

FUNCTIONAL PROPERTIES OF THE COPRECIPITATION OF MUNG AND WHEY PROTEINS

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ABSTRACT

Functional properties of mung bean Flour (MF), mung bean Isolate (MI), Whey protein (W), Whey-mung flour proteins coprecipitate (WMF) and Whey-mung protein isolates coprecipitate (WMI) were studied. Analyses of discrepancy showed differences in water absorption, oil absorption and gelation among the five protein products. Protein coprecipitate of WMI showed higher protein solubility, emulsification capacity and foaming capacity than those of WMF and W. Addition of mung bean flour and mung bean isolate to whey protein improved the functional properties of the coprecipitate protein.

The minimum solubility of whey proteins, WMF and WMI was quite sharp at pH 4.5. Meanwhile, mung bean flour and its isolated proteins exhibited a broad range of minimum solubility at pH range of 4.5 to 5.0. The foam capacity and emulsification capacity increased with increasing sodium chloride concentration to a maximum value (0.6M) and then decreased for all proteins. The foam capacity and emulsification capacity -pH profile of MF, W, MI, WMF and WMI pattern was similar to protein solubility-pH profile

INTRODUCTION

Legumes serve as the main source of protein and calories in many tropical and subtropical areas of the world. Dry legumes and their products are the richest of food protein from plant (Sathe *et al.*, 1984). Thus, legume flours have been used to fortify many products to improve their nutritional values (Deshpande *et al.*, 1983 and McWatters, 1990). Mung bean has about 20 - 7% protein with an essential amino acids content similar to that of soybean and kidney bean (Evans and Bandemer, 1967; Fan and Sosulski, 1974 and Thompson *et al.* 1976).

Functional properties of any protein are very important and helpful in orienting such protein to the right application (Sosulski and Youngs, 1979). The functional properties are the intrinsic physico-chemical characteristics which may affect the behavior of food system during processing and storage, e.g. solubility, foamability, gelation and emulsification properties (Oshodi and Kperigin, 1989). The use of soybean as a source of nutritious food and substitute for meat arises from knowledge of the functional properties of the flour and other products (Kinsella, 1979; Young & Scrimshaw, 1979). Mung bean protein isolate had highest emulsification capacity, oil absorption, foam capacity and stability than mung bean flour (El-Adawy, 1996). The use of mung bean as a protein supplementer is limited due to its bean flavour and dark colour (Thompson *et al.* 1976).

Whey proteins that represent 20% of milk proteins are lost in whey during the manufacture of cheese. The Egyptian public sector dairies produce about 118.8 thousand tons sweet whey, which is disposed in the sewage system (El-Sayed, 1987). Whey proteins which have been precipitated from

sweet salted whey had lower functional properties (*kebary, 1992*). The objectives of this study were preparing mung bean isolate and coprecipitates from whey and mung bean proteins and assessment of the functional properties of the resulting proteins product.

MATERIALS AND METHODES

Preparation of proteins

Protein isolate(MI) was prepared from mung bean flour (MF) as one kilogram of flour was suspended in 10 liters distilled water then adjust to pH 9.0 using 1 M NaOH. The suspension was shacked for 1 hr at room temperature then was centrifuged at 4000 rpm for 20 min. In order to obtain higher yields, the extraction and centrifugation procedures were repeated on the residue. The extracts are combined and acidified to pH 4.5 and 4.8 for mung bean proteins.

The precipitate was recovered by centrifugation at 4000 rpm for 20 min. The precipitate was washed by distilled water several times, and then neutralized by 1.0 M NaOH to pH 7. The neutralized precipitate was dried using freeze-dry (Lab Conco Freeze Dry 64312. Kansas, Missouri), was then milled using household mill (Braun, Germany) and finally sieved through 60 mesh.

Preparation of cheese whey protein (W) was prepared by filtering the bulk of Domiati cheese whey through cheese cloth, adjusting the pH to 4.6 with 1 N HCl heating at 90 °C for 20 minutes in a water bath then cooling to 25°C. Whey was filtered through muslin cloth. The precipitated whey protein was washed several times with distilled water. All products were dried by freeze-dry. Whey –mung flour (WMF) and whey –mung isolate (WMI) protein coprecipitate were prepared as described in Figs (1).

