

Effect of different salt concentrations on water binding capacity of pre-rigor beef.

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INTRODUCTION

Muscle removed 'hot' from the carcass has greater water binding capacity (WBC) than that removed post-rigor and is excellent raw material for sausage manufacture (Hamm, 1972). Recent studies (Jolley, Honikel & Hamm, 1980-81; Honikel *et al.*, 1981a & b) have shown that improvement in WBC by chopping with salt is most effective if done as soon as possible post slaughter; nevertheless, a marked benefit from salt is still achieved right up to onset of rigor.

These earlier studies standardised on a 2% salt concentration. Meat products have a range of salt concentrations associated traditionally with their manufacture, often below 2%. Before the use of pre rigor beef in such products can be recommended, it is necessary to know the effect of lower levels of salt on WBC, and whether this effect changes with increasing delay between slaughter and manufacture. The present work describes an investigation of these factors.

MATERIALS AND METHODS

Beef neck muscles (mainly *M. sternomandibularis* and *M. sternomastoideus*) were obtained within 40 min of slaughter from either a local abattoir or the abattoir of the Meat Research Institute, U.K. The muscles were trimmed of fat and connective tissue and then divided longitudinally into slices 1-2 cm thick and in the weight range 90-200g. Sufficient material was taken for immediate sampling (time 0) and the remaining slices vacuum-packed in individual polypropylene pouches and placed in a cryostat bath at +0.5°C, 7°, or 14°C, to produce marked, slight, and no cold shortening respectively (Bendall, 1973), so that interactive effects of sarcomere length and salt concentration could be checked.

Randomly selected packs were removed at two further sampling times chosen to correspond approximately to two important events in the pattern of ATP hydrolysis post rigor as described by Bendall, 1973. Time 1 approximates to the end of the ATP delay phase when resynthesis of ATP is no longer sufficient to equal its hydrolysis, and consequently the concentration begins to fall from its previously constant value. Similarly, time 2 corresponds to an ATP concentration of 50% of the delay phase, shortly before the onset of rigor (Bendall, 1973). These times were 8 and 12 hours respectively at both 7° and 14°C, and 4.5 and 7 hours respectively at 0.5°C (Jolley *et al.*, 1980-81).

Preparation of salted muscle homogenates

Salted muscle "homogenates" were produced by the method of Jolley *et al.* (1980-81), with minor modification. Muscle strips were minced once through a plate with 4.5 mm holes and weighed. This was repeated with successive packs from the cryostat bath until there was sufficient bulk mince to produce the appropriate number of homogenates. The bulk mince was mixed by hand and 66g samples mixed with appropriate amounts of salt. Finally, 33g of an ice/water mixture was added to each salted mince, mixed, and chopped four times for three seconds in a "Moulinette" (Moulinex, France) chopper. The term "homogenate" is used for convenience, and is not meant to imply a perfectly homogeneous entity either chemically or histologically.

Seven levels of added salt were used: 0, 0.2, 0.5, 1, 1.3, 1.7 and 2% of the final weight of the homogenate. In only one experiment were all seven levels examined, and then only at time 0. Five levels of addition were assessed at each sampling time in all other experiments, either 0, 1, 1.3, 1.7 and 2% (1 experiment at each of the three temperatures) or 0, 0.2, 0.5, 1, and 2% (1 experiment at each of the three temperatures; two further experiments at 7°C, omitting time 2).

Determination of pH

In the early experiments of this study, pH was determined immediately after homogenising 3-5 days of muscle with an approximately equal amount of double distilled water. This was later replaced by homogenising lg samples of mince in 10 ml 150mM KCl/5mM iodoacetic acid (pH 7.0) using a laboratory mixer emulsifier (Silverson Machines Ltd) and determining the pH with a pH M63 Digital pH meter (Radiometer) fitted with a combined glass electrode (Russell pH Ltd). These methods of determination produce similar results, but the latter procedure produces a sample of stable pH.

Water binding capacity

The WBC was determined on 0.3g of each homogenate by the filter paper press method of Grau and Hamm (1952, 1957) and is reported as area of expressed fluid.

WBC was also assessed on all samples by the ability of homogenates to hold water during heating and subsequent centrifugation. Between 2-5g homogenate was accurately weighed into 15 ml glass tubes which were stoppered with glass marbles and placed in a boiling water bath for 15 minutes. Fluid released on heating was discarded and the sample lightly dried on tissue paper. The heated samples were then centrifuged at 15000 rpm for 15 minutes at 5°C, the released fluid discarded and the sample reweighed after lightly drying as before. Total weight loss is expressed as a percentage of the initial sample weight.

RESULTS

There was no marked effect of holding temperature on WBC, in agreement with our earlier results (Jolley et al, 1980-81; Honikel et al, 1981b). In the absence of such effect, results at any one salt concentration have been combined at each of the sampling times.

