# THE CORRECTION FACTOR OF HAIR DENSITY IN THE SKIN OF BUFFALO

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#### ABSTRACT

The hair density of the back region in water buffalo was calculated by counting the number of hair follicles in the horizontal sections prepared by conventional paraffin embedding method and stained with hematoxylin and eosin . The calculated hair density was 3.53 hair follicle/mm<sup>2</sup> of the buffalo skin. All the hair follicles were of primary type and distributed randomly.

The skin samples showed different variable shrinkage percentage during routine histological technique. The shrinkage percentage of surface area of the stained sections was found to be 21 % of the original value. Thus the correction factor of the hair density was calculated to be 0.79, this leads to conclusion that the actual hair density of the back region of the skin of living buffalo is 2.78 hair follicle/mm<sup>2</sup> only .

## العامل المصحح للكثافة الشعرية لجلد الجاموس

مؤيد حسن عبد الرحيم و عبد السلام متي الياس و نزيهة سلطان احمد فرع التشريح والانسجة والاجنة، كلية الطب البيطري، جامعة الموصل، الموصل، العراق

#### الخلاصة

حسبت الكثافة الشعرية لمنطقة الظهر في جلد الجاموس النهري بتعداد جريبات الشعر من المقاطع الافقية المحضرة بطريقة الاسجاء بالبارافين والمصبوغة بصبغة الهيماتوكسلين والايوسين حيث بلغ ٣,٥٣جريبة شعر/ملم من جلد الجاموس وكانت جريبات الشعر من النوع الابتدائي وعشوائية التوزيع. أظهرت عينات الجلد درجات متفاوتة من الانكماش بعد كل خطوة من التمريرات

اظهرت عينات الجلد درجات متفاوتة من الانكماش بعد كل خطوة من التمريرات النسيجية الاعتيادية. وبلغت نسبة الانكماش النهائي في المقاطع النسيجية المصبوغة ٢١% واستناداً الى ذلك فقد بلغ العامل المصحح للكثافة الشعرية لمنطقة الظهر في الجاموس ,٧٩ ويستنتج من ذلك أن الكثافة الشعرية الحقيقية لجلد الظهر في الجاموس الحي هي ٢,٧٨ جريبة شعر / ملم فقط.

#### **INTRODUCTION**

The hair density is usually calculated by examining the number of hair follicles in a definit surface area of the stained horizontal sections of the paraffin embedded skin samples (1, 2, and 3).

Iraqi Journal of Veterinary Sciences, Vol. 20, No.2, 2006 (101-106)

The different chemical substances used in routine histological processing i.e.: fixation, dehydration, clearing and wax embedding affect the surface area of the stained samples , which is decreased if compared to fresh samples (4, 5) this decrease in the surface area reflects a higher hair density in comparison to the fresh skin. Therefore, the false reading of the hair density of the stained skin samples should be corrected to these normally existing in the original fresh skin by applying a correction factor of the hair density.

The correction factor of the hair density in the skin of goats, sheep, cattle and camel has been measured by several authors (6, 7, 8, 9, 10), but the correction factor of the hair density in the skin of local water buffaloes received no attention. The goal of this work is to find out the correction factor of the hair density in this animal.

#### MATERIALS AND METHODS

Samples of skin from the back region of 8 male buffaloes of 1-2 years old were collected by a punching tool of 10 mm diameter. The samples were fixed in a neutral buffered formalin for 24 hours and then immersed in 4% phenol for 24 hours to soften the keratinized material of the skin (10, 11). The samples were dehydrated by ascending grades of ethyl alcohol, cleared with xylene and infiltrated with paraffin wax of 56 °C melting point. Horizontal sections of 5-6  $\mu m$  thick were cut at sebaceous glands level and stained with routine hematoxylin & eosin and trichrome stains (5).

Hair density were calculated by counting the number of hair follicles in a known surface area of the microscopic field, which is confirmed by the counting device of the visopan microscope (Fig.1).



