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## THE PATHOGENESIS OF HYDRALLANTOIS BOVIS

### I. THE CONCENTRATIONS OF SODIUM, POTASSIUM, CHLORIDE AND CREATININE IN THE FOETAL FLUIDS IN CASES OF HYDRALLANTOIS AND DURING NORMAL PREGNANCY\*)

By

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The cause for abnormal regulation of the foetal fluids in cases of hydrallantois is unknown. However, recent investigations on the formation of normal allantoic fluid seem to have opened up the possibility of investigating the development of hydrallantois.

The connection through urachus between the urinary bladder and the allantoic cavity results in a similarity in the composition of allantoic fluid and foetal urine in the rabbit (*Dickerson & McCance 1957*) and sheep (*Jacqué 1902; Alexander et al. 1958*). However, it appears that the composition of the allantoic fluid also is the result of transport across the chorio-allantoic membranes. This is indicated by the finding that the chloride concentrations in the allantoic fluid are lower than in the corresponding foetal bladder urine in sheep (*Alexander et al. 1958*), and the *in vitro* demonstration of active sodium transport across the chorio-allantois of the pig (*Crawford & McCance 1960*).

In investigating the pathogenesis of hydrallantois the alterations in composition of the allantoic fluid are of special interest. *Febvre et al. (1955)* found that in normal cows the allantoic fluid had electrolyte concentrations that deviated strikingly from the concentrations in blood plasma, while *Neal (1956)* in a case of bovine hydrallantois found sodium and potassium concen-

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trations similar to those of blood plasma. This apparent difference between normal pregnancy and hydrallantois indicates a marked change in the function of the foetal kidney and/or for the chorio-allantoic membranes.

In an attempt to further understand these changes, the concentrations of sodium, chloride, potassium and creatinine were measured in foetal fluids from both normal cows and from cows with hydrallantois.

### MATERIAL AND METHODS

The normal material included foetal fluids from 92 normal uteri, opened immediately after the slaughtering. The criteria for normal fluids were:

1. Absence of multiple pregnancy.
2. Absence of apparent signs of congenital deformity.
3. Foetal fluid volumes which did not exceed one standard deviation from the mean monthly values given by *Jakobsen* (1958) and *Arthur* (1957).

The foetal age for Red Danish dairy cattle which makes up the majority of the normal material was calculated on the widest circumference of the foetal head (*Jakobsen* 1959). For the Jersey and the Holstein cattle the foetal age has been judged in weeks or months according to the method of *Franck* (1876).

Foetal fluids recovered from 16 cases of hydrallantois bovis during the last third of the pregnancy were examined. To avoid doubtful cases it was decided to include only patients with a minimum of 75 liters of allantoic fluid.

*Methods of analysis.* The sodium and potassium concentrations were examined by flame photometry (Beckman, model 41). Nine standard solutions between 10 and 160 mEq/l were used. Chloride was estimated ad modum *Schales & Schales* (1941). Creatinine was estimated spectro-photometrically as described by *Poulsen* (1956).

### RESULTS

Table 1 shows the concentrations of sodium, potassium, chloride and creatinine in normal allantoic and amniotic fluids. For the last six months of pregnancy these results are in accordance with those of *Febvre et al.* (1955).

Table 2 shows the hydrallantoic material which in several ways deviated from the normal foetal fluids.

Table 1. Average and standard deviation of concentrations of sodium, potassium, chloride and creatinine in allantoic and amniotic fluids from normal cows.