Figure 1 shows the relationship between area of expressed fluid and salt concentration in all homogenates prepared at time 0 and time 2. Little or no fluid was expressed from homogenates made with 1.3% salt and above, which therefore had maximum WBC by this method. As the amount of salt added was lowered from 1.3%, the area of expressed fluid increased. Within any experiment and at all sample times, this relationship frequently appeared linear but there was considerable variation in the pattern. This is reflected by the size of the error bars in Figure 1, most notably with 0.5 and 1% at time 2.

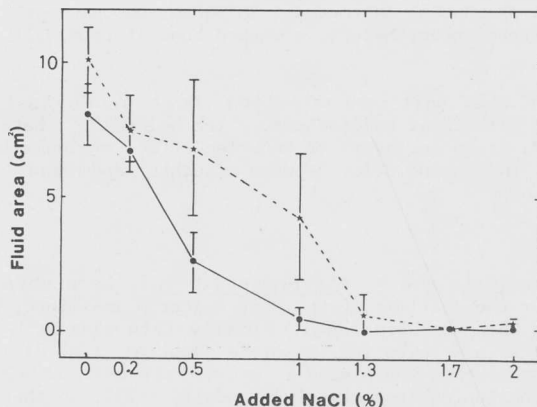


Figure 1. Effect of differing salt concentrations on WBC assessed by the filter paper press method —●— time 0: —★— time 2. Error bars indicate standard error of the mean at the point.

The relationship between total fluid losses on heating and subsequent centrifugation and salt concentration at time 0 and time 2 is illustrated in Figure 2. There was a highly significant ($P < 0.001$) linear correlation between the two factors at all sampling times.

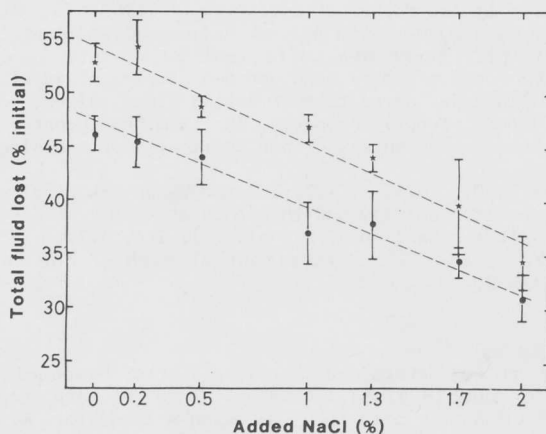


Figure 2. Effect of differing salt concentrations on total fluid loss on heating and subsequent centrifugation, expressed as percent initial weight of sample. Broken lines are those given by linear regression analysis ($P < 0.001$). Other details as Figure 1.

Both Figures 1 & 2 show that the WBC of homogenates produced at time 2 was not as good as those produced at time 0, excluding the previously mentioned results with 1.3% salt and above. Results for time 1 were generally intermediate between those shown for the other sampling times.

DISCUSSION

Three factors are known to be important in explaining the superior WBC of pre-rigor meat: ATP, pH, and ionic strength (Hamm, 1972). The major role of ATP in this context is probably to permit the spatial separation of actin and myosin without the hindrances imposed by cross links (Jolley et al, 1980-81; Honikel et al, 1981a). This separation can presumably occur regardless of sarcomere length in a way analogous to the relaxing of cold shortened muscle when transferred to 20°C before rigor onset (Bendall, 1973).

Table 1 pH at each sampling time.

Sample time	Number of experiments	Mean pH	Standard deviation	Range
Time 0				
Time 1	9	6.86	0.12	6.75 - 7.10
Time 2	8	6.40	0.17	6.18 - 6.67
	6	6.05	0.07	5.95 - 6.13

Mean pH values and observed range at each sampling time are shown in Table 1. The muscle used at all the sample times in this paper had pH >5.9 and was therefore almost certainly pre-rigor (Honikel *et al.*, 1981a). This means that the known variables in WBC of pre rigor meat are reduced to pH and ionic strength. It follows from this that the decrease in WBC observed with post mortem holding of the muscle is due to the fall in pH. Honikel *et al.* (1981, a & b) found that WBC of homogenates made pre rigor with 0 or 2% added salt fell fairly linearly with the pH of the muscle from which they were prepared. A similar trend is seen when the current results for WBC at any one salt concentration are plotted against meat pH. The good WBC of high pH meat, which is well known, probably arises from greater interfilament repulsion due to the higher net negative charge on the contractile proteins the further they are from their iso-electric points (Hamm, 1960, 1975).

Improvement in WBC with increasing salt concentrations encompassing those used here has been frequently reported in the literature with post rigor material (e.g. Grabowska & Hamm, 1979; Moore *et al.*, 1976; Hermansson & Akesson, 1975; Sherman, 1961, 1962; Mahon, 1961). Although the effect is similar, the mechanism involved in the present findings is probably different in that the separation of thick and thin filaments is implicated as a major factor pre rigor. The spatial separation achieved, and hence the WBC, will be some function of the charge on the filaments and the salt concentration.

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