inoculated	No. of guinea pig	Serum titre
Cotton strain 19	20	1/10
	21	1/40
McEwen strain 544	22	1/320
	23	1/160
Local strain 137/23	26	1/40
	27	1/320

Higher serum titres were recorded in guinea pigs inoculated with the known virulent McEwen strain 544 and local strain 137/23 as compared with Cotton strain 19. One guinea pig of each group inoculated with McEwen strain 544 and local strain 137/23 had a serum titre of 1 : 320. The guinea pigs no. 20 & 21 inoculated with strain 19 had a titre of 1 : 10 and 1 : 40 respectively.

Discussion

The general procedure for the detection *Brucella abortus* infection in the animals has been the determination of sero-agglutinin titre. In India animals showing a serum titre of 1 : 40 and above against the standard antigen issued by Indian Veterinary Research Institute is considered to be positive for the infection. Various workers have recorded certain discrepancies in the results of agglutination tests. Polding (1948) reported that in the indigenous herds of U.P., *Brucella* reactors were about 5% and the incidence of clinical abortions was about 1%. In 8 farms situated in or close to Himalayas and in which the abortion was endemic he found the incidence of *Brucella* reactors in 212 non-aborting cows to be approximately 20% and in 118 aborting cows from the same herd as 51%. In the study under report it was observed, that a fairly large number of animals, that had aborted during the year 1956-57, failed to show any significant sero-agglutinin titre at the subsequent agglutination test. But during 1958-59 when the serological, microscopic and cultural examination of the aborted cases were conducted just after abortion, it was observed that out of the twelve animals examined only one cow No. 189/14 failed to show a positive serum titre and no *Brucella* organisms could be demonstrated microscopically or culturally. Buffaloes no. 170/34 and 47/48 which were tested about 9 to 10 months after the abortion showed a residual titre of 1/20 and 1/10 respectively. In buffalo no. 237/45 a serum titre of 1 : 1280 was observed just on the

day of abortion. The observations as recorded in Table IV are suggestive that a major percentage of the abortions in the herd occurred due to *Br. abortus* infection. In the non-reacting aborters one of the possibilities is that infection might have been eliminated from the system after abortion resulting into receding of the serum titres to normal. Beach (1939) reported that 28% of the two herds artificially infected with Bang's disease recovered as shown by the fact that blood titres returned to normal. Doyle (1935) recorded in a cow, falling of serum titre from 1 : 2000 to 1 : 80 within a month. The cow was destroyed and no *Br. abortus* could be isolated from the different organs and lymphatics of this cow. His findings were also suggestive that the chances of recovery of *Brucella* organisms from the organs and lymphatics of animals showing descending serum titres were comparatively less than from animals in which the titres were rising. In the observations under report it is likely that sero-agglutinin titres of some aborters might have receded due to elimination of infection from the system. At the same time the possibility of existence of infection in the system of the animal without manifestation of sero-agglutinin titres or any other symptoms suggestive of this infection, cannot be overlooked. Lamalier (1938) observed that agglutination titre increased as the abortion event approached and the maximum reactions were reached during the second fortnight after the premature birth. The reaction then decreased and sometimes disappeared. The agglutinins again began to augment at the fourth month of gestation when these cows were rebred. The observations indicate the possibility of existence of a period in which the non-pregnant or recently conceived infected animals may also be shedding organisms during this period and thus be spreading the contagion to healthy animals. Hayes and Berger (1923), Carpenter (1926), King (1928), Lourens (1931), Caldwell, *et al* (1934), Doyle (1936) isolated *Br. abortus* from the milk of cows which had negative blood titres against *Brucella* antigen. Hayes & Berger (1935) reported that the cows might shed *Br. abortus* from the udder either before agglutinins appeared in the blood or at a time when the blood agglutinins had receded to negative. Therefore, the normal agglutinin titres of the aborted animals tested a few months after abortion cannot be taken as a conclusive evidence against the abortion being due to Brucellosis. Such non-reactor aborters ought to be looked upon with suspicion and to be kept segregated from the general herd. The infected animals are also likely to give birth to full term calves in spite of the presence of active uterine infection, (Goode *et al* 1957). Gregory (1958) reported isolation of *Br. abortus* from the blood samples and udder of a cow with a negative serum titre. The titre gradually increased and cow aborted a few weeks later. Cow 62/42 and 137/23 had shown similar rising titres before abortion. Therefore, it is felt that for detecting and controlling

the disease in badly infected herds the animals should be tested at regular and shorter intervals. The sera of aborted animals should be tested within 4 to 6 weeks of abortion. The microscopic, cultural and biological examinations of the material from the aborted cows may serve as a good adjunct to the serological tests.