Month of pregnancy	Allantoic fluid						Amniotic fluid						no of fluids
	mEq/l			$\mu\text{g/ml}$			mEq/l			$\mu\text{g/ml}$			
	Na	K	Cl	creatinine	Cl	creatinine	Na	K	Cl	creatinine	Cl	creatinine	
2	103 ± 13	6.2 ± 2.2	70 ± 14	20 ± 5	8	139 ± 4	13 ± 2	118 ± 3	9.6 ± 2.3	6			
3	86 ± 19	7.3 ± 3.6	46 ± 18	66 ± 47	11	136 ± 6	16 ± 4	117 ± 4	9.3 ± 2.1	11			
4	52 ± 28	21 ± 15	31 ± 19	144 ± 59	9	129 ± 6	18 ± 2	118 ± 3	12.1 ± 6.9	10			
5	51 ± 15	20 ± 8	25 ± 11	241 ± 92	12	130 ± 3	16 ± 4	117 ± 4	15.4 ± 5.8	11			
6	43 ± 19	32 ± 21	29 ± 15	366 ± 149	13	133 ± 5	10 ± 2	113 ± 4	32 ± 17	14			
7	54 ± 23	24 ± 24	21 ± 15	822 ± 346	19	132 ± 5	7.6 ± 1.7	113 ± 6	67 ± 26	18			
8	45 ± 20	26 ± 26	15 ± 9	1110 ± 233	13	134 ± 6	5.6 ± 0.8	115 ± 9	77 ± 30	11			
9	53 ± 13	15 ± 13	20 ± 15	1560 ± 683	7	125 ± 7	4.7 ± 0.5	114 ± 9	48 ± 14	4			

Table 2. Concentrations of sodium, potassium, chloride and creatinine in foetal fluids from cows with hydrallantois (7—9 months of pregnancy).

Breed	Month in gestation-period	Amount of allantoic fluid l	Allantoic fluid				Amniotic fluid				
			mEq/l			$\mu\text{g/ml}$	mEq/l			$\mu\text{g/ml}$	
			Na	K	Cl	creatinine	Na	K	Cl	creatinine	
Red Danish	7½	110	116	10	96	—	—	—	—	—	
Jersey	9	100	119	24	61	—	146	4.6	95	—	
Red Danish	9	170	90	4.2	90	230	135	5.9	100	38	
Jersey	8	110	121	10	79	105	131	8.9	114	32	
Red Danish	9	110	126	14	83	180	123	9.0	90	79	
”	”	8	100	122	22	73	290	130	11	85	130
”	”	7½	100	88	26	55	290	119	13	85	95
”	”	7	110	108	18	90	100	105	12	73	39
Jersey	8	100	122	8.7	91	190	130	8.3	101	109	
Red Danish	8	90	118	3.0	70	190	119	3.2	78	148	
Jersey	8	140	139	3.2	90	123	141	4.0	106	52	
Red Danish	7	130	121	9.7	95	120	—	—	—	—	
”	”	8½	170	122	16	95	175	—	—	—	
”	”	8½	100	115	11	73	348	124	7.0	76	89
Jersey	8	120	125	11	74	163	—	—	—	—	
Red Danish	8	90	108	14	81	—	—	—	—	—	

In the beginning of the pregnancy the electrolyte and creatinine concentrations in normal allantoic fluid were approximately identical to extracellular fluid. After about three months of the pregnancy the allantoic fluid had almost lost this character, and the concentrations now showed greater variations.

From the average values the following characteristics for the electrolytes are noticed (Fig. 1):

The chloride concentrations were consistently low relative to plasma levels and decreased until the eighth month of the pregnancy. The sodium and potassium concentrations seemed to show an inverse relationship.

In the last six months of the pregnancy when the average concentrations of sodium varied around 50 mEq/l, the interdependency of sodium and potassium concentrations is illustrated in the following figures: in cases of sodium concentrations above the average value the potassium concentrations had an average value of 12.5 mEq/l, and in cases of sodium concentrations below average the potassium concentration had an average value of 37.5 mEq/l. When the potassium concentrations

