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Lung elastic fibres from embryonic to adult birds

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In mammals it is well known the role of the elastic fibres in the lung development and its fundamental importance in the process of distending and recoil of the organ as a whole [1]. The bird' lungs are compact and virtually non-expansible [2].
After morphological studies on elastic fibres distribution and quantification by image analysis [3] we carried out a study on biochemical methods to quantify elastin in the avian lung, using domestic fowl (*Gallus gallus*) as an experimental model. We made use of two analytical methods for the elastin determination in these animal lungs from the 14th day of hatching until the 42nd day after hatchling.
As analytical methodologies we used high-performance liquid chromatography (HPLC) and a colorimetric method based on the use of a stain that has been only applied in histological techniques.
The HPLC analysis was carried out in a reverse phase system with a binary gradient, detection by UV wavelength set at 254 nm [4]. By this way we were able to quantify desmosine and isodesmosine in the studied samples.
The colorimetric method was used by applying a commercial *kit* ("Fastin™ Elastin Assay") which quantifies soluble tropoelastin and insoluble elastin made soluble (α-elastin) after treatment with hot oxalic acid.
The results obtained by HPLC were statistically different if comparing the incubation period with the after-hatchling days ($p<0,001$). This fact is in agreement with the immaturity of elastin in the period of incubation, without the presence of cross-linkings.
The determinations by the colorimetric method provided results without statistical difference between both analyzed periods ($p=0,581$). This confirms the presence of elastin in the incubation period since this method determines the soluble elastin (α-elastin) as well as the insoluble elastin made soluble after treatment with hot oxalic acid, in the after-hatchling period.
Comparing the results obtained by both analytical methods, it is remarkable that only in the after-hatchling period there is a positive and significative relation between both methods.

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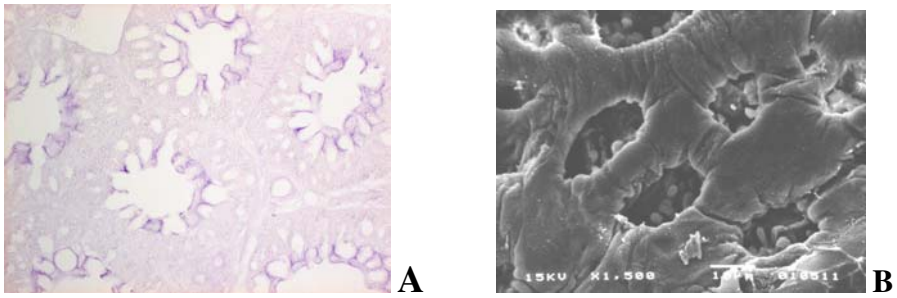

Fig. 1. **A**-Sections of parabronchial units stained by a modification of the Gomori method; elastic fibres can be observed along the axis of interatrial septa. **B**- Parabronchial lumen observed by Scanning Electron Microscopy; interatrial septa can be observed between the atria.

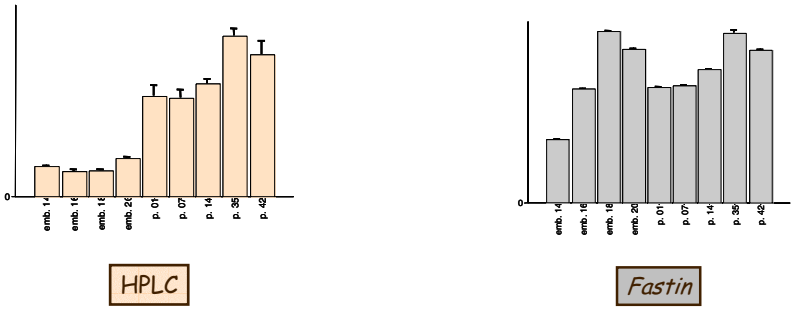

Fig. 2. Quantification of desmosine e isodesmosine by HPLC and quantification of elastin by the colorimetric method "*Fastin*™*, Elastin Assay"*.
Values are expressed as µg of elastin/mg of total protein.

TABLE 1. Amino Acid composition of Soluble and Mature Elastin

		Residues per 850 residues *	
		Soluble Elastin	**Mature Elastin**
Gly	G	275	277
Ala	A	196	197
Pro	P	95	80
HyPro	P	7,5	12
Val	V	110	105
Ile	I	15	16
Leu	L	38	44
Tyr	Y	12	14
Phe	F	24	27
Lys	K	38	6
Cross links	§	Very low	25-30
Arg	R	4,4	5,5
Asp+Asn	D+N	2,7	7,5
Thr	T	12	14
Ser	S	8	11
Glu+Gln	E+Q	16	18
Met+Cys+Trp+His		0	2,6

* Based on an assumed chain length of 850 residues
§ Expressed as Lys equivalents
(Modified from Gray, Sandberg and Foster, 1973)

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