THE NEW ZEALAND VELVET ANTLER INDUSTRY : BACKGROUND AND RESEARCH FINDINGS

J.M. Suttie, P.F. Fennessy, S.R. Haines, M. Sadighi, D.R. Kerr and C. Isaacs* AgResearch, Invermay Agricultural Centre, Private Bag 50034, Mosgiel New Zealand and

*New Zealand Game Industry Board, Wellington, New Zealand

SUMMARY

Deer farming has developed over the last 25 years in New Zealand. Deer were introduced in the last century and still run wild. However, there are now over 1 million farmed deer, most of which are red deer and their hybrids with wapiti.

Velvet antler is an important product and much research has been carried out, mainly at Invermay. Research has included influence of nutrition and breeding on velvet production. More recently, antler composition has been investigated. Minerals, lipids, sphingomyelins and free amino acids have bee: tified. Extracts of velvet antler have been developed and research on the biological effectiveness of these extracts using cell lines *in vitro* has been carried out. This research is integrated with a basic research programme in antler growth in order to advance our knowledge of velvet antler and to discover more of its potential applications for the benefit of velvet consumers.

THE NEW ZEALAND VELVET ANTLER INDUSTRY: BACKGROUND AND RESEARCH FINDINGS

J.M. Suttie, P.F. Fennessy, S.R. Haines, M. Sadighi, D.R. Kerr and C. Isaacs*
AgResearch, Invermay Agricultural Centre, Private Bag 50034, Mosgiel New Zealand
and
*New Zealand Game Industry Board, Wellington, New Zealand

PREAMBLE

Although Agriculture is, and has always been, the mainstay of the New Zealand economy, deer farming is a relatively new land use option. The aim of this paper is firstly to provide a background on the New Zealand deer industry, particularly related to velvet antler production, secondly to describe the research which has taken place on New Zealand velvet antler and finally to evaluate the research findings and consider their implications both in New Zealand and internationally.

INTRODUCTION AND BACKGROUND

Geography

New Zealand lies in the south west Pacific Ocean and consists of two main and several smaller islands. The combined size of 270,500 square kilometres is similar to that of the state of California in the USA. The main North and South Islands lie on an axis running north east to south west and extend from 34° to 47° south latitude. The combined length of the North and South Islands is more than 1600 kilometres but the maximum breadth is only 450 kilometres. Both islands are mountainous with less than a quarter of the land area lying below 200 m above sea level (New Zealand Yearbook, 1994). New Zealand is isolated with the nearest continental land mass is Australia, 1600 kilometres to the west. The New Zealand climate is strongly influenced by the long, narrow mountainous shape of the country and by the large expanse of ocean surrounding it (New Zealand Meteorological Service 1986). New Zealand lies in the zone of predominantly westerly winds which bring cool air from sub-antarctic oceans. These winds meet warmer air from the north, which results in rain followed by cool showery weather. Most of the rain falls to the west of the mountain ranges producing sharp climatic differences from west to east. Overall the North Island is warmer than the South Island (Table 1) and has fewer frosts. Snow lies

permanently above 2500 m and 2000 m in the North and South Islands respectively. Snow falls occur to sea level in the South Island several times each winter.

New Zealand is thus a cool wet country with cold winters in the South. The summers are warmer leading to a high degree of seasonal variation in climate. The vegetation of New Zealand has a high degree of endemism at the species level due to its long isolation from continental land masses. The heavy rainfall leads to an excellent environment for tree production. New Zealand, before colonisation by humans, had only two mammalian species (both bats) and a considerable diversity of bird species (New Zealand Atlas 1976). New Zealand can be thought of as having a cool, temperate seasonal climate, with vegetation and fauna characteristic of its long isolation. Both flora and fauna have been modified by introduced species.

Deer

Deer were first introduced into New Zealand by European settlers in the mid to late 19th century for recreational hunting. Introductions mainly of red deer (Cervus elaphus scoticus) and fallow deer (Dama dama) took place in both North and South Islands. The red deer were predominantly from Scottish wild and English park stock. Smaller numbers of wapiti (C.e. nelson), white tailed deer (Odocoileus virginianus), sika deer (C. nippon) sambar (C. unicolor) rusa (C. timorensis) and moose (Alces alces) were also introduced and established populations. The red deer particularly have become well established in suitable habitats throughout much of the country. A commercial deer meat recovery business began and in 1972-3 yielded a return of NZS9 million in export earnings.

Deer farming was legalised in 1969 and grew slowly through until the late 1970's when it became apparent that deer could compete favourably with more traditional forms of livestock as farmed animals. This facilitated the attainment of high hygiene standards required by markets for venison and co-products, such as velvet, as well as providing an effective farming option. To meet the demands for live deer, innovative capture systems were developed. Most deer were captured from helicopters using dart rifles initially and then 'net guns'. These net guns shot a weighted net over the deer which was then put into a canvas bag and winched to a deer fenced area. New Zealand farmers already had considerable experience in handling sheep and cattle and initially husbandry measures for deer were based on traditional practices. These days deer are typically kept all year round in large paddocks, fenced to a height of 2 meters. The paddocks are sown with clover, grasses and herbs. The farmers understand well the nutritional needs of the deer and the food

supply is carefully managed to ensure the animals have sufficient all year round. This includes the provision of supplementary hay in winter, since pasture does not grow at that time of year in New Zealand's seasonal climate. The New Zealand farming system can be thought of as careful stock management in extensive conditions. There are over 1 million farmed deer, most of which these are red deer but there are also significant numbers of wapiti, wapiti x red deer hybrids and fallow deer. The red deer can be thought of as a dual purpose animal providing both meat (venison) and coproducts (velvet antler, tails, pizzles and hides). The velvet is removed under veterinary supervision and the deer for export are slaughtered in specific deer slaughter premises under inspection by the Ministry of Agriculture and Fisheries. In 150 years the deer has gone from an animal introduced for hunting to an animal of considerable importance to New Zealand and New Zealand farmers.

Velvet antier production

About 340,000 stags are kept on New Zealand deer farms for velvet antler production. In the year ending December 1993, 160 tonnes of dried velvet antler were exported. This was worth NZS47 million. Most of the velvet antler is produced by specialist herds or from specialist herds within a larger farming unit. Stags kept for velvet production are managed carefully. From late winter onwards they are observed frequently and the date of casting of the hard antler buttons is recorded. Casting of the previous hard antler button coincides with the spring flush in natural pasture growth. Consequently over the period of velvet antler growth the deer eat only natural vegetation. The deer are also kept on natural pasture during summer, following antler harvest, and during autumn. During the winter breeding season, the stags are not very interested in eating and use up a lot of their fat reserves which have built up during spring and summer. This is the natural cycle for the stag. After the breeding season, the stags start to eat more food but they do not regain much of the weight lost over the breeding season (rut). During the winter, they are fed well but only manage to maintain or slightly increase their body weight. It is during spring (ie, the period of velvet antler growth) when the stags increase their food intake and gain bodyweight. During summer they continue to build up fat reserves for the breeding season in autumn.

The antiers are then removed either by a veterinarian or by a skilled producer who has been accredited by a veterinarian. About 8 weeks after the casting of the previous hard antier the stags are quietly brought to specially designed indoor pens (yards). The yards have provision for the

hygienic removal and storage of velvet. In the yards, the stags which are ready for antler harvest are separated and those remaining are returned to the paddock. Antler harvest takes place with minimal restraint. Tourniquets made of rubber or plastic are placed round the pedicles to prevent bleeding. The antlers are removed with a saw, allowed to cool and are then frozen, at. The stags are released to pasture. The antlers are processed using traditional methods and most are exported in dried form. Velvet antler production in New Zealand is from stags kept outside on natural pastures and carefully tended throughout the year. Removal takes place under controlled hygienic surroundings.

RESEARCH ON VELVET ANTLERS

Research on deer farming began in New Zealand in 1968 at Lincoln University (then College). In 1973 Dr Ken Drew initiated a Deer Programme at Invermay which since then has grown in size and scope. Some research on deer also takes place at the Ruakura and Grasslands Research Centres and at Massey and Lincoln Universities. Most of the research specifically on velvet antler production has taken place at Invermay. The remainder of this paper is devoted to reviewing antler production, composition and biological effectiveness research.

Antler production

Antler production research has fallen into three categories: optimal harvest time, nutrition and genetics. As almost all the research has taken place on red deer, species will only be described if it differs.

Optimal time of harvest

When deer farming began in New Zealand guidance on when to harvest antiers primarily came from Oriental buyers. At that time the feasibility of keeping specialist velvet herds was under consideration (Elworthy 1976). Observations by producers and by scientists revealed that the period of normal antier growth was 90-120 days and that the Oriental market required about half grown antiers (Wallis 1976, Fennessy 1981). Studies at Invermay showed that the pattern of growth of red deer antiers could be related to the market requirement; antiers were at the optimal stage for harvest when the royal tines had just begun to appear. This took place 55-70 days after

the casting of the previous hard antler (Fennessy 1981, Moore 1984). Moore (1984) studied the optimal timing for harvest in greater detail. Using five year old stags he removed one antler just as the royal tines were developing (Stage 1) and allocated the second antler grown by each stag to harvest either when the royal tines were 0.25 cm (Stage 2) or 0.5 cm (Stage 3) long. Moore found that Stage 2 was reached seven days after Stage 1, and Stage 3 was reached three days after Stage 2. An increase in antler weight was observed (Table 2). Although today the precise removal time is a matter for discussion between producers and buyers, and is based on the standard grading system, Moore (1984) showed that almost daily changes in antler growth could influence grade and hence returns. The principle reason for downgrading antlers which were perceived as over-developed was that they were over-calcified. This means that the process whereby the antler develops into bone was too advanced. Although discussion took place on manipulating this event so that larger velvet antlers would eventuate, no research was ever initiated.

Nutrition

Antler production increases both with age and bodyweight of stags. Moore et al (1988) showed that within stags of the same age antler weight increased 0.1 - 0.2 kg for each 10 kg increase in liveweight. In another study, Fennessy (1989) showed that stags from 2 to 5 years of age from 3 separate herds increased antler production each year (Table 3).

Although early attempts took place to increase antler growth rate, it became clear that improved nutrition could not increase antler weight or growth rate above that of the genetic potential (Fennessy and Suttie 1985). Stags have an annual cycle of weight gain and loss which occurs even when stags are fed high quality feed to appetite all the year round (Pollock 1975, Fennessy et al 1981, Suttie et al 1983). Stags during winter are at their minimum annual bodyweight and outdoors cold weather conditions outdoors put considerable stress on stags to maintain a positive energy balance. Special feeding trials were carried out with stags fed good quality diets to appetite at various times from autumn through to spring. These stags were then compared with other stags fed high quality supplements so that they would eat more total food. The trials showed that increasing the amount of food the stags ate (by increasing feed quality) resulted in only a slight increase in velvet antler yield (Table 4). These results indicate that stags should be well fed after the breeding season, during winter and during antler growth to optimise production. However

feeding high levels of protein to stags during winter did not significantly increase antler weights the following spring (Table 5) (Suttie 1992). On NZ deer farms stags during antler growth are given free access after the breeding season to high quality natural pasture.