Protein content

The nitrogen content of the flour was determined by the micro-Kjeldahl methods according to AOAC 1985.

Protein solubility as a function of pH

The solubility of nitrogen index was determined according to *King et al., (1985)*. Using dried whey and mung bean proteins in distilled water (5%w/v). the pH adjusted in the range of 1 to 12 with 0.5M HCl or NaOH. The nitrogen content in the clear supernatant was determined by micro-Kjeldahl method according to AOAC 1985.

Water and fat absorption

Water and fat absorption were determined according to the methods of *Sosulski (1962) and Sosulski et al.,(1976)*. The results were expressed as water and corn oil retained by 100 g of flour or protein.

Emulsification capacity

The procedure described by *Beuchat et al. (1975)* was used to. Emulsification capacity (E.C.) is expressed as ml of oil emulsified by 1 gm of flour or protein. Emulsification capacity was determined as a function of pH (1 – 12) and sodium chloride concentration (0.2 to 1.2 M).

Mung bean flour (MF) Extract with 0.2% NaOH (Solvent: MF =10:1 V/w) Shake 1hr. at room temp. Centrifuge 4000 rpm / 20 min.

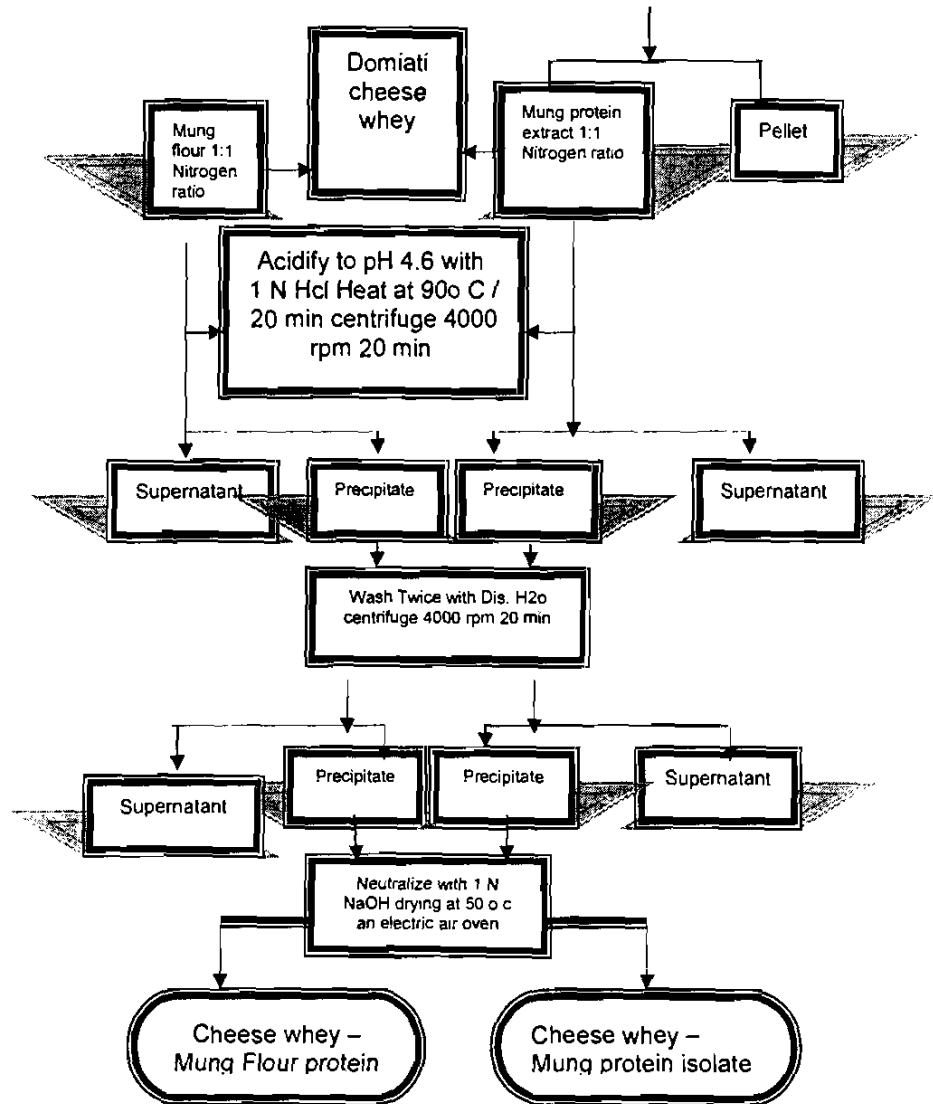


Fig. (1): Preparation of Cheese whey Mung bean Flour proteins coprecipitate (Kebary .1993)

Foaming capacity and stability

Foaming capacity and stability were determined according to the methods of *Lawhon et al.* (1972). Flour sample of 2 gm and 50 ml distilled water were mixed in Braun blender (~ 25°C). The suspension was stirred for 5 min at 1600 rpm (2 nd speed) and the contents along with the formed foam were poured into a 100 ml graduated measuring cylinder. The percentage increase in volume after 30 sec was recorded as foam capacity (FC) according to the following formula:

$$\% \text{ Volume increase or (foam capacity)} = \frac{\text{Total volume} - \text{Initial volume}}{\text{Initial volume}} \times 100$$

The foam volume was recorded after 15, 30, 45, 60, 90 and 120 minutes of standing at room temperature (~ 25°C) as foam stability (FS) according to the following equation:

$$\text{Foam volume (ml) or (foam stability)} = \text{Total volume} - \text{Liquid volume.}$$

The foam capacity and stability were also determined as a function of sodium chloride concentration (0.2 to 1.2 M), and pH (1 to 12).

Gelation

The ability of flour samples to form gel was measured according the method described by *Shigeru and Kinsella* (1985). An aqueous dispersion of different flour concentrations were prepared 6, 7, 8, 9, 10 and 11%, (w/v) in 5 ml distilled water. The test tubes containing these suspensions were heated in a boiling water bath for 30 min followed by rapid cooling to 14°C and then kept at 4°C for 24 hr to ensure complete gelation. The least gelation concentration was determined as that concentration when the sample from the inverted test tube did not fall down or slip. This determination was done in triplicates.

RESUTES AND DISCUSSION

Protein solubility

The effect of pH on protein solubility of MF, W, MI, WMF and WMI is presented in Fig. (2) All samples gave a U-shaped curve in the pH range of 1 to 12 with a solubility minimum, which is similar to many oil seed and legume proteins (*Lawhon, et. Al. 1972*). The minimum solubility of whey proteins, WMI and WMF and was quite sharp at pH 4.5 with 11.3, 14.7 and 16.1 % protein in the solution, respectively, (*Chobert et al., 1988*). Meanwhile, mung bean flour and its isolated proteins exhibited a broad range of minimum solubility at pH range of 4.5 to 5.0 with 18.5 and 20.5% protein in the solution respectively. Whey mung isolate proteins coprecipitate (WMI) had higher protein solubility at any pH than these of whey mung flour coprecipitate (WMF) and whey protein (W). Which means that preparation of coprecipitation of whey and mung improved the whey protein solubility. These results revealed that it is possible to use WMF and WMI in soft drinks and slightly acidic beverages to increase and improve its protein content and nutritional quality.