Figure 1: Explaining the calculation of surface area of the section by using a visopan device.

### Iraqi Journal of Veterinary Sciences, Vol. 20, No.2, 2006 (101-106)

The correction factor is obtained by following the formula  $a_1/a_2$  applied by Carter and Dolling 1954, where  $a_1$ : represents the surface area of the stained section, while  $a_2$ : represents the surface area of the fresh sample of skin (7).

## RESULTS

All hair follicles in the dermis of back skin in native water buffaloes were found to be of primary type and distributed evenly (Figure 2).



Figure 2: Horizontal section of buffalo skin at sebaceous gland level. Note the even distribution of primary hair follicles . Trichrome stain X 190 .

The alterations in the surface area of the skin samples following fixation, phenol treatment, dehydration, clearing, paraffin embedding & sectioning and staining were recorded in Table 1.

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Table 1 : Differences in surface area and shrinkage percentage during routine histological processings of buffalo skin .

Samples	Fresh skin	After fixation	After	After	After clearing	After paraffin	After staining
processings	samples	with neutral	phenol	dehydration	with xylene	embedding &	with
		buffered	treatment	with alcohol		sectioning	hematoxylin &
		formalin					eosin stain
Average of	78.5	73.2 <u>+</u> 2.15	69.4 <u>+</u> 2.51	65.3 <u>+</u> 3.51	61.6 <u>+</u> 3.12	53.0 <u>+</u> 3.25	62.0 <u>+</u> 3.1
surface area							
$(mm^2)$							
Shrinkage %		7	12	17	22	32	21

Hair density of stained sections = 3.53 hair follicle / mm<sup>2</sup>. According to  $a_1 / a_2$  formula the correction factor = 0.79. The actual hair density = 2.78 hair follicle / mm<sup>2</sup>. In the present work, the original surface area of the fresh skin samples was 78.5 mm<sup>2</sup> and it reduced to become 61.2 mm<sup>2</sup> in the mounted stained sections of the same samples. By application of  $a_1/a_2$  formula, the correction factor of the back skin will be 0.79.

The hair density of the stained sections of the back skin in native water buffaloes was 3.53 hair follicle/mm<sup>2</sup>. This density should be corrected. The calculated correction factor was 0.79, so that, the actual hair density of the back skin in water buffalo will be 2.78 hair follicle/mm<sup>2</sup>.

#### DISCUSSION

All hair follicles encountered in the skin of water buffalo are of primary type . No hair grouping was noticed. These findings agree with the results of Nair and Benjamin in their study on the skin of Indian water buffaloes (12).

Different degrees of shrinkage were noticed on the skin samples after fixation, phenol treatment, dehydration, clearing, wax embedding and sectioning. Our suggestion that the reason of this shrinkage is the extraction of the fluid from the skin samples as a result of using hypertonic chemical solutions in the histological processing. The higher shrinkage ratio occurred after paraffin infiltration and sectioning may be due to the wax infiltration as well as the affect of oven heat. These results coincide with the results of other works carried out on the skin of sheep, goats and cattle (4, 6, 7, and 8). The actual hair density in the skin of native water buffalo was 2.78 hair follicle  $/ \text{ mm}^2$  which is confirmed by Jenkinson and Nay whom they found that the hair density was 2.37 hair follicle./mm<sup>2</sup> in water buffaloes and 1.45 hair follicle /mm<sup>2</sup> in the skin of the African buffaloes (9). The hair density in the skin of native water buffalo is very low if compared to other large animals; cattle and camel (7, 9). The low hair density in buffalo skin may be contributed to absence of the secondary hair follicles which considered being the main factor in increasing hair density. This suggestion agrees with the results of other workers who study the hair density of sheep and goats, where they proof that the high hair density in these animals is greatly dependent on the number of secondary follicles (6, 10).

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