Breeding

There is considerable variation in antler size within a group of stags (Table 6). Significant progress in velvet antler production can be made by selecting sire stags (Table 7) but selection is currently based on antler size alone, and it would be desirable to also base this on a concept of defined quality were that available.

A further possibility to increase antler weight is to hybridise farmed red deer with large sub species such as the Canadian wapiti. In New Zealand, although hybrids with red deer are well performed, wapiti imported from Canada have not achieved their genetic potential (Table 8) (Fennessy and Pearse 1990). This has frustrated attempts to greatly increase antler production by using clite sires. In the NZ deer industry as a whole considerable use is made of top antler producing red deer imported fro Europe and wapiti from Canada to increase antler production. Top producing sires have the added advantage that they grow antler of the size and shape preferred by the Korean market.

Antler composition

Antlers grow extremely rapidly and it is to be expected that variations in chemical composition will take place both with time and also with position in the antlers. The following studies were carried out on NZ antler to determine the effect of species, section of antler or time on chemical composition. The studies were carried out in two parts: firstly, mineral composition and total lipid levels were determined and secondly, the levels of specific lipids and free amino acids were measured.

Mineral and lipid composition

Velvet antler from NZ born and bred adult red deer, 2 year old red deer, wapiti and fallow deer were available for analysis (Table 9).

On receipt the dried velvet antlers were weighed, the hair was burned off using a gas burner, and the antlers were reweighed after being cleaned lightly with a damp cloth. Each antler was then cut into 4 main sections for analysis, as outlined in Figure 1. Bez tines were excluded from all analyses. The frozen antlers were cut into sections, freeze-dried and the hair then burned off. Wet and dry weights were recorded. Complete analyses were carried out for all main antler sections. In all cases the dermis was peeled off the velvet antler before analysis. The complete analysis included dry matter (DM, by freeze drying), ash (by incineration in a muffle furnace at 550°C overnight), total lipid (by petroleum ether extraction for 7 hours), nitrogen (by Kjeldahl extraction) and determinations of the following minerals: calcium, phosphorus, sulphur, magnesium, potassium, sodium, manganese, zinc, copper, iron, selenium and cobalt. The nitrogen and mineral analyses were carried out using standard methods for analysis of plant material (Anon 1979).

Adult red deer top grade autler

Table 10 presents the analytical data for the 4 main antler sections for the NZ red antlers. The standard deviation and the 95% confidence limits are also given; the confidence limits indicate the range within which 95% of NZ red velvet antlers cut at the appropriate stage of growth (ie, rounded/flat top as in A, B and C grades) would be expected to fall. The 3 lower sections all comprised approximately equal portions of the antler, with the tip making up only an average 2.7% of the dry weight. The ash and calcium contents of the 4 sections reflect the very low calcification in the tip of the antler and the increasing degree of calcification towards the base. The lipid (or fat, which is the major component of pantocrin) concentration was highest in the tip but, with the tip being only a very small proportion of the antler, the overall lipid content was determined largely by the other sections. The relatively high lipid content of the base section compared with the midsection is noteworthy.

NZ Red two year old antler and the effect of stage of growth on composition

The mean value data for the composition of velvet antler from 2 year old NZ red deer stags are presented in Table 11 (complete analyses for the 4 main sections, as in Table 10). The lower ash content and the higher lipid content of the 55 day 2 year old antler in comparison with the top grade red antler reflect the earlier stage of maturity at harvest of the 2 year old antler. This is not

unexpected as most top grade NZ velvet from older stags would be harvested 55-65 days after casting to be classified in top A, B or C grades.

There were marked effects of the stage of growth (days after casting) on the comparative composition of velvet antler as reflected in the regression relationships between components and days after casting expressed relative to the 55 day antlers (Table 12). The relative contents of ash (total minerals), calcium and phosphorus increased with days of growth while lipid, sulphur, sodium, potassium and selenium decreased (see Figure 2 for graphical display).

Wapiti and Fallow Deer

Data provide a similar picture to red deer (Tables 13 & 14).

Comparisons between Species

Both fallow and wapiti antlers were higher in nitrogen than the red deer (Table 15) and the fallow antlers were lower in calcium and iron. Otherwise the compositions were remarkably similar. Nitrogen as a percentage of total organic matter varied little between the section of antler within species (Table 16). Calcium as a percentage of total ash was lower in fallow deer but the calcium to phosphorus ratio and the lipid as a percentage of organic matter were similar for all species (Table 17).

Specific composition

Acting on information received from Korea that ganglioside and free amino acid levels might form part or all of quality assurance regulations for imported antler (Hong 1991), relevant assays were set up. The aim was to analyse NZ velvet antler and characterise variability with respect to grade and section.

Gangliosides: Sphingomyelins

Gangliosides are animal glycosphingolipids occurring mainly in the central nervous system but also distributed in other tissues. Within tissues they are located in the outer surface of cell membranes and in the synaptic membranes of the central nervous system. Gangliosides consist of fatty acids,

sphingosine, simple sugars (galactose and glucose), galactosamine and sialic acid. They are thought to be involved with cell metabolism and growth.

We had, from an early stage in this programme concerns that the Korean researchers may not have been measuring true gangliosides. After much research, it is now clear that the two components quantified by the Korean researchers are sphingomyelins (Sm), which are phospholipids. Sm, which are also called ceramide phosphocholines, are structural component of cell membranes. It appears that Sm are involved in complex bioregulatory pathways; for example they are potent inhibitors of protein kinase C which plays an important role in signal transduction in cells. Consequently there are grounds to believe that Sm are biologically active, and it may be that the Sm are very important for velvet antler quality.

The assay method chosen for use was a thin layer chromatography (TLC) based system, similar to that used by the Korean scientists for the assay of "gangliosides" in velvet antler. A laser densitometer was used for the quantification of components separated on the TLC plates. Two sphingomyelin bands of relative fronts (Rf) 0.39 and 0.42, respectively (on TLC plates developed in 60:35:8 chloroform:methanol:0.02% CaCl₂) were identified as gangliosides in a pilot study sponsored by the Koreans as potential "quality control" markers of velvet antler. In this report the bands are referred to as "39" and "42". The sections of the antler referred to are shown in Figure 1.

The antlers assayed were as follows:

- 12 adult NZ red deer velvet antlers from 1991-92 (sections 2 and 8); antlers had been graded A (n=2), B (n=4) or C (n=6) and were processed by freeze drying at Invermay.
- 4 adult NZ red deer velvet antlers graded C from 1991-92 (sections 1, 2, 3, 4, 7 and 8); all were processed by freeze drying at Invermay.
- 12 adult NZ red deer velvet antlers from 1992-93 (section 2); antlers had been graded B
 (n=5) or C (n=7) and were processed by freeze drying at Invermay.

The 42 and 39 bands have been analysed separately and as a combined total. The ratio of 42/39 has also been analysed. The data sets for sections 2 and 8 have been analysed for effects of grade and year. The data sets from sections 1-8 have been analysed for antier section.

A typical TLC plate from an assay of red deer standards, samples and purified standards is shown in Figure 3. The 39 and 42 bands are closest to the bottom of the figure. Four other bands that were not characterised and which are labelled A-D in Figure 3, are also visualised by the cupric acetate staining system. These bands do not appear in the purified standards' lanes.

The standard curves relating concentration of standard to the density of the spot (volume of integration) for the 39 and 42 bands were linear (Figure 4). The standard preparation contained slightly more 42 material than 39. Hence although 1, 3 and 5 µg of the combined standard were spotted on to the TLC plate, each band is only a proportion of the combined standard when examined separately.

Both the 42 and 39 Sm components were present in higher concentrations in Section 2 NZ red deer antlers compared with section 8 (Table 18). The ratio of 42/39 was greater than 1.0 for section 2 and less than 1.0 for section 8. The combined 42 and 39 bands were about 4.5 times higher in section 2 compared with section 8. These results indicate that higher quality sections of antler have higher absolute levels of Sm than lower quality sections. In addition higher quality may be associated with a higher 42/39 ratio.

Band 42 and the combined 42 and 39 bands from Section 2 were significantly higher in 1992-93 NZ red deer antler compared with 1991-92 NZ red deer antlers (Table 19). In contrast band 39 was greater in 1991-92 but this difference was not significant. The 42/39 ratio was significantly greater in 1992-93. This result is somewhat strange in that the 1991-92 sample contained two A graded antlers while the 1992-93 sample was composed of B and C graded antlers. However, we know nothing of the causes of differential Sm levels. It is possible that processing technique, time since processing, time frozen before processing and the feed given to the animal could all influence Sm level. The results are consistent overall with the concept that higher levels of band 42 are accompanied by a high ratio of 42/39.

Four NZ red deer antiers were studied in more detail in that Sm levels were measured in more sections (Table 20). There was a highly significant decrease in all Sm levels from the tip of the antier (Section 1) to the base (Section 8). The tips had the highest combined Sm level of 2.92 mg/g and the bases the lowest (0.36 mg/g). Thus as the antier becomes mineralised Sm levels fall. The ratios of 42/39 in the trez tine (Section 3) and the brow tine (Section 7) are low compared with sections 2 and 4 of the main beam. This may reflect differential development patterns.

When the Sm levels in the 1991-92 sample of NZ red deer antier were analysed in their grades, only one significant difference due to grade was found. That is, the Section 2 B grade antiers contained more of the 39 component than either the A or C graded antiers (Table 21). In all cases the Section 2 levels were higher than Section 8. There was a trend that the Section 8 Sm levels in the C grade antiers were lower in than either A or B graded antiers. The Sm ratio for Section 8 was highest for A grade antier and lowest for C grade, but this was not statistically significant.

When the Section 2 NZ red deer antler Sm data for 1992-93 were added to the comparison between grades, highly significant effects due to year were found and also some main effects of grade (Table 22). For this comparison the A and B grade data from 1991-92 were combined because there were no significant differences between A and B grade and because there were no A graded antlers in the 1992-93 sample. Sm band 39 and the combined 42 and 39 bands were slightly higher for the AB antlers compared with the C grade antlers. This may mean that Sm levels are higher in higher graded antler of the same section, but more information would be necessary before any conclusions could be reached.

Antler data from wapiti were normalised for dry matter. This was necessary because commercially prepared antler differed in water content compared with freeze dried material. Sm levels from Sections 2 and 8 of wapiti antlers are compared with the 1991-92 sample of NZ red deer antler in Table 23. Overall the data strongly reinforce the concept that Section 2 has consistently higher Sm levels than Section 8. There were few significant species differences in Sm level but band 42 levels were higher in NZ wapiti compared with red deer, particularly for Section 2. In general, species

which have low levels of total Sm in Section 2 have higher levels in Section 8. This may reflect different patterns of cell differentiation.