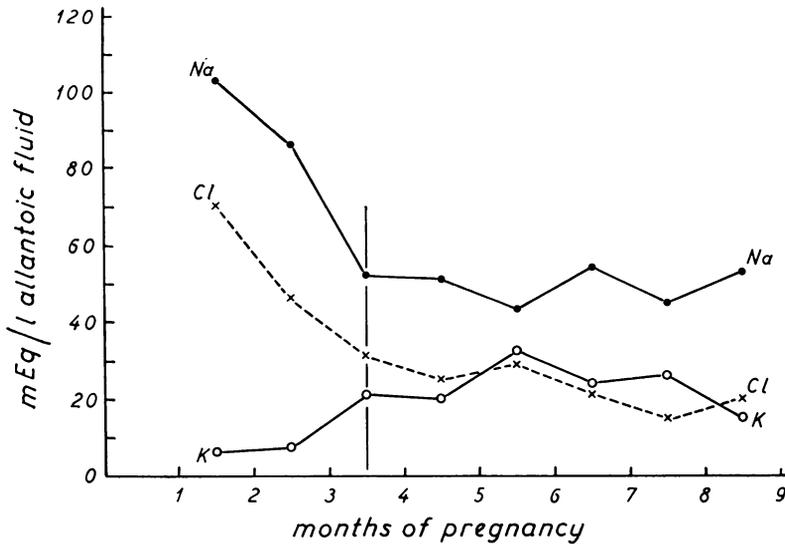


Fig. 1. The average concentrations of sodium, potassium and chloride in the allantoic fluids during pregnancy (normal cows).

were above average the sodium concentrations had an average value of 33 mEq/l, and in cases of potassium concentrations below average the sodium concentrations had an average value of 60 mEq/l.

The creatinine concentrations in normal allantoic fluids increased progressively during the pregnancy as shown in Fig. 2. This increase seemed to be an exponential function of time after

Table 3. Concentrations of sodium, potassium, chloride and creatinine in foetal fluids from cows with hydrallantois and from normal cows. (Average and standard deviation).

	No	mEq/l			$\mu\text{g/ml}$
		Na	K	Cl	creatinine
Allantoic fluid,					
normal	23	52 $\pm$ 20	20 $\pm$ 21	17 $\pm$ 11	1224 $\pm$ 458
hydrallantois	16	116 $\pm$ 13	13 $\pm$ 7	81 $\pm$ 12	193 $\pm$ 73
Amniotic fluid,					
hydrallantois	11	128 $\pm$ 11	7.9 $\pm$ 3.3	91 $\pm$ 13	81 $\pm$ 41
normal	23	132 $\pm$ 7	5.5 $\pm$ 0.9	115 $\pm$ 8	70 $\pm$ 26
Blood plasma					
in normal cows*		140 $\pm$ 4	4.0 $\pm$ 0.3	101 $\pm$ 4	13.9 $\pm$ 1.8

\* The electrolyte values originate from Knudsen (1960).