Ropp (1945) recorded a mean agglutination titre of 1:5,120 in guinea pigs after 9 weeks of inoculation of 300 viable McEwen strain (544) organisms. A mean agglutination titre of 1:1280 was recorded after six weeks and 1:10,000 after nine weeks in guinea pigs inoculated with three million viable organisms. The end serum titres recorded in the study under report after six weeks of inoculation of two guinea pigs with 10,000 McEwen strain 544 organisms each, were 1:320 and 1:160 respectively. These serum titres were not comparable with the results reported by Ropp (*loc. cit.*). But the local strain 137/23 was almost similar in agglutinogenic behaviour. Two guinea pigs, which were inoculated subcutaneously with 10,000 organisms each, developed serum titres of 1:320 and 1:40 respectively. Two guinea pigs inoculated with a similar dose of Cotton strain 19 showed serum titre of 1:40 and 1:10 respectively after 6 weeks of inoculation.

Although, Ropp (*loc. cit.*), after intramuscular inoculation of 300 viable organisms of strain 544 reported a recovery of 100,000 organisms per gm of spleen six weeks after inoculation, the authors failed to recover *Brucella* organisms from all the guinea pigs inoculated with different strain. Doyle (1936) could not isolate *Brucella* organisms from guinea pigs which had developed serum titres of 1:25 and 1:200 (not end titre) forty four days after the inoculation of infected milk. It is likely that even the pathogenic strains of *Br. abortus* may be eliminated from the system of guinea pigs before 6 weeks.

Summary

1. A fairly large number of animals with a past history of abortion had failed to show positive sero-agglutinin titres against *Brucella* antigen.

2. Microscopic, cultural and serological examination of the animals conducted at the time of abortion revealed that most of the abortions were due to *Brucella* infection. Ascending serum titres were recorded in some of the animals which later aborted.

3. McEwen strain 544, local strain 137/23 and Cotton Strain 19 could not be isolated six weeks after inoculation from the spleen of guinea pigs, inoculated with 10,000 organisms subcutaneously;

4. The agglutinogenic behaviour of the local strain 137/23 was almost similar to McEwen strain 544.

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THE CHEMICAL COMPOSITION AND NUTRITIVE VALUE OF GREEN MANURE CROP

Leucaena glauca, Benth

By

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Leucaena glauca (Horse tamarind) is a green manure cum fodder tree which is a native of Tropical America. It is a quick growing variety which attains a height of 10 to 12 feet in three years. It has been found established in almost all the districts of Madras state. It is also a good hedge plant which responds to pruning.

Botanical Description :—It is a deep rooted shrub belonging to Mimisoidae family growing into small trees. Leaflets are thin and they are 20 to 30 in number. The plant fruits profusely. The pods are straight, flattened at the sides, 5 to 6 inches long and about half an inch broad. Each pod contains on an average 15 to 20 seeds which are dark brown in colour and covered by a hard shining testa.

Propagation :—They are sown around its vicinity by the sudden opening of the mature pods. Seeds can be sown direct or in the nursery and then transplanted. Heat treatment between 70-80°C for five minutes increases germination rate.

Experiment :—In order to study the nutrient composition of the pods and seeds of *Leucaena glauca*, it was grown in the experimental plots of Madras Veterinary College. The mature and dry pods were collected during three different periods and subjected to a study of ratio of pod cover to seed by weight; and chemical analysis for its nutrient content. The ratio of seeds to pod cover by weight was found to be of the order of 4 : 2 on an average. The results of chemical analysis of the pod cover and the seed are shown in Table I.

TABLE I

Constituents	Expressed as % on Fresh matter basis	
	Seeds	Pod Cover
Moisture	15.53	11.02
Total Ash	3.82	6.61
Insoluble Ash	0.29	0.75
Crude Protein	35.16	13.31
Ether Extractives	6.73	0.32
Crude Fibre	19.55	55.23
Nitrogen free extracts	19.21	13.51
Calcium	0.57	1.15
Phosphorus	3.22	1.08

Discussion

The presence of hard testa necessitates crushing of the seeds before feeding. The high content of protein (35.16%) and the mineral content especially Phosphorus (3.22%) in the seed suggest the possibility of using these seeds in the feeding of animals.