Free Amino Acids

Free amino acids (FAA) are the basic building blocks from which proteins are constructed. They thus constitute essential nutrients for cell growth and, as high concentrations of FAA are found in rapidly growing tissues, should be in abundance in velvet antler. Furthermore, a number of amino acids are termed "essential amino acids" (which means they cannot be synthesised by animals from other materials in their diet) and must be fed to young animals, including humans, so that optimal growth can take place. Velvet antler, as a dietary supplement or traditional medicine, might be expected to provide high levels of some or all of the essential amino acids.

FAA were measured using a reversed phase high performance liquid chromatography procedure. FAA derivatised using phenyl isothiocyanate (PITC) and were determined in all samples analysed in the sphingomyelin assay. Each amino acid quantified in the FAA assay was analysed separately. Where individual acids were not detected, values were treated as zero in the analysis if the derivatised AA was well resolved from other peaks in the chromatogram or as missing if poorly resolved. Some pairs of derivatised FAA were not resolved (alanine/histidine, and ornithine/tryptophan) and were analysed as aggregates.

The total levels of FAA were also analysed, as were the totals of hydrophobic amino acids (leucine, isoleucine, valine, phenylalanine, proline, methionine), of hydrophilic (threonine, serine, hydroxylysine) and of essential amino acids (excluding histidine and tryptophan, ie, arginine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, and valine). All of these data were analysed both as absolute amounts, expressed as nmol/g, and as a percentage of the total FAA. Only absolute data have been presented in the tables for reasons of space.

The various data sets were analysed for the same effects as were the sphingomyelin data sets.

In the 1991-92 velvet antlers a significant effect of section was found on FAA concentrations in sections 2 and 8 (Table 24). These differences were highly significant (p<0.001) for most amino

acids. In all cases, absolute amounts of FAA were lower in section 8 than in section 2, but for some amino acids (particularly taurine) the percentage values showed the opposite trend.

The distribution of FAA in different parts of the antler was examined more fully in four of the 1991-92 antlers (Table 25). When the main beam sections are compared (1, 2, 4 and 8), the levels of most AA decreased from tip to base. However, individual FAA displayed distinctly different patterns of distribution, especially when FAA levels were expressed as percentages. For example, levels of glutamic acid were particularly high in the tips of the antlers both in absolute and in relative terms. Other FAA (eg, glycine, arginine, alanine, isoleucine, taurine and tyrosine) were found in equally high levels in both tips and trez tines. Hydrophobic FAA, on the other hand, were generally found in lower levels in the tip than the main beam or trez tine. The relative amount of hydroxyproline was markedly increased in section 8, while for lysine the peak occurred in section 4. It is highly probable that these differences in distribution patterns are of real physiological significance, and may be related to rates and types of protein synthesis (and possibly protein degradation).

The effect of grade and section (2 and 8) for the 1991-92 NZ red deer antlers (Table 25) showed large effects of antler section for all FAA, and some effects of grade. An effect of antler grade on levels of FAA was also found in the NZ red deer antlers Section 2 comparison (Table 26) when both years data were analysed together. Levels of most amino acids were higher in A and B graded as compared to C graded antlers, and this difference reached statistical significance for alanine/histidine, γ-aminobutyric acid (GABA), isoleucine, lysine, ornithine/tryptophan, proline (and valine). The difference was also significant for the total levels of FAA in these antlers. The magnitude of the difference between grades was often greater in one year than the other, as indicated by the significant interaction between grade and year for some FAA.

This data set also exhibited a highly significant effect of year on total levels of FAA, and on levels of many individual FAA (Table 26). The 1991-92 antlers contained almost 50% more FAA in section 2 than the 1992-93 antlers. Of interest, though, are the observations that levels of the essential amino acids in A and B graded antlers displayed the opposite trend and were higher in 1992/93 than in 1991-92, and were unaffected by year in C graded antlers. In 1992-93 these amino

acids constituted half of the total FAA antlers of all grades. Marked differences were also apparent between years in the ratios of hydrophobic to hydropholic amino acids ("H ratio"), of leucine to isoleucine, and of asparagine to glycine. The causes and potential physiological implications of changes in these quantities are unclear at present, but they may prove to be particularly sensitive indices of changes in antler quality.

FAA levels in antlers from (oven dried) wapiti were compared to the (freeze-dried) 1991-92 red deer antlers (Table 27) after the data from the latter were normalised to zero residual water content. The wapiti and red deer antlers generally had similar FAA concentrations.

Appraisal of sphingomyelins and FAA as quality criteria

In red deer there was a strong linear relationship between total Sm level and total FAA within the antler (Figure 5). This means that parts of the antler which are regarded as being in general of higher quality have higher levels of the components selected by the Koreans as potential quality assurance criteria. However, individual FAA show different patterns of relative concentrations with some FAA having their maximum concentration in the upper portions while others exhibit their maximum concentrations in the base of the antler.

Although higher graded antlers and less mature parts of the antler have higher levels of Sm and FAA, red deer and wapiti antlers are similar. In this study several important variables were not studied: these include stag nutrition, species outside NZ, processing technique and stage of development at harvest. It may be that the SM/FAA data are only relevant in a gross sense as quality criteria but insensitive to some important variable of quality.

Extract production from antiers

Considerable interest has been shown in New Zealand and internationally in development of extracts which concentrate biologically and clinically active substances from velvet antiers. Classically Russian pantocrine is made in a 50:50 v/v mixture of water and alcohol. At Invermay we have developed both aqueous and organic extraction systems. We have measured the effects of several antier variables on yield of these extracts.

The yield of extractable substances from both extracts is higher in the antler tips compared with the bases (Table 28). This is so even from dried velver, probably reflecting a increase in soluble components in these sections. A and B graded antlers had similar yields for both extractions (Table 29). In contrast (Table 30) freeze dried C grade antlers had lower yield of extractables then B grade antlers, but there was no grade difference in antlers commercially processed. Commercial processing using conventional heat drying technology reduced the yield compared with freezedrying for both aqueous and organic extracts. It is likely that heat used in commercial processes reduces soluble components.

Although details of the processes cannot be presented, the data show that both the part of the velvet antier and processing technique affected yields. In contrast grade of antier did not substantially affect yield.

Research on biological effectiveness of organic extracts

Organic extracts from antiers have been tested in an antitumour assay. Samples of interest are incubated for 72 hours with P388 (murine leukaemia) cells. The concentration of sample required to reduce the P388 cell growth by 50% (compared to control cells) is determined. The results are expressed as an ICso in µg of extract/ml of solvent (2:1 dichloromethanol/methanol).

Biological activity in the P388 assay decreases from upper sections to lower sections (ie, it requires a higher concentration of extract to have the same inhibitory effect; Table 31). Enigmatically in these studies lower grade antler had higher activity than A or B graded antlers (Table 32). Processing technique also influences activity (Table 33). These results indicate that there is potent anti-tumour activity in organic extracts from NZ red velvet antler and this is influenced by section of the antler, grade and processing technique. Data, however, are highly variable both between and within antlers. It is now important to carry out further studies on the reasons for this variability and whether stags consistently produce antlers of higher activity than others. Ways of handling and processing antlers to maximise activity must also be sought and optimal systems developed.

Research on biological effectiveness of aqueous extracts

The aqueous extracts described above have been evaluated for biological effectiveness by measuring their effects on growth of antier cells in culture. The method used for the antier cell culture has been published (Sadighi et al 1994) but a synopsis of the method, with the specific variations required to evaluate antier extracts, is given below.

Velvet antiers were obtained under normal husbandry conditions from farmed red deer (Cerus elaphus) stags 60 days after casting of the previous hard antier. The epidermis and dermis were dissected from the underlying tissues and sections from the most proliferative zone, 0.75 cm distalt to the tip, and the maturation zone, 0.75-1.5 cm distalt to the tip, were dissected free of dermis and loose connective tissue with a sterile scalpel. The cells are referred to as fibroblast zone (line x) and cartilage zone (line y) respectively. The tissue sections were transferred to Petri dishes and rinsed with a balanced salt solution and the cells were desegregated.

The dispersed cells were seeded in tissue culture flasks in medium containing Fitton-Jackson Modification (BGJs, 45%), F₁₂ nutrient (45%), fetal bovine serum (FBS; 10%) penicillin (100 U/ml), and streptomycin (100 μg/ml), at a density of 10⁴ cell/cm². They were incubated in a humidified incubator in 95% air and 5% CO2 at 37°C. The medium was changed every 3 days. Cells were transplanted on reaching confluence (every 2 weeks) using trypsin-EDTA (Sigma) to lift them from the dish. Cells used in the present study had been through two passages. Cell viability was measured using Trypan blue and was always above 85%. Aliquots of cells were frozen in liquid nitrogen in media containing BGJs (40%), F₁₂ nutrient (40%), FBS (10%), penicillin (100 U/ml), streptomycin (100 μg/ml) and dimethyl sulphoxide (Sigma; 10%). Prior to use, cells were taken out of the liquid nitrogen and put straight into a 37°C waterbath to thaw. Cells were counted in a hemocytometer.

To evaluate the proliferative activity of aqueous antler extracts, x and y zone cells were seeded in 24-well plates at a density of $2x10^4$ cell/cm². The cells were incubated for 48 h after which time the medium was changed to either 10% FBS or to serum-free media (SFM) and the cells were incubated for a further 24 h. This was followed by a 24-h incubation in 10% FBS or SFM to which varying doses of antler extract had been added.

The results of each experiment are the means of triplicates performed at the same time.

In the first series of studies extracts were made from whole combined A and B grade NZ red velvet antlers. The dose response of the extracts on mitogenicity of antler cells from the x and y lines (Figure 6) revealed that there was a peak at 1.25 - 2.5 mg/ml after which mitogenicity declined. In a second series of studies separate extracts of upper (1-4) middle (5) and lower (6 and 8) antler sections were examined in the x cell line only (Figure 7). This showed that the mitogenicity of upper and mid extracts exhibited a different response pattern to that of extracts from lower sections. Peak mitogenicity occurred at 0.125 mg/ml for both the upper and mid extracts but at 2.5 mg/ml for the lower extract. Overall, peak mitogenicity occurred at lowest concentrations for the mid section extracts.

These results, taken together, reveal that there are potent stimulators of cell division in aqueous antler extracts. Also extracts from different antlers and different antler sections vary widely in potency. Exploring the precise reasons for these effects represents a major, but exciting challenge for the research group.

HARVEST TECHNOLOGY AND COLOUR

In the Korean marketplace New Zealand velvet is noted for its rich dark red colour which contrasts to Russian velvet. We have begun a preliminary investigation into the factors that influence colour of processed velvet antler. While these studies are not sufficiently complete to present detailed results it must be stated that removal technique appears to play only a minor role in determining colour. Much more important is the processing technique, particularly the processing temperature. We consider in the future that antler processing will become more specific and techniques will vary according to the requirements of end users. Traditional users of cut antler slices may require a very different process compared with makers of the various antler extracts.

