Dental microwear in dinosaurs: a comparative analysis of polysiloxane replication

Vince Williams, Mark Purnell and **Sarah Gabbott**
Department of Geology, University of Leicester

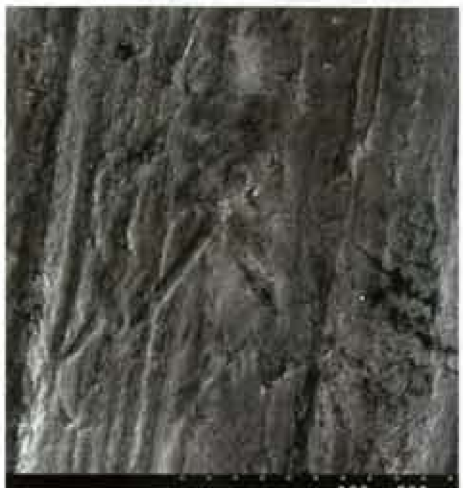
EVERY time a tooth comes into contact with food or with another tooth, it is damaged. Abrasive particles cause microscopic scratching and pitting, and while this may seem undesirable from the point of view of the tooth's owner, it is good news for palaeontologists because the "microwear" that accumulates on the tooth surface provides a record of what has been eaten and the range of tooth movements involved.

This is true of humans and any other animals with teeth, both extant and fossil, and studies of living mammals with controlled or known diets have established the relationship between feeding and microwear sufficiently well for the microwear footprint on fossil teeth to be used to interpret diet.

This is an extremely powerful tool with which to analyse feeding in extinct animals and has been applied extensively to fossil hominoids in order to evaluate the role of dietary changes in human evolution (Teaford and Ungar, 2000).

It has also been applied to extinct mammals and has revealed in surprising detail how feeding in mammals has tracked past environmental change (Semprebon *et al* 2004; Solounias and Hayek, 1993). But can the same technique be applied to dinosaurs?

We are attempting to find out by conducting the first systematic study of dinosaur microwear. Our main aims are to understand how diet and feeding changed during the evolution of a major group of herbivorous dinosaurs, and assess whether dietary changes were related to known changes in the environment and vegetation.



Above: FIG 2a – SEM image of microwear on a fossil hyena canine tooth
Below: FIG 2b – SEM image of microwear on a cast made from a GC Exafast mould

For example, we know from the fossil record that between 150 and 60 million years ago, the gymnosperm conifer forests – which had dominated the global flora for more than 100 million years – were replaced by angiosperm flowering plants. It seems quite likely that such a major change in food supply would have had a significant effect on herbivorous dinosaurs and their evolution.

More surprisingly, it has also been suggested that the grazing activities of dinosaurs may have been an important factor in causing this change in vegetation.

Before we can evaluate what the microwear footprint of dinosaurs can tell us, however, we need to understand two things. Firstly, we need to see what microwear in dinosaur teeth looks like, because dinosaur teeth are very different to those of mammals.

Mammals replace their teeth just once, so adult teeth are retained in the mouth for a long time over which microwear can accumulate. Dinosaurs, on the other hand, shed worn teeth and continued to grow replacements throughout their life.

To further complicate matters, dinosaur jaw articulation is very different to that of mammals and they did not bite or chew their food in the same way. In fact the jaws of most dinosaurs, including all of the earliest forms, could only move up and down, meaning that their tooth surfaces could not slide across each other like those of mammals do, making chewing impossible.

Secondly, we cannot conduct the experiments required to establish the relationship between food type and microwear in dinosaurs because most of them are extinct and those that survive (birds) have no teeth. The nearest extant relatives with teeth are reptiles, so we need to investigate microwear and feeding in living reptiles.

Our dinosaur study will focus on ornithopods, a large group of herbivorous dinosaurs including well-known characters like *Iguanodon*. They make an ideal group to investigate because they are known from strata of earliest Jurassic through to latest Cretaceous age, an interval of 140 million years, with their continuous evolutionary history spanning most of the time that dinosaurs existed.