Summary

1. Botanical description, the qualities and the method of propagation of *Leucaena glauca* - Benth are briefly stated.

2. The ratio of seeds to the pod cover by weight and the nutrient composition of both are presented.

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Clinical Article

SUCCESSFUL CAESARIAN SECTION IN A COW WITH TORSION OF UTERUS

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Introduction

Torsion of uterus is defined as twisting of gravid horn on its long axis. It is more frequent in cows than in sheep and goats, but rare in mare and small multiparous animals. Torsion of uterus was first mentioned by Bourtrioller of France as early as 1776. It was only in the 19th century that more attention was drawn to this condition. Torsion of uterus may be complete or partial; the foetus will not be delivered if the torsion is pronounced. It was reported from Germany that incidence is 10% in cattle. In Switzerland 70 to 75 of dystokia is

due to torsion of uterus. This is mostly due to the hilly nature of the country. In Sweden where the country is mountainous, the incidence of torsio-uteri has been found to be very high.

History of the case

On 17-8-59, a heifer in advanced pregnancy was admitted as indoor patient, case No. 576. It was reported by the owner that since last five days the animal was restless, and straining much without any avail. Recto-vaginal examination revealed that it was a case of torsion of uterus. The gravid horn close to body of uterus was twisted like a cord with several turns. On vaginal exploration the vaginal mucus folds were felt; os uteri was closed.

The indirect detorsion of the uterus was done by rolling the cow as direct detorsion was not possible due to closure of cervix. The pregnancy was in the right horn and it was right side torsion. The limbs were tied and she was rolled on the same side (right). The control was made by putting the hand in the vagina for judging the detorsion. Three quick successive rollings were given and with this method two fingers could be introduced into the os uteri. The cow was under observation till evening. The cow did not have any sign of straining.

18-8-59. On rectal examination the condition was the same. It was decided to adopt continuous rolling with extra abdominal pressure. Attempts were not successful.

20-9-59. Finding no improvement, caesarian section was undertaken. The animal looked depressed and its appetite was not good. Right flank was selected as the site of operation. It was cleaned and shaved. Epidural anaesthesia with 2% Procaine Hydrochloride 20 c.c. and Paravertebral anaesthesia at the sites of 1st, 2nd and 3rd lumbar spinal nerves with 20 c.c. at each site was given. Chloral-hydras was given with gum-acacia and treacle. The right flank was sterilised with 50% alcohol. A linear infiltration with 2% Procaine Hydrochloride was given in right flank on the proposed line of incision. It was covered with a sterilised drape with central opening of 12 inches and fixed in position with towel clips. A bold incision was made commencing in the mid region between right angle of ilium and last rib and proceeded downwards and forwards parallel to the fibres of the abdominal muscles and bleeding was mopped with sterilised swab. At this stage to supplement the anaesthetic action ethyl chloride was sprayed and abdominal muscles were divided and the peritoneum was secured with an artery forceps and incised with a blunt pointed straight scissors and the opening was enlarged with the help of finger. The divided muscles and peritoneum were secured by Balfour's retractor. The loops of the intestine were set aside

and packed with sterilized gauze soaked with normal saline solution. The gravid horn was visible and was outside the bursasupra-omentalis. The gravid horn looked dark red and congested. It was brought near the site of opening and secured at two places by long handled artery forceps. Every step was taken to drain out uterine fluid by packing gauze round the gravid horn. Incision was made with blunt pointed scissors and the opening was enlarged with fingers. The amniotic fluid was drained. The foetus was visible with posterior presentation. The foetus and placenta were removed. The uterine opening was sutured with double rows of chromic cat-gut No. 2 in continuous and interrupted Lambert fashion. After suturing the incised uterine horn the torsion which looked like twisted rope was corrected. Before closing abdominal opening, Sulphanilamide powder was sprinkled and the gravid horn was covered with omentum. The peritoneum and abdominal muscles were sutured separately with cat-gut and the second line of suture was given to the abdominal muscles with No. 2 cat-gut in continuous and interrupted manner. The skin was sutured in Halsted fashion with silk thread. The cutaneous suturing was accelerated with the aid of sterilized cobbler needle. The cutaneous line of suture was painted with weak Tr. Iodine and sprinkled with Sulphanilamide and was covered with sterilized gauze maintained by three retention sutures. The site of wound was covered with a clean abdominal bandage. The animal's condition did not show any abnormality throughout the operation. After operation glucose saline 200 c.c. was given subcutaneously. Procaine Penicillin 20 lac units were given intramuscularly and to allay pain and surgical shock, Pathedine Hyd. chlor (Delantion) 200 mg. intramuscularly was also given. The operation was started at 3-30 P.M. and was over by 4-45 P.M. At night again 200 mg. Pathedine hyd. chor. was given as there were signs of restlessness. At late hours in the night, animal had slight laboured breathing and the following mixture was given.—

R/-

Tr. digitalis	5ii
Tr. gingeris	5iv
Aqua camphor	Oi

21-8-59. The condition of the animal was hopeful and temperature was 102.6°F. Procaine Penicillin 20 lac units twelve hourly was given. Normal saline solution 100 c.c. intravenously morning and evening, Calcium Borogluconate 25% solution 100 c.c. morning and evening subcutaneously. In the noon the temperature went up to 104.4°F. and came down to 103°F. at 4-30 P.M. The animal tried to get up but was made to rest.