RESEARCH ON MECHANISMS OF ANTLER GROWTH

There has been a considerable effort at AgResearch Invermay since 1981 on the physiological mechanisms underlying velvet antler growth. The approaches have included the role of steroids and growth factors (both in vivo and in vitto), the role of genes in controlling antler growth, and the role of blood supply. A full review of this work is outside the scope of this manuscript but the majority of this research is already published (Fennessy and Suttie 1985, Suttie and Fennessy 1990, Suttie et al 1989, Sadighi et al 1994, Li and Suttie 1993, Suttie et al 1994).

CONCLUSIONS AND OVERALL SUMMARY

Since deer farming began in New Zealand, integrated research has accompanied and contributed to the rapid advances of the industry. Although much of the earlier work was of a production and a mechanistic nature, the direction and scope has altered over the years and will continue to do so. Since the late 1980's, the demands of the developing velvet antler industry have dictated that a fuller evaluation of value-added product development should take place. In parallel to this approach has been the need to develop accurate and sensitive indicators of antler quality and the factors which influence antler quality. A decision was made at the outset of our research that the performance of clinical trials would need to wait until repeatable procedures for the production of biologically active preparations could be developed. The products must be free from microbiological contamination, safe for the user and likely to be effective. This point has now been reached.

Taking our research of almost 20 years altogether, our key achievement has been to provide the necessary information to support the initiation, development and maturation of a new animal industry in New Zealand and to take a product of this industry to the point where it is now ready for clinical testing. We have been able to help build up a deer industry that is based on quality, and which combines New Zealand's twin advantages of a wide genetic stock of deer and a cool, clean, unpolluted environment. We have not attempted to artificially manipulate antier growth but rather have developed systems of targeted feeding to ensure genetic potential growth is attained. The cell culture systems we use to measure one aspect of biological activity come directly from our studies

of the mechanisms of antler growth: to understand the product you must know how it grows and vice versa. Overall our antler research is aimed at producing, maintaining and enhancing antler quality.

Quality, as it applies to the antler, needs some definition. Much of the early research in New Zealand concentrated on quantity as the sole indicator of quality. However, our research has shown that this is not necessarily the case. There are many effects on quality which are quite distinct from quantity. It may be that there are several indicators of quality. What is quality for traditional dried product may differ substantially from quality for extract production. It is not known how feeding, temperature, time of harvest or processing actually effects quality. It may be that no single chemical or biological indicator of quality is sufficiently reliable or sensitive to measure quality. Our research programme, because it begins with the stag and ends with a value-added product and its use, puts us in a unique position internationally to follow at all stages the factors which determine quality. This is our key competitive advantage in antler research today.

Our programme while complete in some aspects has proceeded at such a rapid pace that some gaps have appeared and some key unknowns have emerged. We do not fully understand the interactions between species, location and feeding on antler quality. Although these have each been researched individually no integrated study has taken place. We note variability from year to year in Sm and FAA level in the same stags, but have no clues as to why this happens. Although Sm and FAA crudely correlate with the part of the antler we do not know whether these actually explain specific or unique biological or clinical activity. To arrive at meaningful quality standards, answers to these and many more questions must be found. Invermay is uniquely placed to find them.

Research has played a role in the Deer Industry in New Zealand since its outset. The programme currently advances at pace and moves into new areas. The success of our integrated approach to deer - from the pasture via the animal, to the product, to the value added product and, soon, to the consumer - has brought considerable benefits to New Zealand. The future is exciting and carries with it the potential to bring benefits to numerous health-conscious people in various parts of the world.

REFERENCES

- Anon (1976) New Zealand Atlas. A.R. Shearer, Government Printer, Wellington.
- Anon (1979) Plant and Analytical Chemistry Group Bulletin. Ruakura Soil and Plant Research Station, Hamilton.
- Anon (1986) Climate of New Zealand. NZ Meteorological Service, Information Publication 15, Wellington.
- Anon (1994) New Zealand Official Yearbook. Statistics New Zealand, Wellington.
- Elworthy, P. (1976) Production of velvet. New Zealand Deer Farming Annual 1976. NZDFA, P21.
- Fennessy, P.F. (1981) Antler growth in deer. Proc. Deer Course for Veterinarians, Queenstown, 17-21.
- Fennessy, P.F. (1989) High returns from velvet antler. Proc. Deer Industry Conference, Ruakura Agricultural Centre, 21-25.
- Fennessy, P.F. and Pearse, A.J. (1990) The relative performance of Canadian wapiti and their hybrids. Proc. Aust. Assoc. Anim. Breed. Genet. 8, 498-500.
- Fennessy, P.F. and Suttie, J.M. (1985) Antler growth: nutritional and endocrine factors. <u>In</u> Biology of Deer Production, P.F. Fennessy and K.R. Drew (eds). <u>Royal Soc. of N.Z. Bull.</u> #22, Wellington, 239-250.
- Fennessy, P.F., Corson, I.D. and Moore, G.H. (1981) Energy requirements of red deer. Proc. N.Z. Soc. Anim. Prod. 41, 167-173.

- Hong, N-D. (1991) Studies on the components and analysis method of Cervi spp. Unpublished Report. Kyung-Hee Medical Center.
- Li, C. and Suttie, J.M. (1993) Pedicle and antler development following sectioning of the sensory nerves to the antlerogenic region of red deer (*Cerrus elaphus*). J. Exp. Zool. <u>267</u>, 188-197.
- Moore, G.H. (1984) Removal of antler in velvet: Time, method, grades. Proc. N.Z. D.F.A. Conf. 9, 21-33.
- Moore, G.H., Littlejohn, R.P. and Cowie, G.M. (1988) Liveweights, growth rates and antler measurements of farmed red deer stags and their usefulness as predictors of performance. N.Z. J. Agric. Res. 31, 285 – 291
- Pollock, A.M. (1975) Seasonal changes in appetite and sexual condition in red deer stags maintained on a six-month photoperiod. J. Physiol. Lond. 244, 95-96.
- Sadighi, M., Haines, S.R., Skottner, A., Harris, A.J. and Suttie, J.M. (1994) Effects of insulin-like growth factor 1 (IGF 1) and IGF 11 on the growth of antler cells *in vitro*. J. Endocrinol. (In Press).
- Suttie, J.M. (1992) Good nutrition is as good as gold. The Deer Farmer, July, 16-17.
- Suttie, J.M. and Fennessy, P.F. (1990) Recent advances in the physiological control of velvet antler growth. <u>In</u> The Biology of Deer, R.D. Brown (ed), Springer-Verlag, New York, 471-486.
- Suttie, J.M., Goodall, E.D., Pennie, K. and Kay, R.N.B. (1983) Winter food restriction and summer compensation in red deer stags. Brit. J. Nutr. 50, 737-747.

- Suttie, J.M., Fennessy, P.F., Corson, I.D., Lazs, F.J., Crosbie, S.F., Butler J.H. and Gluckman, P.D. (1989) Pulsatile growth hormone, insulin-like growth factors and antier development in red deer (Cervus elaphus scoticus) stags. J. Endocrinol. 121, 351-360.
- Suttie, J.M., Li, C, Sadighi, M., Gunn, J. and Fleming, J.S. (1994) Physiological control of antler growth. Proc. 3rd Biology of Deer Symposium (In Press).
- Wallis, T. (1976) Some points on harvesting velvet. New Zealand Deer Farming Annual 1976, NZDFA P22.

Table 1: Summary of climate information at selected stations. The mean daily temperature is the average of the maximum and minimum temperature for a given day. A ground frost occurs when the grass minimum thermometer (25 mm above short grass) reads -1.0°C or lower (NZ Meteorological Service 1986).

	Height Above Sea Level (m)						No of	
		Mean Jan	Daily July	Daily Jan	Max July	Daily Jan	Min July	Ground Frosts
North Island								
Taupo Wellington	376 126	17.3 16.4	6.5 8.2	23.5 20	11 10.9	11 12.8	1.9 5.5	71
South Island						12.0	3.5	15
Queenstown Dunedin	329 2	15.8 15	3.7 6.4	21.7 19.0	7.7 9.9	9.9 11.1	-0.4 2.9	141 78

Table 2: Effect of stage of antler at harvest on yield in kg (from Moore 1984).

		Stage of Royal Tine	
	Developing	0.25 cm long	0.5 cm long
Velvet weight (kg)	2.20	2.54	2,58

Table 3: Velvet antler production (kg) by age in herds of red deer stags. In herd A 30% of stags were sold as 2 year olds but no culling took place from herds B or C (from Fennessy 1989).

		Herd	
Age (years)	Α	В	С
ber of stags	1.43 2.05 2.60 2.96 301	1.42 2.03 2.50 2.85 49	1.01 1.60 1.93 2.32 36

Table 4: Effect of level of nutrition at different stages of the antier cycle on subsequent velvet antier production (After Fennessy 1989).

			Season	
	Autumn (Post rut)	Winter	Late Winter	Spring (Antler Growth)
Number of trials	1	4		
Days of trial	50	80	2 50	1 65
Velvet antier yield (kg/stag) for				
stags fed at different levels	2.70	1.80	2.06	2.20
Difference (%)	2.40	1.66	1.94	1.87
	13	. 8	6	. 18

Table 5: Effect of feeding diets containing only soluble or protected protein sources to 3 year old stags during winter on subsequent antler weight and stag liveweights. Antler weight is adjusted for velvet cutting date. Enerpro is a diet containing high levels of groundnut meal as a protein source.

	Diet					
	Soluble protein	Protected protein	Enerpro			
Winter liveweight (kg)	138	137	138			
Spring liveweight (kg)	144	146	145			
Ander weight (kg)	2.1	1.9	2.0			

table 6: vervet after production (kg) by age for groups of stags categorised according to their two year old velvet antier production (n=90) (from Fennessy 1989).

Rank of two year old velvet production	Velve	Cumulative (2-			
	2	3	4	5	5 years)
Top 1/6 Next 1/3 Next 1/3 Bottom 1/6	2.18 1.55 1.29 1.02	2.62 2.22 2.08 1.89	2.99 2.46 2.25 2.11	3.58 3.12 2.85 2.67	11.37 9.35 8.47 7.69
Overall mean ± Std deviation (SD)	1.48 0.43	2.18 0.42	2.42 0.47	3.03 0.56	9.11 1.60

Table 7: Progeny test of five red sire stags: comparison of the mean cumulative velvet ander production (two to five years of age) and the three year old winter lean liveweight for the progeny groups.