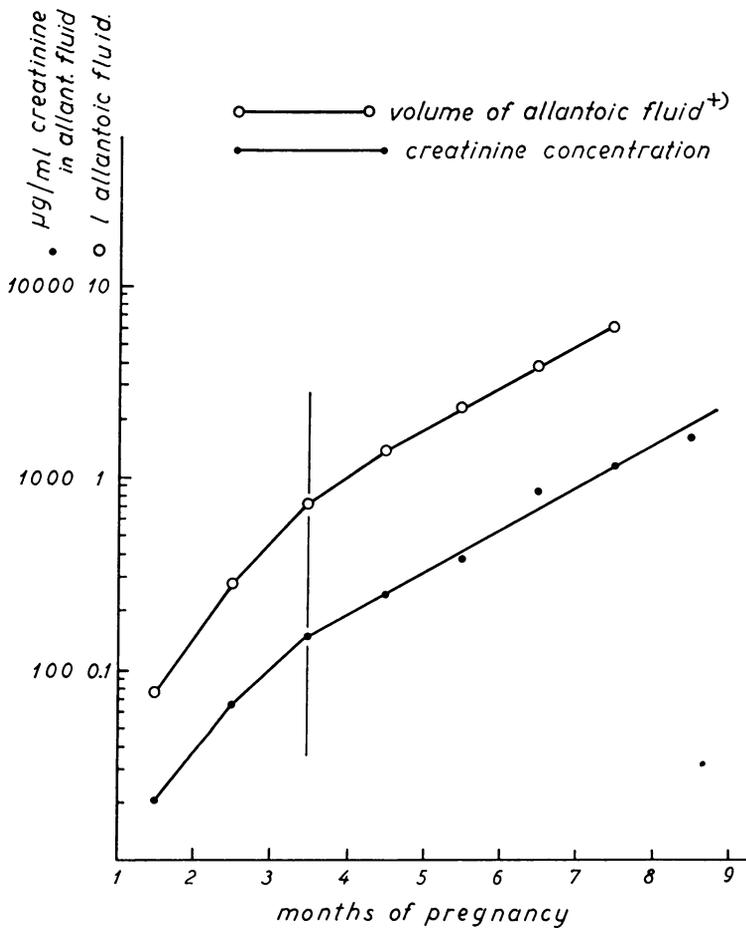


Fig. 2. The monthly average concentrations of creatinine in the allantoic fluids showing the increase of creatinine concentrations during normal pregnancy in the cow.

\*) The increase of allantoic fluid volume is shown with figures given by *Jakobsen* (1958).

three months of pregnancy. The results in *Jakobsen's* (1958) and *Arthur's* (1957) investigations on the foetal fluids show that the volume of allantoic fluid also increases exponentially with time. *Jakobsen's* volume results are included in Fig. 1.

In cases of hydrallantois the allantoic fluids deviated from the corresponding normal fluids. The electrolyte concentrations obviously approached those of extracellular fluid and the stand-

Table 4. Average of total contents of sodium, potassium, chloride, creatinine and water in the allantoic fluids from 16 cases of hydrallantois bovis and from normal bovine pregnancy (8th month).

	mol			l	g
	Cl	Na	K	water	creatinine
Normal allantoic fluid	0.13	0.37	0.16	7.6*	12
Hydrallantoic fluid	9.4	13.5	1.9	116	22
Hydrallantoic fluid	72	37	12	15	2
Normal allantoic fluid					

\* from figures given by *Arthur* (1957) and *Jakobsen* (1958), calculated in accordance with the exponential volume increase during pregnancy.

ard deviations were considerably smaller. This is shown in Table 3 which lists the average values and standard deviations for the foetal fluids of normal animals and animals with hydrallantois during the last 12 weeks of pregnancy. The table shows furthermore that creatinine concentrations of the allantoic fluids are considerably lower in cases of hydrallantois although the standard deviations were approximately the same.

Table 4 gives the mean quantities of chloride, sodium, potassium and creatinine in the allantoic fluids from hydrallantois and normal pregnancy. The mean fluid volumes are also given. The hydrallantois/normal pregnancy ratios showed that in the cases of hydrallantois the increased accumulation in the allantoic fluids was greatest for chloride and sodium, lower for potassium and water, and lowest for creatinine.

In *normal amniotic fluid* the sodium and chloride concentrations examined were found to be in the range of those found in extracellular fluid (Table 1). The potassium concentrations, however, were relatively high until near the end of the pregnancy. The creatinine concentrations increased above normal extracellular concentrations after about the fifth month.

*The amniotic fluids from cases of hydrallantois* were slightly altered in composition. Chloride concentration was lower than normal and the average concentrations of potassium and creatinine were slightly higher than normal. The variability tended to increase especially for potassium, chloride and creatinine concentrations (Table 3). Fig. 3 shows that in normal amniotic