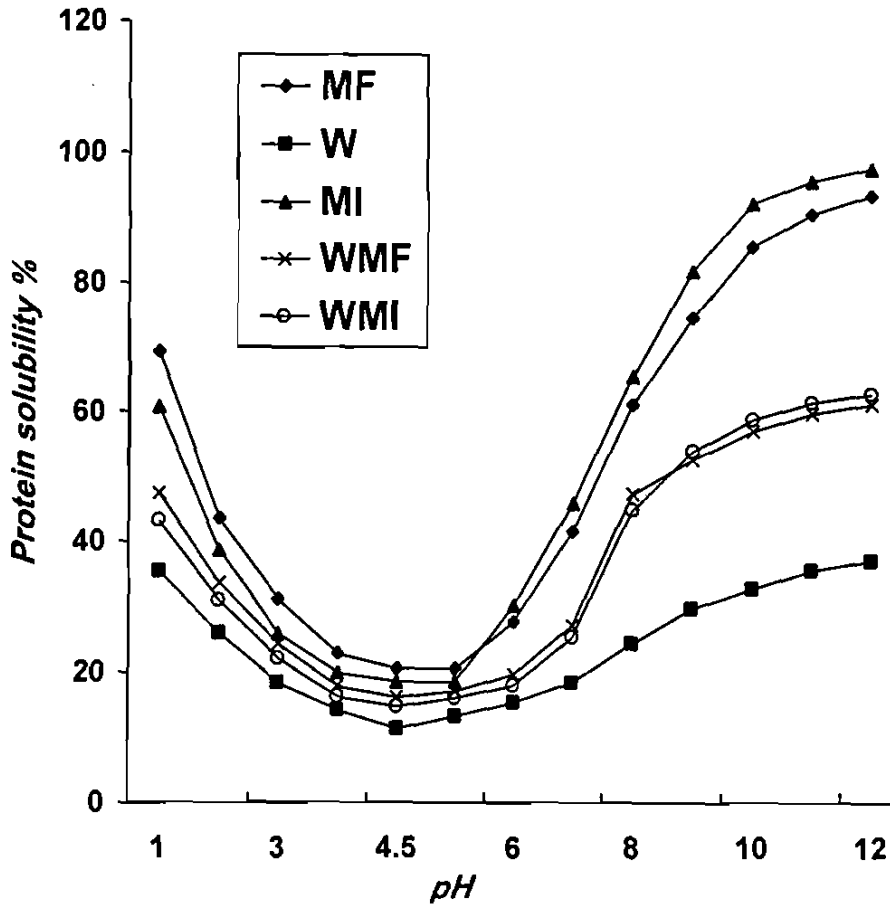


Fig (2): Effect of pH on protein solubility of (MF), (MI), (W), (WMF) and (WMI). See table 1

Water and fat absorption capacity

The water and fat absorption of MF, W, MI, WMF and WMI are shown in Table (1). Mung bean seed flour had the highest water absorption capacity. The values for MF, W, MI, WMF and WMI were 209.5, 140.6, 151.9, 189.7 and 174.8 gm H₂O/100 gm flour, respectively. These results are in agreement with those reported by EL-Adawy (1996) for mung bean seed flour, who found that water absorption capacity value was 216.4 gm/100 gm flour. The differences in the water absorption capacities may be due to the conformational features of the proteins, also some other chemical compounds rather than protein particularly starch and crude fibre may take place in water binding capacity (Kuntz, 1971). The results of water absorption capacity showed an advantage for mung bean flour and coprecipitate WMF and WMI utilization in some bakery products or as meat extenders, which require holding more water.

Table (1): Water, Fat absorption and gelatin of Mung bean and Whey proteins coprecipitate .

Property	Protein samples				
	MF	W	MI	WMF	WMI
Water absorption Capacity g/100g	209.5 ±1.91	140.6 ±1.50	151.9 ±1.34	189.7±1.83	174.83 ±1.25
Fat absorption Capacity g/100g	99.4 ±1.26	78.3 ±0.82	103 ±0.91	83.5±0.74	92.65 ±0.83
Gelation (% protein)	4.5	7	6.5	5.5	6.5

MF : Mung bean flour.

W : Whey proteins.

MI : Mung bean protein isolate.

WMF : Cheese whey -Mung bean flour proteins coprecipitate.

WMI : Cheese whey -Mung bean proteins isolate coprecipitate

The ability of proteins to bind fat is an important phenomenon since fats act as flavour retainer and increase the mouth feel of food (Kinsella, 1976). MI exhibited the highest values of fat absorption followed by MF, WMI, WMF and W. The preparation of coprecipitate of whey and mung bean proteins improved the water and oil absorption. Kinsella and Fox (1986) and Morr (1992) reported that the differences in processing equipment design and operating conditions affect the water and fat absorption of the same whey protein concentrate.