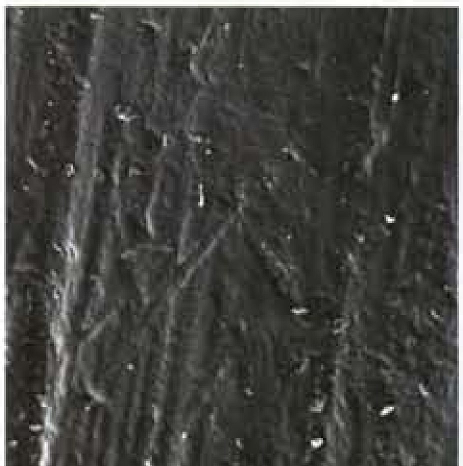


FIG 2c – SEM image of microwear on a cast made from a Speedex Light mould

FIG 1a – Lambeosaurus lambei *skull, from the Upper Cretaceous Oldman Formation of Alberta, Canada. YPM 3222*

The earliest ornithopods had no ability to masticate food as their jaws occluded in the vertical plane only, slicing and shredding vegetation in a manner similar to that of living iguanas. Later ornithopods, however, evolved two different but highly successful mechanisms for masticating food. The most advanced forms evolved a hinge in the top of their skulls that allowed the upper jaws to slide laterally over the lower jaws and grind plant material between them, in essence allowing them to chew.

FIG 1b – Speedex Light moulds of Hypsilophodon foxi *isolated teeth, from the Wessex Formation, Isle of Wight. MIWG.6362*

To carry out our research we need to examine teeth under high magnifications in a scanning electron microscope (SEM), but ornithopod specimens are distributed in various museums and private collections around the world, and most are either too large to fit in a SEM or unavailable for loan.

Consequently we need to obtain casts of teeth, and this requires a moulding material that will replicate tooth microwear with high fidelity and, crucially, leave fragile dinosaur material undamaged.

FIG 1c – Araldite 20/20 cast of Hypsilophodon foxii *tooth, made from Speedex Light mould, mounted on SEM stub and splutter-coated with gold*

This is a tough challenge because after millions of years dinosaur teeth tend to be rather brittle and cracked, and are usually held together with a cross between glue and varnish known as a consolidant. The varnish-like sheen can be seen in **Fig 1a**, a *Lambeosaurus lambei* skull that is coated in consolidant.

Obviously this has to be removed from the tooth surface to get at the microwear, so what starts out as an already fragile fossil suddenly becomes very fragile indeed. Similar studies on fossil mammal teeth have used room temperature vulcanising (RTV) silicone rubbers, but these are unsuitable for a number of reasons.

The silicon fluid they contain tends to penetrate and stain the surface of fossils, and many have a high tear strength, meaning that unless a release agent is used, parts of the fossil can be pulled away as the mould is removed – and this is not good. In addition most RTV silicone rubbers have long cure times, and while this may be acceptable for use on fossils, it is unworkable where live reptiles, an important part of our study, are concerned.

Polysiloxane impression mediums have none of these drawbacks and look to be ideal for our purposes. Their low tear strength in particular means that they are likely to give way before a fossil tooth gets pulled apart.

The only aspect of using polysiloxanes about which we were uncertain was the fidelity with which they can replicate microwear.

The scratches and pits we are interested in are measured in microns (thousandths of a millimetre), and the replicated surface must pick up every feature or the statistical analysis of the microwear patterns will be fatally flawed.

In order to evaluate whether polysiloxanes were up to the job, we performed a comparative test of a number of polysiloxane dental impression mediums, including Aquasil, Reprosil, GC Examix, GC Exafast, Coltène President Jet and Coltène Speedex.

After removal of any consolidant, tooth

surfaces were cleaned with a fine, soft brush. Impression medium was mixed according to the manufacturer's instructions; small quantities (less than 30ml) were generally used, but this varied according to the size of the specimen being moulded.

After surface moulding (**Fig 1b**), casts were prepared using low viscosity Araldite 20/20 epoxy resin. These were then mounted for SEM and splutter-coated with gold (**Fig 1c**).