22-8-59. The temperature was 102.6°F and the rest of treatment was as on 21-8-59.

23-8-59. The temperature came to 101.8°F. and the following was given in electuary—

R/-

Sodium salicylas	5iv
Soda bicarb	5iv
Pulv. liquorice	5iv
Treacle	Q.S

and normal saline solution 100 c.c. intravenously. The Procaine Penicillin 20 lacs was given. Sulphamezathine solution 33½% was given 50 c.c. subcutaneously once a day. The animal stood and was given liquid diet in the form of Satu and Gur in drinking water and rice gruel.

24-8-59. The line of suture was dressed with weak solution of Tr. Iodine and Sulphanilamide powder. The wound was healthy and healing under first intention. The temperature was varying from 101 to 103°F. The following drugs were given in the form of electuary twice a day, in addition to other medicines as on 23-8-59 —

R/-

Sodium salicylas	5iv
Pot. acetat,	5ii
Sodi citras	5ii
Tr. digitalis	5ii
Pulv. liquorice	5iv
Treacle	Q.S

The animal was bright and ate green grass and had good rumination.

25-8-59—28-8-59. The temperature varied from 101 to 103°F. and the rest of the treatment was the same.

29-8-59. The temperature ranged from 101 to 103°F. The Penicillin was discontinued as the owner could not afford to provide. The Sulphamezathine was continued upto 2nd September 1959.

3-9-59. The Sulphamezathine was discontinued as the temperature came to normal.

4-9-59. Stitches were removed and dressed with weak solution of Tr. Iodine and Sulphanilamide. The temperature remained normal and the animal was healthy. The animal was discharged as cured.

Discussion

Torsio-uteri in cattle is rare in this country. The statistical data of percentage of occurrence among Indian cattle are not available. In this case the owner revealed that there was an elevated ground where the animal used to be tied and every day the animal had to ascend and descend many times during pregnancy. The exciting cause of the torsion of the uterus may be the elevated nature of the place similar to the high incidence in hilly tracts of the country like Switzerland and Sweden.

In the abdominal cavity the gravid horn was found outside the bursa supra-omentalis. Normally the gravid horn develops in the cow inside the bursa supra-omentalis but sometimes this development occurs outside the omentum and the good fixation of the uteri in the abdominal cavity is lost and this condition predisposes to torsion. This development of gravid uterus outside the bursa supra-omentalis cannot be called an anatomical defect but it is quite physiological. Exact incidence of torsion due to the development outside the omentum is difficult to know clinically until laparotomy is performed. The operation for caesarian section in large animal is attended with some risk. The aseptic technique and post operative care have to be adopted with meticulous care. It is very essential to control the post operative shock. In human beings the use of pathedine Hyd. Chlor. is very much in use to allay pain and surgical shock. The use of Pathedine Hyd. Chlor in large animals may not be extensive. Some Veterinarians have used it in small animals. Pathedine Hyd. Chlor. can give good results in large animals also.

The administration of Glucose Saline, large doses of antibiotics and Pathedine Hyd. Chlor. after operation proved highly useful.

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Abstracts

The effect of Chemotherapy on the numbers of *Bacterium Coli* in the Faeces of Calves:—By H. WILLIAM SMITH and W. E. CRABB. *Jour. of Comp. Pathology and Therapeutics*. Vol. 70, No. 1, Jany. 1960.

In undertaking this work, the authors realised the difficulty in producing the natural disease experimentally and this hampered their efforts to study and to compare the therapeutic effects of different chemotherapeutic agents in a controlled manner. The field trials are also fraught with difficulties. Since the faeces of healthy as well as scouring calves contain large numbers of *B. coli*, it was decided to determine the comparative effect of drugs, particularly those commonly used in treating calf scours, on the *B. coli* population of the faeces of healthy calves. The results showed that the greatest and most prolonged reduction in the *B. coli* faecal count was obtained in calves treated with Streptomycin or Neomycin. Polymixin treatment consistently resulted in a marked reduction, while Terramycin, Aureomycin, Sulphamezathine and Phthalylsulphathiazole had a considerable depressing effect on the *B. coli* faecal count, although in a small proportion of calves no decrease was found. Administration of Chloramphenicol had no effect on the *B. coli* faecal count and similar results were also obtained in calves treated with Furazolidone or Furamazole. A much greater and more prolonged reduction was obtained in the *B. coli* faecal count of the calves given oral treatment than those given subcutaneous treatment.