Sire	Number of male	Deviations from average (kg)			
	progeny	Cumulative velvet antler weight	3 year old winter lear liveweight		
Α	29	-0.46			
В	32		+4.6		
Ċ		-0.45	-1.4		
-	22	-0.27	-0.7		
D	17	÷0.37	+4.6		
E	35	+0.61			
		+0.01	+2.0		
Whole herd	301	Means: 9.11	126		

Table 8: Typical velvet antler yields (kg; and days of growth) for NZ red (NZR), Canadian wapiti (CW) x NZR hybrids, and CW at Invermay and the expected velvet antler weight for CW² assuming no hybrid vigour and the ratio of actual to expected velvet antler weight for CW.

	Actual velvet antier weights			Expected velvet antier weight	Actual/expected velvet weight
	NZR	CW X NZR	CW	of CW ¹	for CW
2y 3y	1.01 (55) 1.60 (58)	2.05 (64) 2.63 (64)	1.99 (68) 2.50 (71)	3.09 3.66	0.64 0.68

NZR = NZ red deer, CW = Canadian wapiu

² Expected velvet antler weight of CW = 2 ($CW \times NZR$)-NZR

Table 9: Species, number and description of antler used in mineral and lipid compositional studies. Species

Species	Number	Description
NZ top grade (adult red deer)	17	10 dried commercially 7 frozen
Wapiti	6	2 dried commercially 4 frozen
Fallow	6	6 dried commercially
2 year old red deer	6 pairs	One antler of each pair was cut at 55 days and the

contra-lateral antier cut at 43 to 67 days; all frozen

Table 10: Composition of adult NZ red deer velvet ander (n=17): mean values with standard deviation (SD) and the 95% confidence limits for the components in each of the 4 main sections of the antler and the combined total.

		Antler	section		
	Tip (100)	Upper (200)	Mid (300)	Base (400)	Complete (100 + 200 + 300 - 400)
Mean values % of total dry weight in section	2.7	35.3	29.8	32.5	100
% of DM in processe ander!	d 81.1	85.5	85.9	86.2	84.7
Components (as %	of DM; ± SD with ra	inges being the 95%	confidence limits)		
Ash	6.6	28.4	37.8	38.8	34.0
	(0.75; 5.0-8.2)	(2.43; 23.2-33.5)	(2.62; 32.2-43.4)	(2.28; 33.9-43.6)	(1.96; 29.9-38.2)
Lipid	5.58	2.65	1.98	2.57	2.50
	(1.32; 2.78-8.38)	(0.69; 1.19-4.11)	(0.47; 0.99-2.98)	(0.65; 1.19-3.94)	(0.56; 1.31-3.70)
Nivogen (N)	12.2	9.1	8.1	7.6	8.4
	(0.63; 10.9-13.5)	(0.52; 8.0-10.2)	(0.63; 6.8-9.5)	(0.61; 6.3-8.9)	(0.51; 7.3-9.5)
Calcium (Ca)	0.29	9.3	13.5	14.7	12.1
	(0.22; 0-0.76)	(1.02; 7.2-11.5)	·(1.15; 11.0-15.9)	(1.76; 10.9-18.4)	(1.11; 9.7-14.4)
Phosphorus (P)	0.64	5.0	6.3	6.5	5.8
	(0.11; 0.41-0.88)	(0.75; 3.4-6.5)	(0.51; 5.2-7.4)	(0.36; 5.7-7.3)	(0.32; 5.1-6.4)
Sulphur (S)	0.85	0.54	0.35	0.34	0.43
	(0.11; 0.62-1.08)	(0.026; 0.48-0.60)	(0.030; 0.29-0.41)	(0.035; 0.27-0.42)	(0.030; 0.37-0.49)
Magnesium (Mg)	0.052	0.21	0.27	0.28	0.25
	(0.011; 0.029-0.075)	(0.024; 0.16-0.26)	(0.021; 0.23-0.32)	(0.023; 0.24-0.33)	(0.019; 0.21-0.29)
Sodium (Na)	1.09	0.90	0.80	0.77	0.83
	(0.14; 0.77-1.42)	(0.08; 0.73-1.07)	(0.040; 0.71-0.88)	(0.045; 0.67-0.86)	(0.039; 0.75-0.91)
Potassium (K)	0.91	0.59	0.33	0.29	0.42
	(0.12; 0.66-1.16)	(0.06; 0.46-0.72)	(0.055; 0.21-0.44)	(0.037; 0.22-0.37)	(0.43; 0.33-0.51)
Trace mineral comp	onents (as mg per k	g of DM; ± SD with	ranges being the 95	% confidence limits)	
Manganese (Mn)	2.6	3.2	3.4	3.5	3.4
	(1.4; 0-5.6)	(0.8; 1.6-4.8)	(0.6; 2.1-4.7)	(0.8; 1.8-5.2)	(0.4; 2.4-4.3)
Zinc (Zn)	46 (7.9; 29-63)	72 (9.1; 53-92)	67 (10.1; 46-89)	68 (12.0; 43-94)	(9.2; 49-88)
Copper (Cu)	5.2	5.1	5.6	5.3	5.3
	(1.1; 2.9-7.5)	(0.7; 3.7-6.5)	(0.8; 3.9-7.3)	(0.8; 3.7-6.9)	(0.5; 4.3-6.3)
Iron (Fe)	462 (227; 0-943)	472 (92; 277-667)	288 (100; 77-499)	179 (53; 67-291)	· 319 · (69; 173-465)
Selenium (Se)	0.35	0.25	0.14	0.13	0.18
	(0.12; 0.10-0.59)	(0.09; 0.06-0.44)	(0.06; 0.02-0.26)	(0.05; 0.02-0.24)	(0.07; 0.04-0.32)
Cobalt (Co)	0.05 (0.05; 0-0.16) processed anders; DN	0.04 (0.06; 0-0.17)	0.03 (0.03; 0-0.09)	0.03 (0.03; 0-0.09)	0.04 (0.03; 0-0.11)

Table 11: Composition of NZ red deer velvet ander (n=6) from 2 year old stags banvested at 55 days after bard ander casting.

		Antle	r section		
	Tip (100)	Upper (200)	Mid (300)	Base (400)	Complete (100 + 200 + 300
Mean values % of total dry we	ight				400)
in section				•	100
Components (as	% of DM; ± SD with r	anges being the 050	confidence timites	•	
Ash	8.0				
••••	(1.10; 5.2-10.9)	24.9 (3.66; 15.5-34.3)	35.2 (2.12; 29.8-40.7)	38.8 (1.49; 35.0-42.6)	33.1 (1.24; 29.9-36.3)
Lipid	6.33 (0.75; 4.39-8.26)	3.82 (0.51; 2.50-5.13)	2.68 (0.27; 1.99-3.37)	2.31	₹ 3.16
Nitrogen (N)	11.8			(0.53; 1.44-4.18)	(0.49; 1.90-4.42)
Calcium (Ca)	(0.24; 11.2-12.4)	8.7 (0.74; 6.8-10.6)	7.5 (0.25; 6.8-8.1)	7.2 (0.41; 6.2-8.2)	7.8 (0.17; 7.4-8.2)
	0.076 (0.059; 0-0.23)	7.9 (1.64; 3.7-12.2)	12.2 (0.72; 10.3-14.0)	13.5 (1.12; 10.6-16.3)	11.2
Phosphorus (P)	0.75 (0.06; 0.60-0.90)	3.7 (0.79; 1.6-5.7)	6.2 (0.47; 5.0-7,4)	6.4	(0.63; 9.6-12.8) 5.4
Sulphur (S)	0.74		•	(0.91; 4.0-8.7)	(0.50; 4.1-6.7)
Moonadian O.C.	(0.04; 0.63-0.85)	0.54 (0.13; 0.20-0.88)	0.37 (0.03; 0.30-0.46)	0.34 (0.03; 0.27-0.41)	0.42 (0.032; 0.34-0.50)
Magnesium (Mg)	0.063 (0.008; 0.042-0.084)	0.19 (0.029; 0.12-0.27)	0.25 (0.024; 0.19-0.31)	0.27 (0.023; 0.21-0.33)	0.24
Sodium (Na)	1.12	1.01	0.76		(0.019; 0.19-0.29)
Potassium (K)	(0.11; 0.83-1.41) 1.13	(0.14; 0.65-1.36)	(0.02; 0.71-0.81)	0.71 (0.03; 0.62-0.80)	0.82 (0.046; 0.70-0.94)
	(0.12; 0.81-1.44)	0.73 (0.11; 0.44-1.01)	0.41 (0.07; 0.23-0.58)	0.30 (0.03; 0.22-0.38)	0.47 (0.037; 0.37-0.56)
race mineral con	nponents (as mg per kį	g of DM; ± SD with	ranges being the 959	o confidence limits)	(4-42-7) (0.57-0.50)
Manganese (Mn)	2.1	3.0	3.1		
Zinc (Zn)	(1.0; 0-5.3) 40	(0.7; 1.2-4.9)	(1.0; 0.7-5.6)	3.2 (0.9; 1.1-5.4)	3.1 (0.7; 1.3-5.0)
Copper (Cu)	(3.7; 30-49)	56 (7.0; 38-74)	58 (11.0; 29-86)	59 (3.4; 50-67)	57 (4.6; 46-69)
	5.3 (0.8; 3.2-7.4)	4.3 (0.4; 3.4-5.3)	3.3 (0.3; 2.5-4.1)	3.1 (0.2; 2.5-3.7)	3.6
ron (Fe)	432 (139; 76-788)	399 (33; 316-483)	288 (89; 59-518)	142	(0.3; 2.8-4.4) 267
Gelenium (Se)	0.29	0.24		(32; 61-224)	(35; 177-357)
Cobalt (Co)	(0.03; 0.22-0.37)	(0.02; 0.20-0.29)	0.16 (0.02; 0.10-0.21)	0.11 (0.02; 0.07-0.14)	0.16 (0.01; 0.14-0.19)
	0.03 (0.01; 0-0.06)	0.02 (0.02; 0-0.06)	0.02 (0.02; 0-0.06)	0.02 (0.01; 0-0.05)	0.02 (0.01; 0-0.05)

Table 12: Significant regression relationships between velvet antier components (relative concentration) and days of growth expressed relative to controlateral antier cut at 55 days after hard antier casting.

				cr casuity.
	Regression Intercept coefficient	L²	SE of regression coefficient	Mean concentration of component in DM
Positive relationsh	ips (increasing with time)		Tomponent III Div
Relative ash Relative calcium Relative phosphorus	= 0.0077 Days + 0.60 = 0.0128 Days + 0.27 = 0.0088 Days + 0.49	0.93** 0.46(*) 0.57*	0.0010 0.0056 0.0032	33.1% 11.2% 5.4%
Negative relationsh	ips (decreasing with time			
Relative lipid Relative sulphur Relative sodium Relative potassium Relative selenium	= -0.0132 Days + 1.69 = -0.0142 Days + 1.79 = 0.0062 Days + 1.36 = 0.0154 Days + 1.86 = -0.0123 Days + 1.65	0.95** 0.87** 0.67* 0.64* 0.70*	0.0014 0.0024 0.0019 0.0049 0.0035	3.16% 0.42% 0.82% 0.47% 0.16 mg/kg

Statistical significance levels: (*) P<0.10; *P<0.05; **P<0.01.