The results showed that all proteins were not able to form gel at lower concentration than 4.5%w/v.gelation capacity of whey protein was the lowest. Generally, the difference in the gelling ability of the different samples is not only a function of the quantity of protein but also to the type of protein and non-protein components present in the sample such as starch (Sathe and Salunkhe 1981, and Bencini, 1986). Preparation of coprecipitate of whey and mung bean proteins improved the gelation ability.

Emulsification capacity

Effect of ionic strength on the emulsification capacity

The effect of sodium chloride concentration on emulsification capacity of MF, W, MI, WMF and WMI is presented in Fig. (3). There was an observed increase in emulsification capacity with increasing sodium chloride concentration up to 0.6 M then decreased. In water, the emulsification capacity was 93.6, 25.5, 98.4, 53.6 and 65.7 for MF, W, MI, WMF and WMI ml oil/gm protein, respectively. Emulsification capacity is known to increase with increasing moderate salt concentration due to salting-in effect of the proteins. At higher salt concentrations the emulsification capacity does not increase as there is likely proteins undergo salting-out effect. The data discussed above confirm this pattern. Also, Rahma (1979), Nath and Narasinga Rao (1981), Narayana and Narasinga Rao (1982), Tasneem et al. (1982). WMI had higher emulsification capacity at any sodium chloride concentration than both WMF and W, while its emulsification capacity was lower than those of mung bean flour and mung bean isolate.

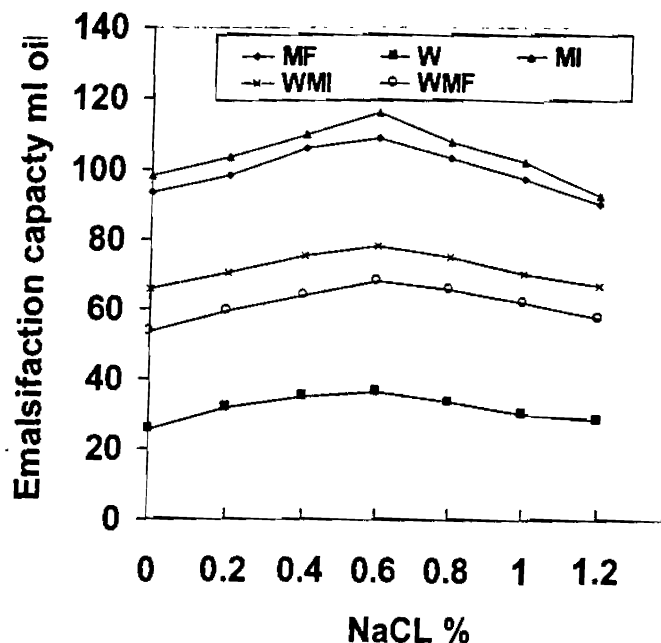


Fig (3)Effect of sodium chloride concentration on emulsification capacity of MF,W,MI,WMF and WMI. See table 1

Effect of pH on the emulsification capacity

The emulsification capacity of MF, W, MI, WMF and WMI as affected by pH (1 to 12) is presented in Fig. (4). The emulsification capacity vs pH profile of all samples showed a similar pattern to the protein solubility vs pH profile suggesting that emulsifying property is mainly due to the soluble protein. The emulsification capacity values were higher on both sides of the pH 4.5-5.0 for all proteins (the minimum solubility pH of each protein). Whey mung isolate proteins coprecipitate (WMI) had higher emulsification capacity at any pH than these of whey mung flour coprecipitate (WMF) and whey protein (W), but was lower than MF and MI. The emulsification values of all samples were higher in alkaline pHs than in acidic pH's. The emulsification capacity vs pH profile was simulate those obtained by *Crenwelge et al. (1974)* for soybean, *Rahma (1979)* for sunflower, *Narayana and Narasinga Rao (1982)* for winged bean.

Foam capacity and stability

The foam capacity was 66, 17.9, 112.8, 32.6 and 62.9 % for MF, W, MI, WMF and WMI, respectively (Fig.5). Foam stability at room temperature decreased markedly within the first 15 min and then the decrease was gradually up to 90 min and was almost stable after that. Mung bean isolate (MI) had the highest foam stability followed by MF, WMI, WMF and W. This decrease may be due to collapsing and bursting of the formed air bubbles (*Kinsella, 1976*).