A. MUKERJI.

Oral treatment of Spontaneous Ringworm in Cats with Griseofulvin:—By W. KAPLAN and L. AJELLO. *J.A.V.M.A.*, 135: 253-261.

The introduction of Griseofulvin, the metabolic product of mycelium of *Penicillium* for the successful treatment of ringworm is a land-mark in the history of the therapy of dermatophytoses. This wonder drug was administered orally to 22 cats naturally infected with *Microsporum canis* and manifesting varying degree of lesions. The drug was given in single daily doses of approximately 60 mg. to cats 3 months old and over and 40 mg. to one month old kittens. The cats were treated every day until they became clinically normal and thereafter twice a week. There was marked clinical improvement in 1-2 weeks and complete disappearance of lesions in 3-5 weeks. The possible prophylactic power of the drug is also indicated.

V. S. ALWAR.

Preserving Anatomical Specimens in a Dry State:—By FRANCOI B. SAKLA, 1960. *Anat. Rec.*, 135 (2): 149.

Anatomical specimens were fixed in 10% formalin, and they were drained from excess formalin by squeezing gently and left for next day covered with gauze. They were dehydrated by painting them daily for a fortnight with a mixture of 50% glycerine, 25% 96% alcohol and 25% acetone and then left to dry for a week. This was followed by painting them several times with pure acetone and then with 3% celloidin solution. These specimens were kept without deterioration for ten years and still kept in the museum of Anatomy Department of Abbassia Medical Faculty, Cairo.

B. PANDA.

Induction of reversal of the Heart Beat in Chick embryos by injection of Novocain into the vein:—By S. SANGVICHIAN, 1959. *Anat. Rec.*, 135 (3): 185.

Co₂ introduced into the chamber occupied by the chick embryo can cause reversal of the heart beat (Sangvichien '52). In order to demonstrate that the sino-atrial portion could be suppressed directly, 1% novocaine tinted with very dilute methylene blue was injected into the vein of the embryo. The drug suppressed the sinus-end of the heart allowing the parts of the heart which are previously formed to resume dominancy thus causing reversal of the heart beat. (Author's).

B. PANDA.

Dried semen successful:—*Agri Leaders' Digest*. Jan. '60, pp. 22.

A grade Ayrshire cow at the University of Maryland has been successfully bred with dried semen. The technique of the dried semen was the result of work by Dr. H. T. Meryman and E. Kafig at the naval medical research institute, M.D. The sperm cells are suspended on a piece of nylon gauze. The gauze bearing the sperm cells is then placed in the vacuum. While under vacuum, rapid evaporation actually freezes the sperm cells and moisture is removed under the vacuum until the material is dry. This work has been cited as an important research development but further research is needed before the technique can be practically adapted.

B. PANDA.

Histochemical distribution of Succinic Dehydrogenase in Bone and Cartilage:—By SCHAJOWICE, F. and R. L. CABRINI, 1960. *Science*, Vol. 31: 1043.

Large amount of succinic dehydrogenase have been demonstrated histochemically in osteoclasts and chondroclasts. The same enzyme was also found in the giant cells of the giant cell tumours of the bone.

It has been shown histochemically that osteoclasts and chondroclasts contain several enzymes such as alkaline phosphatase, acid phosphatase, esterases, amino polypeptidases, and beta-glucuronidase.

The results seem to confirm the hypothesis that these cells are not passive elements but cells with a high metabolic activity and suggest a relation to bone and cartilage resorption.

B. PANDA.

Review

Aids to Biochemistry.—By S. P. DATTA, B.Sc., M.B.,B.S. and J. H. OTTAWAY, B.Sc., Ph.D., A.R.I.C. 5th Ed., 1960. Bailliere, Tindall & Cox, London, W.C.-2. Price 15 sh.

This is a small handbook containing only 264 pages, with 41 illustrations and 29 tables, and is one of the "Students' Aids" series of the above Publishers. The material of the book is based on the course of biochemistry for pre-clinical medical students at the University College, London. The authors have confined themselves to human biochemistry, and little mention has been made of the comparative biochemistry of animals. A number of new techniques such as the use of isotopic tracers, chromatography, electrophoresis, etc., have been described in the book. The biochemical bases of physiological phenomena are more or less the same in man and animals and as such the book will be found useful by students of Veterinary Science.

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