Table 13: Composition of NZ wapiti velvet antler (n=6): mean values with standard deviation (SD) and the 95% confidence limits for the components in each of the 4 main sections of the antler.

		An	tler section		
	Tip (100)	Upper (200)	Mid (300)	Base (400)	Complete (100 + 200 + 300
Mean values % of total dry w in section		43.8	23.6	29.8	400)
% of DM in pro- ander	cessed 89.1	90.1	91.5	91.4	100
Components (as	% of DM; ± SD with				
Ash		ranges being the 95	જ confidence limits)	1	
Lipid	7.4 (2.80; 1.5-13.4)	28.0 (5.45; 15.8-39.0)	20 9 .	20.7	34.0
Libia	4.28 (1.32; 1.46-7.10)	2.50 (0.54; 1.34-3.65)	1.70	2.21	(2.70; 28.2-39.7)
Nitrogen (N)	13.4	10.2	(0.39; 0.86-2.54) 8.8	17 3.00)	(0.21; 1.77-2.66)
Calcium (Ca)	(1.52; 10.2-16.7)	(1.19; 7.7-12.8)	(1.09; 6.5-11.1)	8.6 (0.92; 6.6-10.5)	9.4 (0.81; 7.7-11.2)
Phosphorus (P)	(1.03; 0-2.78) 0.77	9.16 (2.42; 4-14.32)	13.97 (1.72; 10.3-17.63)	14.54 (1.48; 11.4-17.70)	12.0 (0.77; 9.7-14.3)
Sulphur (S)	(0.48; 0-1.79)	4.60 (1.36; 1.7-7.49)	6.81 (0.64; 5.5-8.16)	6.96 (0.60; 5.7-8.23)	5.9
	0.83 (0.12; 0.58-1.07)	0.57 (0.18; 0.18-0.96)	0.31 (0.03; 0.24-0.39)	0.30	(0.47; 4.9-6.9) 0.41
Magnesium (Mg)	0.06 (0.02; 0.01-0.10)	0.19	0.26	(0.05; 0.18-0.41) 0.28	(0.031; 0.340.47)
Sodium (Na)	1.23	(0.03; 0.12-0.26) 1.05	(0.04; 0.17-0.36) 0.78	(0.04; 0.20-0.36)	0.24 (0.030; 0.17-0.30)
Potassium (K)	(0.17; 0.88-1.59) 0.97	(0.27; 0.49-1.61)	(0.07; 0.63-0.93)	0.76 (0.09 0.47-0.95)	0.86 (0.075; 0.70-1.02)
Trace mineral com	(0.21; 0.53-1.40)	0.64 (0.19; 0.24-1.04)	0.27 (0.06; 0.16-0.39)	0.27 (0.06; 0.15-0 30)	0.42
	nponents (as mg per kg	of DM; ± SD with	ranges being the asc	7 71	(0.031; 0.35-0.49)
winniganese (Mn)	1.18 (0.98; 0-3.9)	2,4	3.1		
Zinc (Zn)	45	(0.48; 1.3-3.4) 73	(0.19; 2.7-3.5)	3.2 (0.41; 2.3-4.0)	2.8 (0.2; 2.3-3.3)
Copper (Cu)	(13.4; 16-74) 5.0	(12.4; 46-99)	69 (8.7; 50-87)	72 (6.8; 57-86)	71 (6.5; 57-85)
ron (Fe)	(2.1; 0.5-9.5)	5.1 (1.3; 2.3-8.0)	5.0 (0.7; 3.5-6.5)	4.7 (0.5; 3.6-5.8)	4.8
elenium (Se)	546 (253; 6-1085)	467 (131; 187-744)	241 (116; 0-488)	161 (56; 42-280)	(0.6; 3.5-6.1)
	0.29 (0.11; 0.07-0.52)	0.21 (0.74; 0.05-0.37)	0.12 (0.04; 0.03-0.21)	0.10	(74; 171-488) 0.16
Cobalt (Co)	0.05 (0.08; 0-0.22)	0.02	0.01	(0.04; 0.02-0.19) 0.02	(0.06; 0.03-0.29)
	, 0 0.22)	(0.02; 0-0.06)	(0.01; 0-0.03)	(0.01; 0-0.04)	0.02 (0.02; 0-0.05)

Table 14: Composition of NZ fallow deer antler (n=6): mean values with standard deviation (SD) and the 95% confidence limits for the components in each of the 4 main sections of the antler.

		Antler s	section		
	Tip (100)	Upper (200)	Mid (300)	Base (400)	Complete (100 + 200 + 300 + 400)
Mean values % of total dry weight in section	8.9	25.3	26.5	. 39.3	100
% of DM in processe untler	d 85.6	87.0	86.8	86.9	
Components (as % o	of DM; ± SD with re	inges being the 95%	confidence limits)		
Ash	10.1	30.4	35.9	37.0	32.6
	(3.64; 2.38-17.90)	(2.09; 25.91-34.79)	(1.\$8; 31.87-39.\$8)	(2.68; 31.26-42.67)	(1.59; 29.3-36.0)
Lipid	5.81	2.68	2.06	2.50	2.70
	(1.51; 2.60-9.02)	(0.71; 1.18-4.18)	(0.31; 1.40-2.73)	(0.61; 1.19-3.81)	(0.47; 1.71-3.70)
Nitrogen (N)	12.7	9.60	9.20	9.00	9.5
	(1.06; 10.46-14.97)	(0.99; 7.51-11.72)	(0.63; 7.88-10.55)	(0.37; 8.17-9.74)	(0.32; 8.8-10.2)
Calcium (Ca)	1.41	9.13	. 11.93	12.47	10.5
	(1.43; 0-4.48)	(0.70; 7.64-10.63)	(0.95; 9.91-13.96)	(1.51; 9.25-15.69)	(0.96; 8.5-12.6)
Phosphorus (P)	0.96	0.64	0.40	0.35	5.4
	(0.62; 0-2.61)	(1.256; 2.24-7.57)	(0.44; 4.496-6.81)	(0.49; 5.13-7.21)	(0.37; 4.6-6.2)
Sulphur (S)	0.79	0.56	0.37	0.35	0.45
	(0.05; 0.69-0.89)	(0.12; 0.30-0.81)	(0.04; 0.29-0.45)	(0.05; 0.25-0.46)	(0.041; 0.36-0.53)
Magnesium (Mg)	. 0.09	0.23	0.27	0.29	0.25
	(0.039; 0.01-0.17)	(0.021; 0.19-0.28)	(0.030; 0.21-0.33)	(0.051; 0.18-0.40)	(0.033; 0.18-0.32)
Sodium (Na)	0.96	0.64	0.40	0.35	0.81
	(0.15; 0.76-1.39)	(0.20; 0.50-1.33)	(0.09; 0.54-0.93)	(0.10; 0.51-0.96)	(0.098; 0.60-1.01)
Potassium (K)	0.96	0.64	0.40	0.35	0.49
	(0.11; 0.73-1.20)	(0.08; 0.48-0.80)	(0.07; 0.26-0.54)	(0.09; 0.17-0.54)	(0.053; 0.38-0.60)
Trace mineral comp	onents (as mg per k	g of DM; ± SD with	ranges being the 959	6 confidence limits)	
Manganese (Mn)	3.0	3.4	3.0	3.3	3.2
	(1.0; 0.9-5.1)	(0.5; 2.3-4.6)	(0.6; 1.8-4.2)	(0.5; 2.2-4.3)	(0.3; 2.6-3.9)
Zinc (Zn)	56	73	70	68	. 70
	(11.6; 31-81)	(7.7; 57-90)	(16.2; 36-105)	(9.7; 47-89)	(9.5; 49-89)
Copper (Cu)	4.6	5.1	5.0	4.9	5.0
	(0.5; 3.4-5.7)	(0.7; 3.7-6.6)	(0.6; 3.8-6.2)	(0.2; 4.5-5.4)	(0.3; 4.3-5.7)
Iron (Fe)	198	289	240	152	216
	(71; 46-349)	(74; 131-4+8)	(82; 67-414)	(46; 55-249)	(58; 92-340)
Selenium (Se)	0.31	0.23	0.17	0.13	0.18
	(0.03; 0.24-0.38)	(0.02; 0.18-0.28)	(0.02; 0.13-0.21)	(0.02; 0.09-0.17)	(0.02; 0.13-0.23)
Cobalt (Co)	0.06	0.0 1	0.01	0.01	0.02
	(0.06; 0-0.20)	(0.02; 0-0.05)	(0.01; 0-0.04)	(0.01; 0-0.04)	(0.01; 0-0.05)
			•		

Table 15: Comparisons of mean values for components (= SE) in the complete velvet ander between other species and NZ red deer (*, p<0.05; **, p<0.01; ***, p<0.01; otherwise comparisons not significantly different).

	Red n=17			Wapiti n=6		llow =6
	Mean	SE	Mean	SE	Mean	SE
Dry matter	88.9	1.03	90.8	1.62	86.3	0.37
Components (as % o	of DM)					0.57
Ash	34.0	0.49	34.0	1.10	32.6	0.60
Lipid	2.51	0.14	2.22	0.09	2.70	0.13
Nitrogen (N)	8.4	0.13	9.4	0.33***	9.5	0.13
Phosphorus (P)	5.7	0.08	5.9	0.19	5.4	0.14
Sulphur (S)	0.43	0.01	0.41	0.01	0.45	0.02
Magnesium (Mg)	0.25	0.01	0.24	0.01	0.25	0.01
Calcium (Ca)	12.1	0.28	12.0	0.44	10.5	0.36**
Sodium (Na)	0.83	0.01	0.86	0.03	0.81	0.04
Potassium (K)	0.42	0.01	0.42	0.01	0.49	0.02***
Trace mineral comp	onents (as m	g per kg of I	OM)			
Manganese (Mn)	3.4	0.11	2.8	0.10**	3.2	0.12
Zinc (Zn)	69	2.3	71	2.7	69	3.6
Copper (Cu)	5.3	0.12	4.8	0.25	5.0	0.13
iron (Fe)	320	17	330	30	216	22**
Selenium (Se)	0.18	0.02	0.16	0.02	0.18	0.01
Cobalt (Co)	0.01	0.01	0.02	0.01	0.02	0.01

Table 16: Nitrogen as a percentage of total organic matter in velvet antler from three species (mean \pm SD).

		N as % of	Total OM	
		Ander	section	
	Tip (100)	Upper (200)	Mid (300)	Base (400)
Red Wapiti Fallow	13.1 ± 0.61 14.5 ± 1.50 14.1 ± 0.82	12.7 ± 0.88 14.1 ± 1.35 13.8 ± 1.07	13.1 ± 0.94 14.6 ± 1.51 14.4 ± 0.93	12.5 ± 0.92 14.1 ± 1.07 14.2 ± 0.95

Table 17: Composition of velvet antler from three species (mean \pm SD) for selected components (*, p<0.05, compared with NZ red deer).