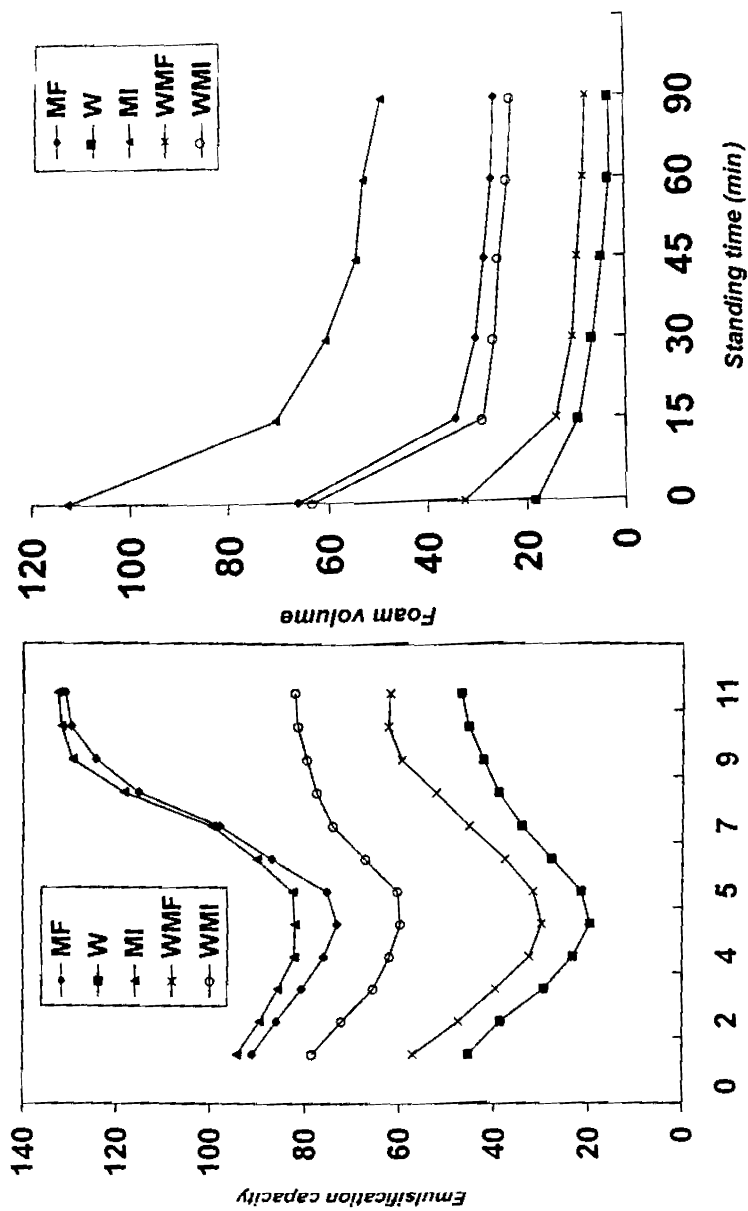


Fig (5) Foam capacity and stability of MF, W, MI, WMF and WMI. See table 1

Fig (4): Effect of pH on the emulsification capacity of MF, W, MI, WMF and WMI. See table 1

Effect of ionic strength on the foam capacity

Fig. (6) shows the effect of sodium chloride concentration on the foam capacity of MF,W,MI,WMF and WMI in the range of 0.0 to 1.2 M. NaCl. All samples, the foam capacity of all protein products increased with increasing sodium chloride concentration to a maximum value and then decreased. Generally, the maximum foam capacity was occurred at 0.6 M sodium chloride for all proteins. Mung bean isolates had the highest foaming capacity followed by MF, WMI, WMF and W. This increase and decrease of foaming capacity by increasing the sodium chloride concentration might be mainly due to the protein solubility (De Witt, 1989)