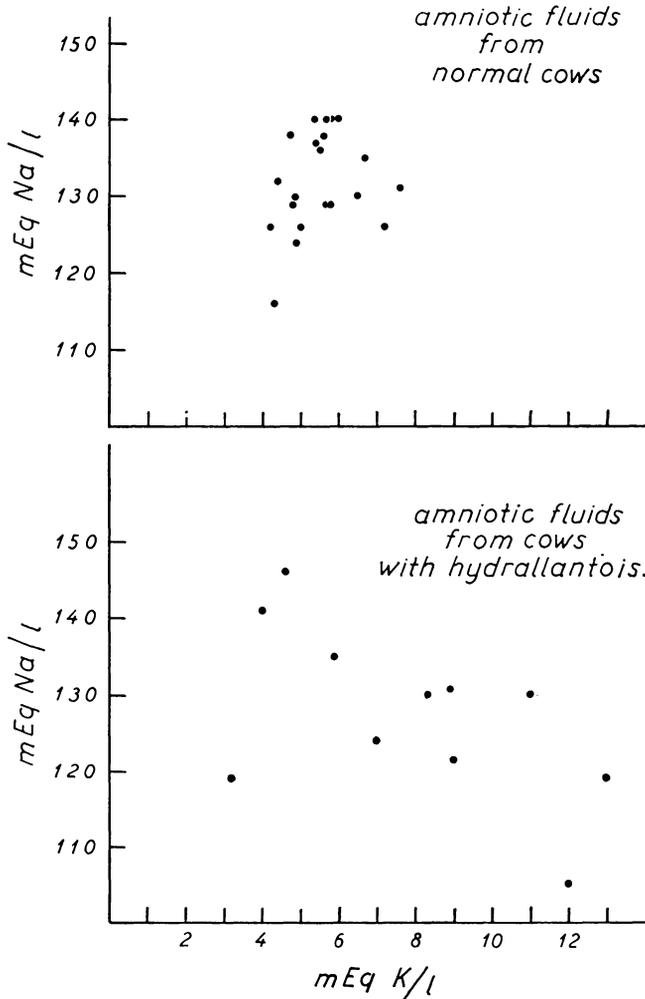


Fig. 3. The relationship between sodium and potassium concentrations in the amniotic fluids.

fluids the concentrations of sodium and potassium varied independently, whereas in cases of hydrallantois there did appear to be a definite inverse variation.

#### DISCUSSION

The normal allantoic fluid evidently undergoes alteration from the second to the fourth month of pregnancy. This alteration coincides with the structural maturation of the placenta which

seems to be complete after about three months of pregnancy (*Björkman* 1954). It also coincides with the development of a functional mesonephros which is completely developed towards the end of the first month (*Bremer* 1915; *Shumkina* 1960) and terminated before 3 months of pregnancy (*Zietzschmann & Kröling* 1955).

Comparative studies indicate that the size and duration of the mesonephros are closely related to the size of the allantoic cavity (*Bremer* 1916), and the composition of the allantoic fluid is influenced by mesonephric excretion in the rabbit (*Davies & Routh* 1957). However, complete similarity between allantoic fluid and foetal urine has never been found, either in the mesonephric period (investigated on pig and rabbit by *Stanier* 1960), or in the metanephric period of animals which retain the allantoic cavity after the involution of mesonephros (investigated in pigs by *McCance & Stanier* 1960, and in sheep by *Alexander et al.* 1958). Therefore transport across the allantoic membranes must have taken place early in the examined animals, and the allantoic cavity should not be exclusively regarded as a reservoir of foetal urine.

The question of whether the kidney or the placenta takes care of the foetal excretion should be regarded as only part of the greater question of exchange between the fluid compartments of foetus and dam. At an early foetal stage the allantoic fluid expands considerably before any renal excretion can be developed and the fluid resembles plasma dialysate. Later, the mesonephric urine arriving in the allantoic cavity still resembles plasma dialysate in spite of a considerable development of tubular reabsorption (*Stanier* 1960). However, the composition of the allantoic fluid begins deviating from that of plasma water. Finally, metanephric urine arrives in the allantoic cavity and although progressive increase in the creatinine content indicates accumulation of urine, this urine is modified in composition. This is indicated, for example, by the difference in the chloride concentrations of the allantoic fluid and corresponding bladder urine (*Alexander et al.* 1958; *McCance & Stanier* 1960). In mammals the allantoic fluid thus seems to be a constant part of the common fluid exchange.

An active sodium transport away from the allantoic cavity is probably quite important. *Crawford & McCance* (1960) demonstrated such a transport across the chorio-allantoic membranes

in pigs. The electrical potential gradients resulting from active sodium transport would make the dam's blood electropositive to the allantoic fluid, and such electric potential gradients have been found in rabbits (*Krespie & Davies 1963; Wright 1963*), goats (*Meschia et al. 1958*), sheep and cats (*Widdas 1961*) and in cows (*Skydsgaard 1965*). Although comparisons should be made with reservation, and although electric potential gradients between dissimilar solutions may be due to both active ion transport and chemical gradients, it appears that there is a transport of ions and water across the bovine allantoic membranes throughout pregnancy. After the differentiation of placenta and involution of the mesonephros, an equilibrium appears to be developed and maintained the last six months of pregnancy. The equilibrium between fluid arrival and removal is presumably also reflected by the exponential accumulation of water and creatinine in the allantoic cavity after about three months.