	N as % of total OM	Ca as % of total ash	Ca:P ratio	Lipid as % of total OM
Red	12.8 ± 0.76 14.3 ± 1.31 14.1 ± 0.60	34.5 ± 1.64	2.1 ± 0.14	3.8 ± 0.79
Wapiti		34.5 ± 0.85	2.0 ± 0.08	3.4 ± 0.32
Fallow		30.8 ± 1.57*	1.9 ± 0.14	3.9 ± 0.66

Table 18: Effect of section, (Section 2 or Section 8) on sphingomyelin levels (mg/g) and ratio in adult NZ red deer velvet antlers (n=12) (1991-2). SED is the standard error of the difference; **, p<0.01; ****, p<0.001.

Band -	Sec	tion	
	2	8	SED
42	0.99	0.20	.052***
39	0.81	0.21	.038***
42 + 39	1.81	0.41	.062***
42/39	1.26	0.96	0.10**

Table 19: Effect of year of growth on sphingomyelin levels (mg/g) and ratios in Section 2 adult NZ red deer velvet antlers. SED is the standard error of the difference; ns, means the any differences are not statistically significant; *, p<0.05; ****, p<0.001.

Band	Y		
	91-92 (12)	92-93 (12)	SED
42	0.99	1.27	.061**
39	0.81	0.73	
42 + 39	1.81	2.00	.051ns
42/39	1.26	1.75	.087* 0.09***

Table 20. Effect of section on sphingomyelin levels (mg/g) and ratios in adult NZ red deer velvet antlers (n=4). SED is the standard error of the difference; ***, p<0.001.

Band			Sec	ction			
	1	2	3	4	7	8	- SED
42	1.57	1.05	0.92	0.87	0.32	0.15	000
39	1.36	0.73	0.78	0.48	0.25		.082***
42 + 39	2.92	1.78	1.70	1.35		0.21	.077***
42/39	1.16	1.46			0.57	0.36	.150***
		1.40	1.19	1.81	1.23	0.80	0.12***

Table 21. Effect of grade and section on sphingomyelin levels (mg/g) and ratios in adult NZ red deer antlers from 1991-92. SED refers to the standard error of the difference among the grades; ns. not significant; *, p<0.05. In all cases the main effect of section is significantly different (p<0.05, see Table 1).

Band	Section -	·	Grade (n)			
		A (2)	B (4)	C (6)	_ SED (grade)	
42	2 .	.99	1.00	.99		
42	8	.24	.25	.14	.109ns	
39	2	.71	.98	.73		
39	8	.22	.26	.18	.083*	
42 + 39	2	1.71	1.98	1.72		
42 + 39	8	.46	.51	.32	.154ns	
42/39	2	1.39	1.04	1.36		
42/39	8	1.09	0.98	0.89	0.18ns	

Table 22. Effect of grade and year on sphingomyelin level (mg/g) and ratio in section 2. The A and B grades for 1991/92 were combined as there were no significant differences between them (Table 21) and because there were no A grade antlers from 1992-93 available for analysis. The SED is the standard error of the difference for main effects of grade and year respectively. "Grade" and "year" refer to the comparisons between grade within year and within grade between year respectively. ns, not significant; *, p<0.05; **, p<0.01; ***, p<0.001.

Band	Year	Grade		SED	SED
	ı caı	AB	С	(grade)	(year)
42	91-92	1.00	.99		
42	92-93	1.36	1.20	.060ns	.060**
39	91-92	.89	.73		
39	92-93	.77	.70	.046**	.046*
42 + 39	91-92	1.89	1.72		
42 + 39	92-93	2.14	2.00	.078**	.078**
42/39	91-92	1.16	1.36		
42/39	92-93	1.78	1.72	0.09ns	0.09**

Table 23. Effect of species and section on sphingomyelin levels (mg/g) and ratios. The data are expressed after it was normalised to a constant dry matter because antler received from processors differed in its water content. The SED refers to comparisons between sections or species. ns, not significant; *, p<0.05; ***, p<0.01; ****, p<0.001.

		Spec	cies	SED	
Band	Section	91-92 NZ red	NZ wapiti	Section (2 with 8)	Species
42	2	.99	1.29		
42	8	.20	.38	.064***	.096*
39	2	.81	.93		
39	8	.21	.31	.045***	.083ns
42 + 39	2	1.81	2.22		
42 + 39	8	.41	.68	.093***	.153ns
42/39	2	1.26	1.39		
42/39	8	.96	1.26	.13**	.22ns

Table 24. Effect of section on FAA levels (nmol/g) of NZ red deer antiers (n=4), *, p<0.05; **, p<0.01; ***, p<0.001. ND, not detected (note: sections 1, 2, 4 and 8 are main beam sections; 3 and 7 are tines).

Amino Acid		··-	S	ection			
	1	2	3	4	7	8	SED
AANB AAD	81	114	56	28	23	17	
ALA/HIS	325	271	294	119	65	27	29 *
ARG	22451	13651	16369	6892	5338		28 ***
ARG ASN	1916	1471	2038	951	1050	2338	785 ***
	2006	1335	1250	411	328	372	124 ***
BAIB	372	543	695	407	247	72	136 ***
BALA	2649	2212	3028	1547	1320	45	55 ***
CAR	145	251	116	188		666	I17 ***
CIT	1015	645	1164	620	30	17	71 *
CYS	322	167	227		339	126	47 ***
GABA	146	85	122	93	62	25	52 ***
GLU	25524	12683		57	106	71	10 ***
GLY	10001	7728	10730	3461	2134	1143	1155 ***
HYL	247	253	8766	5128	4027	1849	572 ***
HYP	976		263	163	162	37	72
ILE	2071	1536	1102	507	389	458	186 ***
LEU	4619	1544	1915	1091	739	262	76 ***
LYS	4215	8123	7953	5988	3977	1061	781 ***
MET		3560	4666	3889	2220	651	197 ***
1MH	1028	1528	1257	1134	760	190	193 ***
3MH	289	225	173	131	33	ND	193
ORN/TRP	1452	921	1113	556	268	209	
PEA	1598	996	1146	1054	316	179	100
	ΝD	31	48	19	ND		124 ***
PHE	1423	2680	2792	1948	1268	7	27
PRO	3564	3270	4385	2594	1897	372	247 ***
PSE	107	661	700	349	226	847	182 ***
SER	4955	2889	3348	1703		91	127 ***
ŢAU	7530	5367	7414	4606	1458	511	211 ***
THR	4373	3017	3208	1758	4377	2067	465 ***
ΓYR	1664	1242	1634	903	1293	553	183 ***
VAL	5646	4633	5187		627	234	67 ***
i Ratio ¹			3107	3457	2264	874	257 ***
	1.8	2.9	3.0	3.9	3.3	2.4	2.00
LEU)/(ILE)	2.2	5.3	4.2	5.5	5.3	2.4	0.32 ***
ASN)/(GLY)	0.20	0.17	0.14	0.08	. 0.08	4.1	0.59 ***
ssential	25292	20556				0.04	0.012 ***
lydrophobic	18351	26556	29017	20215	13571	4334	1684 ***
Hydrophilic		21777	23489	16212	10906	3606	1405 ***
otal FAA	10595	7695	7921	4130	3302	1559	562 ***
	111380	83415	92100	51341	37319	15316	4317 ***

¹ H ratio = ratio of total hydrophobic to hydrophillic AA

Table 25: Effect of grade and section on FAA levels (nmol/g) of NZ red deer antiers. *, p<0.05; **, p<0.01; ***, p<0.001. ND, not detected.

Amino acid		1991-92	Antiers			SED	
	Sect	ion 2	Sect	ion 8	Grade	Section	Grade :
	AB	С	AB	С	_		Section
AANB	99	98	20	15	20	16***	26
AAD	311	260	41	29	30	30***	42
ALA/HIS	17610	14272	3099	2339	877*	859***	1228
ARG	1404	1495	425	366	106	88***	138
ASN	1260	1245	76	72	97	96***	137
BAIB	597	519	50	45	· 27	35***	44
BALA	2653	2280	704	661	138	98***	169
CAR	142	194	6	20	27	25***	37
CIT	279	648	119	132	46**	42**	
CYS	143	157	51	25	12	12***	62*
GABA	192	97	127	73	12***		17
GLU	13282	11834	1407	1150		12**	17
GLY	8958	7757	2375	1826	1048	1016***	1460
HYL	231	271			557	421***	698
HYP	1339		56	37	27	26***	37 -
ILE	1717	1426	341	381	176	94***	199
LEU	8721	1546	369	262	51*	53***	73
LYS		8253	1700	1047	305	242***	390
MET	4371	3600	965	638	198*	90***	217*
1MH	1379	1499	290	184	110	88***	141
	330	185	ND	ND	67	-	-
3MH	1288	1070	298	221	102	94***	139
ORN/TRP	1635	985	244	174	76***	68***	102**
PEA	46	34	ND	5	11	12*	. 16
PHE	2466	2787	458	357	252	253***	357
PRO	3888	3334	1151	834	125**	111***	167
PSE	732	764	144	96	88	79***	118
SER	3126	2761	598	499	173	144***	225
TAU	6215	5648	2470	2157	235	258***	349
THR	3165	2964	585	520	141	131***	193
TYR	1424	1270	307	231 `	52	49***	72
VAL	4905	4674	1208	858	144	141***	202
H Ratio¹	3.0	3.0	3.3	2.6	0.29	0.10	0.31**
(LEU)/(ILE)	5.1	5.3	4.5	4.0	0.24	0.10	0.31**
(ASN)/(GLY)	0.14	0.16	0.03	0.04	0.008	0.010***	0.30
Essential	28128	26818	6000	4232	893	743***	1162
Hydrophobic	23076	22093	5177	3541	768	677*	
Hydrophilic	7861	7421	1602	1437	453		1024
Total FAA	93556	83649	19622	15189	433 3189*	357*** 3306***	577 4593

¹ H ratio = ratio of total hydrophobic to hydrophillic AA.

Table 26. Effect of grade and year on FAA levels (nmol/g) in NZ red deer antlers. *, P<0.05; **, p<0.01; ***, p<0.001. ND, not detected.