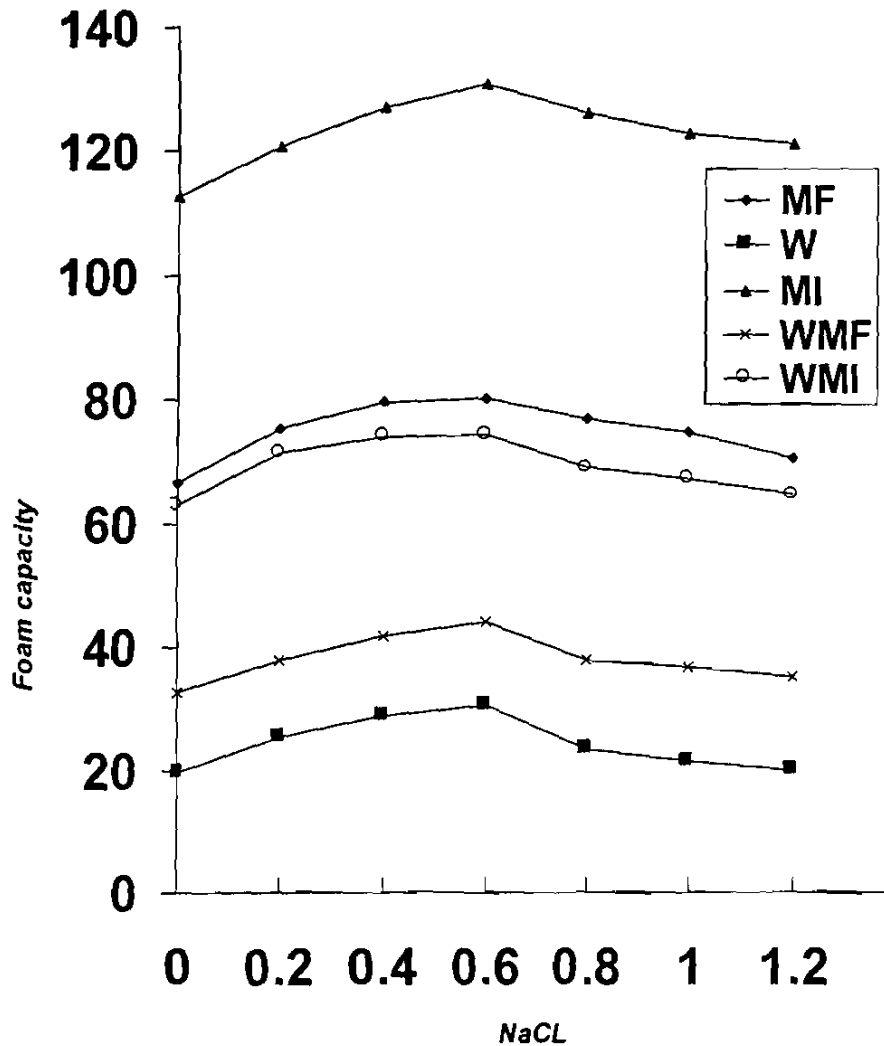


Fig (6): Effect of pH on the foam capacity of MF, W, MI, WMF and WMI.

See table 1

Effect of pH on the foam capacity

The foam capacity-pH profile of MF,W,MI,WMF and WMI is presented in Fig. (7). In general the foam capacity-pH pattern also was similar to protein solubility-pH profile, suggesting that foaming property is also mainly due to soluble proteins. The values being at low pH 4.5 for W,WMF and WMI and 4.5-5 for MF and MI. The foam capacity values at the minimum pH's were 42,10,54,17 and 33% for MF,W,MI,WMF and WMI, respectively.