The inverse relationship between the sodium and potassium concentrations in allantoic fluid is a general characteristic of body fluids due to the exchange of these ions across the cell membranes, and in the allantoic fluid the inverse relationship might be due to such ion exchange in the renal tubules of the foetus and/or the border membranes of the allantoic cavity.

In cases of hydrallantois the allantoic fluid showed an abnormal increase in volume with a simultaneous approach to the concentrations of electrolytes found in plasma. As the production of allantoic fluid depends on the foetal kidney function as well as on the transport across allantochorion such a "hydrallantoic fluid" may, according to our present knowledge, be due to at least three different factors.

1. A marked tubular dysfunction of the foetal kidney tubules with a normal glomerular filtration rate resulting in a considerably increased urine volume with electrolyte concentrations close to those of plasma. This coupled with minimal reabsorption from the allantoic cavity would give the above results.
2. If an essential function of the chorio-allantoic membranes is active sodium transport with secondary passive transport of other ions and water, a decreased active sodium transport could result in the accumulation of sodium, chloride and water in the allantoic cavity.
3. Increased permeability of the chorio-allantoic membranes to sodium, chloride and potassium could result in equilibration

between allantoic fluid and plasma in spite of continued active transport of sodium. This would also decrease the rate of water reabsorption.

A renal tubular dysfunction alone does not explain the pathogenesis of hydrallantois unless one assumes a very low rate of reabsorption from the allantoic fluid which seems to be incompatible with our present knowledge. It thus appears most likely that a structurally or functionally defective in the allantochorion is involved.

Although the amniotic fluid in case of hydrallantois deviates little from normal, signs of certain changes in the formation-reabsorption reveal themselves. The tendency towards lower sodium and chloride concentrations, higher potassium and creatinine concentrations and increased variability suggests that the fluid exchange involved in the formation of the amniotic fluid is not able to mask the urine contamination as much as in normal pregnancy. This is further indicated by the appearance of an inverse variation between the sodium and potassium concentrations in the amniotic fluid (Fig. 3). Little is known about the formation-reabsorption of the amniotic fluid, but tracer experiments (reviewed by *Plentl & Gray* 1957) have shown that the amniotic fluid is exchanged so rapidly that urine admixture at best plays a much subordinated part. Therefore, the change in characteristics of amniotic fluid in cases of hydrallantois suggests either a decreased fluid exchange or an increased urine flow to the amniotic cavity.

### CONCLUSIONS

1. In the beginning of the normal bovine pregnancy the concentrations of sodium, potassium and chloride in allantoic fluid resembled those found in extracellular fluid, but this similarity completely disappeared after about three months of pregnancy. Then low chloride concentrations were found together with a decreased sodium concentration and increased potassium concentration, the latter two showing great variations with tendency to inverse relationship.
2. The amount of creatinine in the allantoic fluid increased exponentially with time after about three months of pregnancy.
3. The allantoic fluid from normal cows is characterized partly by foetal urine, partly by transport activity across the chorio-allantoic membranes.

4. In cases of hydrallantois the electrolyte composition of the allantoic fluid differs from the corresponding normal allantoic fluid and closely resembles that of extracellular fluid. The amniotic fluid seems to be more marked by urine admixture than normal amniotic fluid.
5. The pathogenesis of hydrallantois seems to be associated with structural or functional changes in the chorio-allantoic membranes. A contemporary dysfunction of the foetal renal tubules cannot be excluded.

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#### SUMMARY

The concentrations of sodium, potassium, chloride and creatinine in allantoic fluid have been investigated in normal cows and in cases of hydrallantois. After three months of pregnancy the ion concentrations in normal allantoic fluid lost their initial resemblance to extracellular fluid. During pregnancy the concentrations of creatinine in allantoic fluid increased, and after about three months the increase became slower but exponential with time.