				•						
Amino Acid		Sect	ion 2				S	ED		
	199	91-92	199	92-93	Gr	ađe	Y	ear	Gra	ade x
	AB	С	AB	С	_					ear
AANB	99	98	29	41	19		19	**	27	
AAD	311	260	130	85	33		33	***	46	
ALA/HIS	17610	14272	8516	7198	1030	***	1030	***	1457	
ARG	1404	1495	1671	1403	110		110		155	
ASN	1260	1245	1395	865	158		158		223	
BAIB	597	519	690	505	.43	**	43		61	
BALA	2653	2280	1826	1871	140		140	** *	197	
CAR	142	194	114	109	35		35		49	
CIT	279	648	740	727	124		124		175	
CYS	143	157	207	180	15		15	*	22	
GABA	192	97	25	28	10	***	10	***		***
GLU	13282	11834	4920	4222	1134		1134	***	14	•••
GLY	8958	7757	3287	2851	524		524	***	1603	
HYL	231	271	302	334	43		43		741	
HYP	1339	1426	653	958	268		268	*	60	
ILE	1717	1546	1012	914	59	*	59	***	379	
LEU	8721	8253	13188	9980	434	***	434	***	83	
LYS	4371	3600	4195	3035	206	***	206		613	**
MET	1379	1499	3047	1884	295		295	**	291	
1MH	330	185	140	198	47		47		418	
3МН	1288	1070	798	636	127		127	**	66	
ORN/TRP	1635	985	1021	468	216	*	216	*	179	
PEA	46	34	ND	ND	11		410	•	306	
PHE	2466	2787	3044	2904	365		365		-	
PRO	3888	3334	1522	1336	136	*	136	***	516	
PSE	732	764	359	289	89		130	***	193	
SER	3126	2761	1377	1221	164		164	***	126	
TAU	6215	5648	4587	4390	317			***	231	
THR	3165	2964	2358	2051	156		317 156	***	448	
TYR	1424	1270	1061	959	71		71	***	221	
VAL	4905	4674	5547	4344	205	**	205	***	100	_
H Ratio¹	3.0	3.0	5.9							*
(LEU)/(ILE)	5.1	5.0 5.3	3.9 13.1	5.0	0.31		0.31	***	0.44	
(ASN)/(GLY)	0.14			11.2	0.60		0.60	***	0.85	
		0.16	0.43	0.36	0.018		0.018	***	0.025	*
Essential	28128	26818	34063	26829	1225	**	1225	*	1732	* .
Hydrophobic	23076	22093	27360	21676	1020	**	1020		1443	
Hydrophilic	7861	7421	4691	4565	479		479	***	677	
Total FAA	93556	83649	67678	56130	3741	*	3741	***	5290	

¹ H ratio = ratio of total hydrophobic to hydrophillie AA.

Table 27. Effect of species on FAA levels (nmol/g). *, p<0.05; **, p<0.01; ***, p<0.001. ND, not detected

Amino Acid	Secti	ion 2	Sect	ion 8	SED
	Wapiti	Red	Wapiti	Red	
AANB	154	91	15	17	
AAD	223	265	31	34	47 65
ALA/HIS	14210	14768	2050	2580	55 3142
ARG	1689	1343	380	375	497
ASN	1073	1160	104	70	
BAIB	486	517	58	45	151 102
BALA	1924	2285	774.	647	245
CAR	163	155	26	12	
CIT	553	487	91	119	126
CYS	185	139	28	39	84
GABA	137	134	73		57
GLU	8519	11634	754	95	51
GLY	7543	7742	1524	1213	1990
HYL	367	232	53	1993	2940
LE	1449	1511	26 -	43	83
_EU	8765	7862	1554	299	329
LYS	4284	3692		1303	1207
MET	1987	1333	707	760	613
MH	333	229	273	225	179 ***
MH	1106	1093	ND	ND	96
ORN/TRP	1239	1093	317	246	195
EA	. 39	37	159	198	514
HE	3726	2433	ND	2	29
RO	4035		494	387	417
SE	5 60	3345	732	942	1488
ER	2301	693	36	114	94
AU .	5062	2727	304	520	629
HR	3522	5495	2051	2195	490
YR	1489	2839	5 31	524	493
/AL		1248	273	255	236
	5221	4437	875	980	698
I Ratio¹	3.03	3.02	2.40	2.00	
LEU)/(ILE)	7.14	5.24	6.25	2.90	0.416 *
ASN)/(GLY)	0.16	0.15	0.23	4.27 0.04	0.712 *** 0.023
ssential	30643	25451	5078		
ydrophobic	25183	20922	4192	4854	3802 *
ydrophilic	8234	7079		4136	3615
otal FAA	84001	82081	1766 15321	1435 16514	1483 13809

¹ H ratio = ratio of total hydrophobic to hydrophillic AA.

Table 28: The effect of the section of NZ red deer velvet antlers on yields of extractables after aqueous or organic extraction (± standard deviation).

	į			Average	; yield (%)	Average yield (%) from dried NZ red deer velvet antlers Section	J NZ retion	l deer velv	ct antlers			
Extract		-		2		3		4		7		~
Aqueous Organic	28.4	(5.86)	27.5 5.04	(4.96)	22.0	(3.06)	28.8	(3.74)	13.9	(2.81)	12.57 1.78	(1.61)

Table 29: The effect of grade of NZ red deer velvet antler on the yield (%) of extracts with aqueous or organic extraction (\pm standard deviation).

			Sect	ion	
Extract	Grade		2	;	3
Aqueous	A	39.2	(2.26)	14.4	(1.54)
	В	39.6	(1.77)	13.8	(2.55)
		39.4	(2.01)	14.2	(1.87)
Mean			•		(1.0.)
		2.99	(0.23)	1.08	(0.16)
Organic	Α	3.05	(0.17)	0.93	(0.19)
	В	3.01	(0.20)	1.02	(0.18)
Mean					

Table 30: The effect of grade and process technique on the yield (%) of extract from dried NZ red deer antlers.

	_	Secti	on 2		Secti	ion 8	
		Gra	ıde		Gr	ade	=
Extract	Drying method	В	С	Mean	В	С	Mean
Aqueous	Freeze dried Commercial 1 Commercial 2	35.5 25.5	31.5 26.5	33.2 26.0	11.8 9.3	7.7 6.3	9.6 7.8
Organic	Freeze dried	22.6 2.70	22.9 2.59	22.8	7.6 1.07	6.6 0.94	6.9 0.99
	Commercial 1 Commercial 2	2.33 1.93	2.20 1.90	2.26 1.91	0.94 1.02	0.72 0.88	0.83 0.92

Table 31: The effect of antler section (of C grade NZ red velvet antler) on the activity (IC $_{50}$) of organic antler extracts in the P388 assay (SED = standard error of the difference between means).

_			Section		
	2	4	7	8	SED
IC ₅₀ dose (μg/ml)	35	32	378	409	77

Table 32: The effect of velvet ander grade (NZ red) on organic extract activity (IC_{so})in the P383 assay.

		Gra	ade	
	A	В	С	SED
IC _{so} dose (µg/ml)	750	155	35	154

Table 33: The effect of drying method and antler section on the activity of organic extract from NZ red velvet antler. Results expressed as IC_{50} ($\mu g/mI$) in the P388 assay. Results are not corrected for differences in dry matter content or extract yield.

	Freeze dried	Commercial 1	Commercial 2	SED
Section 2	860	197	195	148
Section 8	720	318	469	696

Figure 1: Antler sections used for compositional analysis.

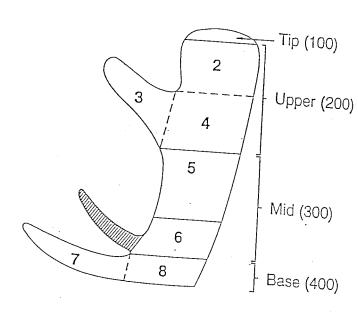


Figure 2: Relative contents of ander constituents with days of growth.

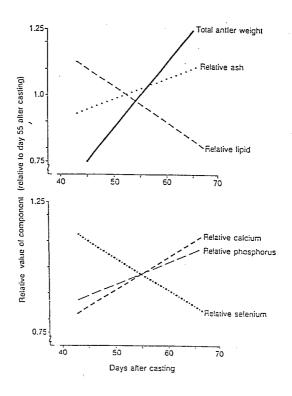
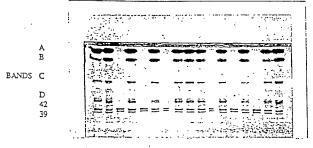


Figure 3: A TLC plate of a sphingomyelin assay of red deer standards, samples and purified standards.

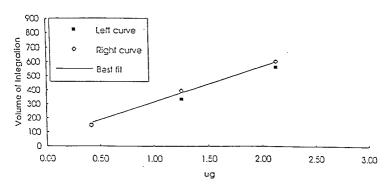
Lane	Extract/Sample
1	Quality control red deer Section 2. This sample was run in all plates. Data from it were used to calculate coefficient of assay variability
2	Section 2 NZ red deer 1992-93
3	5 µg standard (mixed 39 and 42 material)
4	Quality control red deer - same as lane 1
5	3 µg standard
6	Section 2 NZ red deer 1992-93
7	1 μg standard.
8	Section 2 NZ red deer 1992-93
9	Quality control red deer - same as lane 1.
10	Section 2 NZ red deer sample
11	l μg standard
12	Section 2 NZ red deer sample
13	3 µg standard
14	Section 2 NZ red deer sample
15	5 μg standard
16	Section 2 NZ red deer sample
17	Quality control red deer - same as lane I
	•



1 2 3 4 5 6 7 8 9 40 11 12 13 14 15 16 17 LANE

Figure 4: A standard curve of sphingomyelin values using the densitometry output





Sphingomyelin (42 band)

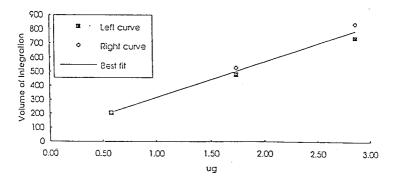
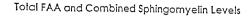


Figure 5: Relationship between total free amino acids (FAA) and sphingomyelin levels. The data are from sections 1, 2, 3, 4, 7, 8 from NZ red deer antlers.



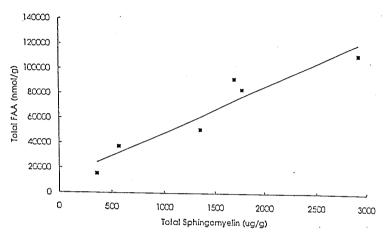


Figure 6: Mitogenicity of antier extracts: the relationship between cell growth response of antier X and Y cell lines (uptake of tritiated thymidine) and the concentration of aqueous velvet antier extracts in the medium (extracts from NZ red deer velvet antier).

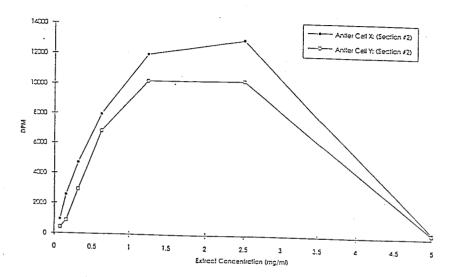


Figure 7: The relationship between cell growth response of antler X cell lines (uptake of tritiated thymidine) and the concentration of aqueous velvet antler extract in the medium (from NZ red deer velvet antlers).