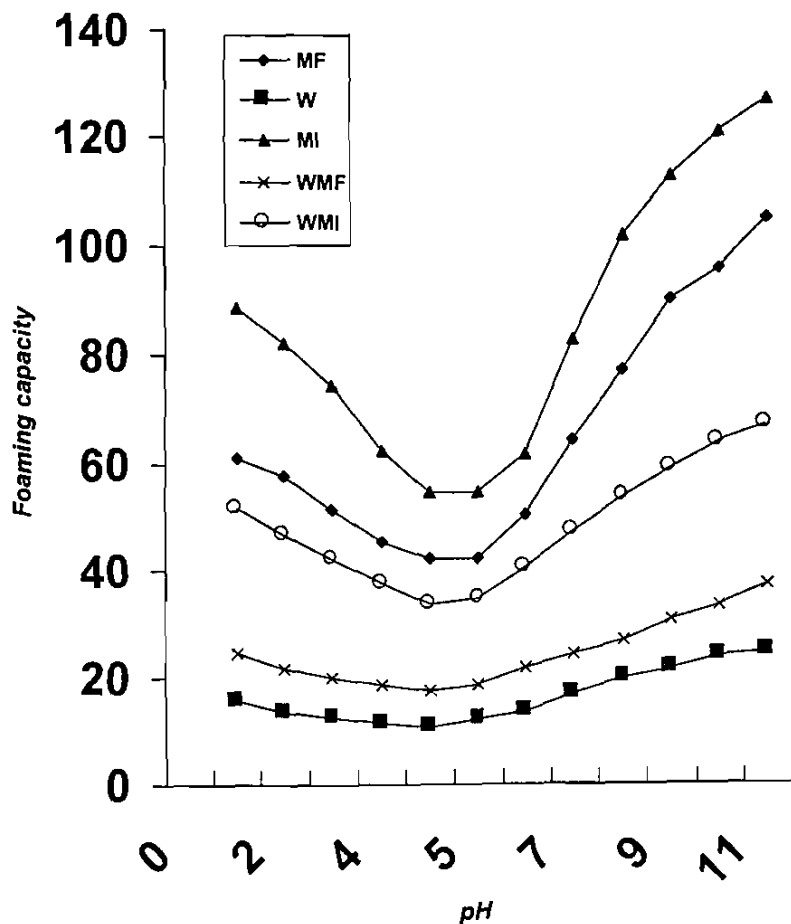


Fig (7): Effect of pH on the foam capacity of MF, W, MI, WMF and WMI. See table 1

Generally, whey protein had lower foam capacity compared to other samples at pH 2-11. Also, foam capacity of all proteins was higher in the alkaline region of pH compared to the acidic side. Similar observations have been reported for soy protein isolate, caseinate and whey protein concentrate (Hermansson, 1975). The observed minimum foam capacity at previous pH's, is due to low solubility of protein. Also, the strong intermolecular forces which prevent the unfolding and spreading of the protein molecules. This also is proved by the phenomena that protein molecules has almost no net electrical charges at the isoelectric pH, thus exists at the minimum solubility.

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الخواص الوظيفية لمعدّات بروتين فول الماتج وبروتين الشرش

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تم دراسة الخواص الوظيفية لكل من دقيق فول الماتج والبروتين المعزول من فول الماتج ، وبروتين الشرش ، ومعدّات بروتين دقيق الماتج مع الشرش أيضا معدّ البروتين المعزول من الماتج و الشرش . وقد أثبت النتائج المتحصل عليها إن هناك اختلاف في معدل امتصاص الماء ، معدل امتصاص الزيت أيضا وجد اختلاف في نسب تكون الجل بين الخمس أنواع من البروتينات المنتجة. اظهر معدّ البروتين المعزول للماتج مع بروتين الشرش أعلى معدل ذائنية البروتين ، سعة الاستحلاب و أيضا سعة الرغوة عن معدّ دقيق الماتج مع بروتين الشرش وكذلك بروتينات الشرش. لوحظ ان معدّات بروتينات الشرش مع كل من دقيق الماتج وكذا البروتين المعزول من الماتج حسنت الخواص الوظيفية لمعدّات بروتين الشرش.

كانت أقل ذائنية لمعدّات بروتين دقيق الماتج مع الشرش أيضا معدّ البروتين المعزول من الماتج و الشرش عند pH 4.5 بينما كان شكل المنحني متسعا لكل من دقيق فول الماتج والبروتين المعزول من فول الماتج وتتراوح pH 4.5 - 5.0. لوحظ ارتفاع قيم سعة الرغوة والاستحلاب مع زيادة تركيز كلوريد الصوديوم حتى تركيز 0.6 ثم تخفض وذلك لكل البروتينات. منحنيات القدرة على تكوين الرغوة وسعة الاستحلاب على درجات حموضة مختلفة تماثلت مع منحنيات ذائنية البروتين على درجات حموضة مختلفة لكل البروتينات .