The excretion-reabsorption mechanisms responsible for the characteristics of the allantoic fluid the last 6 months of normal pregnancy appear to be altered or ineffective in cases of hydrallantois. The sodium, potassium, and chloride concentrations contrary to normal conditions resemble extracellular concentrations, and the creatinine concentrations were much lower than normal.

In hydrallantois a structural or functional change of chorio-allantois seems to be present although a dysfunction of the foetal kidney cannot be excluded. The problem requires further investigation of ion transport and permeability across the chorio-allantoic membranes.

### ZUSAMMENFASSUNG

#### *Die Pathogenese der Hydrallantois bovis.*

#### *I. Die Konzentrationen von Natrium, Kalium, Chlorid und Kreatinin der Allantoisflüssigkeit bei der Hydrallantois und bei normaler Trächtigkeit.*

Es wurden die Konzentrationen von Natrium, Kalium, Chlorid und Kreatinin in der Allantoisflüssigkeit bei normalen Kühen und bei Kühen mit Hydrallantois untersucht. Nach einer Tragezeit von 3 Monaten verloren die Ionenkonzentrationen in der normalen Allantoisflüssigkeit ihre anfängliche Gleichheit mit den Konzentrationen in der Flüssigkeitsphase der Extrazellularflüssigkeit. Die Kreatininkonzentration zeigte durch die Trächtigkeit hindurch ein progressives Ansteigen, das sich nach ca. 3 monatiger Trächtigkeit verlangsamte, jedoch exponential mit der Zeit.

Der Exkretions-reabsorptionsmechanismus, welcher der Allantoisflüssigkeit während der letzten 6 Monate der Trächtigkeit ihre charakteristische Zusammensetzung gibt, scheint beim Vorliegen einer Hydrallantois verändert zu sein oder weniger wirksam zu sein. Die Konzentrationen von Natrium, Kalium, und Chlorid ähneln im Gegensatz zu den normalen Konzentrationen denen der extrazellulären Flüssigkeitsphase und die Kreatininkonzentration lag weit unter dem Normalen.

Bei der Hydrallantois scheint eine strukturelle oder funktionelle Veränderung der Allanto-chorion vorzuliegen, obgleich eine gleichzeitige Dysfunktion der embryonalen Nierenfunktion nicht ausgeschlossen werden kann. Das Problem erfordert eine weitere Untersuchung des Ionentransportes und der Permeabilität der Eihautmembranen.

### SAMMENDRAG

#### *Patogenesen ved hydrallantois bovis.*

#### *I. Koncentrationerne af natrium, kalium, klorid og kreatinin i allantoisvæsken ved hydrallantois og under normal drægtighed.*

Koncentrationerne af natrium, kalium, klorid og kreatinin i allantoisvæsken er blevet undersøgt blandt normale køer og køer med hydrallantois. Efter 3 måneders drægtighed mistede ionkoncentratio-

nerne i normal allantoisvæske deres initiale lighed med koncentrationerne i extracellulærvæskens vandfase. Gennem drægtighedsperioden viste kreatininkoncentrationen en progressiv stigning, som efter ca. 3 måneders drægtighed blev langsommere, men exponentiel med tiden.

Den ekskretions-reabsorptions mekanisme, som giver allantoisvæsken dens karakteristiske sammensætning i drægtighedsperiodens sidste 6 måneder, synes at være ændret eller mindre effektiv i tilfælde af hydrallantois. Natrium, kalium og klorid koncentrationerne ligner i modsætning til normale koncentrationer extracellulærvæskens vandfase, og kreatininkoncentrationerne var langt mindre end normalt.

Ved hydrallantois synes en strukturel eller funktionel forandring af allanto-chorion at være til stede, selvom en samtidig dysfunktion af fosternyrefunktionen ikke kan udelukkes. Problemet kræver yderligere undersøgelse af iontransport og permeabilitet gennem allanto-chorion.

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