Due to the fragile nature of dinosaur teeth, the initial testing was performed on hyena teeth, progressing to dinosaur teeth only when the moulding process was proven safe.

Results

The results of comparative testing showed that the condensation polymerising polysiloxanes gave the best performance, and out of these the best was Coltène Whaledent's Speedex Light, which produced near perfect replication of the dental microwear once the casts were splutter-coated in gold.

Fig 2 compares microwear on a fossil hyena tooth (**Fig 2a**) to that replicated on casts made from a GC Exafast mould (**Fig 2b**) and a Speedex Light mould (**Fig 2c**). (GC Exafast was the next best performing medium after Speedex Light).

Fig 3 compares microware, at a much higher magnification, on a dinosaur (*Hypsilophodont foxii*) tooth (**Fig 3a**) to that replicated on a cast made from Speedex Light (**Fig 3b**); the microwear in which we are interested is highlighted in **Fig 3c**.

Here the cast gives better definition than the original tooth; this is typical and relates to the coating in gold which cannot be applied to the original tooth.

Multiple casts were taken from the same Speedex Light mould with no appreciable difference in the quality of the casts and there was no significant shrinkage of the moulds over three months.

Summary

Care needs to be taken when de-moulding as the polysiloxanes, while reasonably elastic, will tear if more than gentle "teasing" is attempted. Immediately after de-moulding, the polysiloxanes are susceptible to condensation and the moulds must be dried thoroughly before casting. Many of the polysiloxanes require mixing guns and while these were very easy to use, the ability to be hand-mixed proved to be a great advantage as it allowed the working time to be varied.

Due to the low viscosity of the polysiloxanes, it was necessary to create a "dam" around some specimens to prevent overflow from the area to be moulded. No damage was caused to any of our specimens by Speedex Light or GC Exafast.

Our results demonstrate that Speedex Light can replicate tooth microware at magnifications in excess of 300 times and this is more than sufficient for our research needs. Using this medium, our research will progress to form a systematic investigation of tooth microwear in ornithopod dinosaurs, building on previous microwear work. Our studies will have clear implications for understanding the diet and the evolution of feeding mechanisms in dinosaurs and present day reptiles.

Acknowledgements

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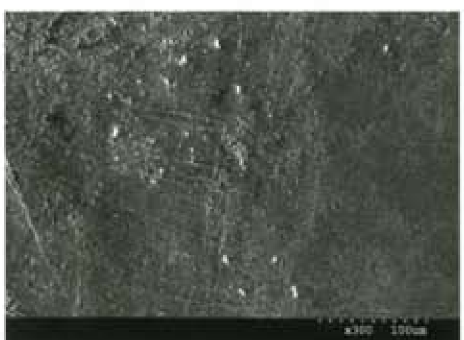

FIG 3a – SEM image of microwear on a Hypsilophodon foxii *tooth. MIWG.6362*

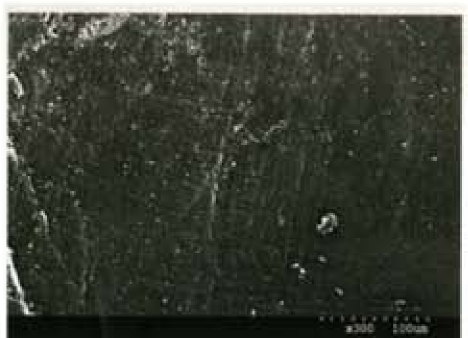

FIG 3b – SEM image of microwear on a cast made from a Speedex Light mould

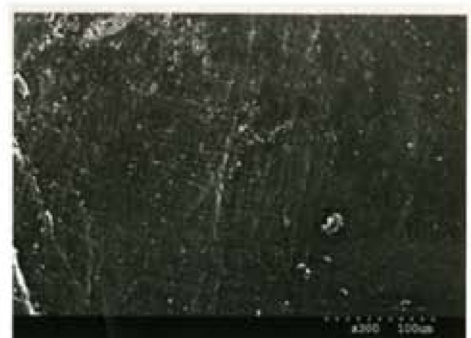

FIG 3b – SEM image of microwear on a cast made from a Speedex Light mould

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